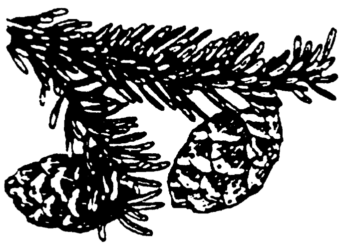
CANADIAN FOREST GENETICS ASSOCIATION
ASSOCIATION CANADIENNE DE GÉNÉTIQUE FORESTIÈRE



Tree Seed Working Group

NEWS BULLETIN

No. 54 December 2011

THIS ISSUE AT A GLANCE

CHAIR'S 'ARMCHAIR' REPORT

Hello! I hope this finds you healthy for the upcoming holiday season. I'd like to first of all thank the organizing committee of Canadian Forest Genetics Association (CFGA) 2011 for a wonderful meeting and for hosting a Tree Seed Workshop <u>and</u> field tour. It was a great opportunity to meet some maturing friends and make some new ones. Highlights at the CFGA for me were many of the invited speakers, Kakabeka Falls, and the world famous (for Lakehead graduates) Hoito Restaurant. It was great to see Ben Wang at the meeting and also representing Ontario, the four musketeers from the Ontario Tree Seed Plant.

In terms of CFGA 2013, planning is underway and our co-chairs Sally Aitken and Lee Charleson are leading the charge. The location is Whistler, BC and the dates are July 22–25 with venue confirmation coming soon. We are looking to host a Tree Seed Workshop on July 22nd and it will most probably be in Surrey or Langley to allow for a tour of our Tree Seed Centre. Thoughts regarding topics and/or speakers are always appreciated. Although early in the planning stages, we will probably have a bus to take participants from the Tree Seed Centre to Whistler for the conference proper.

It looks like seedling production is continuing to rise again in BC this year (or people are putting in requests earlier) as we are already at 190 million seedlings requested which is 30 million ahead of last year at this time. An ongoing target area for BC is the production of a climate based seed transfer system. I'm sure you'll hear more about this at CFGA 2013. Cone and seed processing is at a decade-low with most collections from mountain pine beetle ravaged areas already completed and a dip in many seed orchard crops after a very productive 2010 season. In terms of sowing, last year 55.8% of our seed sown was from seed orchards and will continue to rise over the next decade.

Wishing everyone the very best over the holiday season. Hope you have some time off to share with your loved ones.

Dave Kolotelo
TSWG Chairperson

EDITOR'S NOTES

This is an open issue which contains a variety of great articles. Firstly, I want to welcome Lindsay Robb, Alberta's provincial seed specialist. She tells us about herself and her experiences in the first article. Taxonomy is not static which is exemplified in an article about the reclassification of yellow cypress and followed by updated seed pretreatments for yellow cypress seed. The Quebec provincial government is making progress in developing seed storage containers for *ex situ* conservation. There are several newsy articles covering highlights of the Tree Seed Workshop at the CFGA conference, a perspective of the CFGA conference, and an update on seed orchard activities by JD Irving. There is an article about the impact on seed quality of hand- and mechanically-picked white spruce cones followed by an update on the Mexico Tree Seed Centre. An article about upgrading white birch seed completes this issue.

I wish everybody a Joyful Holiday Season and a healthy, prosperous New Year.

Dale Simpson
Editor

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

All issues of the News Bulletin are available at: http://www.for.gov.bc.ca/hti/treeseedcentre/tsc/tswg.htm

ALBERTA'S NEW PROVINCIAL SEED SPECIALIST!

In September, the Alberta Tree Improvement & Seed Centre welcomed Lindsay Robb as their Provincial Seed Specialist.

Lindsay was born in Kingston, Ontario and completed a BSc (Honours) at Trent University and a MSc at Laurentian University, both focused on botanical restoration and nitrogen cycling. Shortly after completing her MSc defence, Lindsay moved to the United Kingdom where she lived for 10 years. Below she writes about her previous job, living in Europe, travelling the world, and her future at ATISC.

Donna Palamarek

I worked as a Germination and Longevity Specialist for 4.5 years at the Millennium Seed Bank (MSB), part of the Royal Botanic Gardens, Kew. The seed bank was started in the early 1970's and is located within Kew's 180 acre 'country garden' of Wakehurst Place, about 60 km south of London, England. The Millennium Seed Bank Partnership was conceived after the Earth Summit in Rio in 1992 and in 2000 the current building was completed. In addition to the banking and research facilities, there are 14 bedrooms available for visiting students and colleagues. There is also an interactive exhibition that allows the public to learn how seeds are collected from the wild, what happens to them when they arrive and, most importantly, why it is important that seeds are stored for the future.

Visitors are able to look in on most work areas and see actual staff and not actors. Imagine having to work all day with someone watching you through your window! It took a little while to get used to it.

There are three main aims of the Partnership: 1) to conserve all UK seed bearing plants (nearly complete), 2) to conserve 10% of the world's seed bearing plants by 2010 (completed) and 25% by 2020, and 3) to develop research and partnerships around the world in order to support and advance plant conservation. To date, the MSB Partnership has partners and collaborators in more than 120 institutions in over 50 countries around the world, and focuses on prioritising the collection of the 3 'E'S – endemic, endangered, and economically important species. The site of the Millennium Seed Bank is the most genetically rich spot on the planet. The bomb, earthquake, and flood proof banks store nearly 60 000 collections of over 30 000 species from 128 countries. The processing teams and volunteers clean, dry, and store over 6000 collections and set up over 10 000 germination tests each year.

My role at the MSB evolved and changed quite a lot over the 4.5 years I was there. Not having a large background in seed science or botany, it was a very steep learning curve for the first two years! Initially, my general responsibility was for solving germination problems arising from the MSB germination testing program and for applied research into novel approaches to overcome seed dormancy. My research was heavily involved in developing an online search tool for the germination of wild seeds. I was part of a small team developing and testing a user interface tool, which searches the MSB database and utilizes WorldClim climate data to enable seed germination temperatures and treatments to be more accurately predicted by MSB staff. This tool has been completed internally for the world and the UK information is now live on the RBG, Kew website for public use: www.kew.org/science-research-data/databases-publications/uk-germination-tool-box/index.htm.

Much of my later time at the MSB was divided between two related projects. I was part of a team using probit analysis to analyse 40 years of MSB germination test data in an effort to use taxonomy, climate, seed morphology, and/or habitat to determine and possibly predict which new seed collections may be shorter-lived under seed bank conditions and whether alternative storage measures (e.g., cryo-storage, ultra-dry storage, low oxygen storage) should be considered in an effort to improve longevity. This work also included the development of a shortened comparative longevity procedure, which is faster and uses less seed than previous methods, while still giving good estimated results. The understanding derived from these studies is being incorporated into the Department's standard seed intake protocols in order to maximise and actively manage the quality of the MSB seed conservation collections.

The second focus surrounded the further management of the seed collections by developing and implementing new and updated standard operating procedures for initial germination tests, retesting collections, chemical viability testing, and the management and prioritisation of short-lived and important collections by creating criteria for storing seeds in our new cryopreservation facility and/or regenerating seed collections to bulk up our accessions.

While I enjoyed nearly all aspects of my position at the MSB, I have to say that my favourite role was as a student supervisor and trainer. I provided training in seed germination and dormancy, tetrazolium chloride testing, seed longevity, and seed storage behaviour, both at the MSB site and abroad, which can challenge your communication abilities to the maximum! I trained dozens of visiting students and colleagues from many countries around the world including Sweden, Argentina, Burkina Faso, China, and yes, even British Columbia! But the most enjoyable aspect of training was when I was required to go in-country. The MSB encourages this type of training, since students/colleagues will have to eventually go back to their place of work and attempt to implement what they have learned. It is much more efficient to be able to see what access they have to equipment and supplies and modify the training to suit their situation and needs. My favourite training destination was by far the Tanzania Tree Seed Agency in Morogoro, Tanzania. The MSB held a 2-week Seed Conservation Course for our new and existing African conservation seed bank partners and a new restoration seed bank by Rio Tinto. It was very challenging and we had a fabulous time with a great bunch of students. The locals are so friendly and the pace of life is very relaxed. I came back with the 'Africa Bug' and will go back again soon I'm sure. I've always wanted to see the Okavango Delta in Botswana.

I loved my job at the MSB but the economy in the UK has been hit very hard and the standard of living and the amount of disposable income is very different than in Canada. My new job is a great opportunity and a fantastic challenge to implement everything I have learned about seed science. I really want to help build one of the best seed conservation programs in Canada.

Initially, I have been coming to grips with the equipment and methods employed, as well as the forest industry in general. The next step will be to review, assess, and provide equipment and accurate protocols for storage and testing and implement a good quality monitoring program.

As ATISC starts to take in non-commercial tree (e.g., whitebark pine) and woody shrub seeds, new testing protocols will need to be set up and there will be opportunities for research. I will be providing seed storage and germination advice to Wild Rose Consulting Inc. and novaNAIT with respect to reclamation work on the oil sands in the Fort McMurray area of Alberta. I have already begun providing some advice to the Smoky Lake Forest Nursery next door on some of their problem woody shrubs. This is the part of my job that I am very passionate about and I am happy to be able to continue, and hopefully expand, teaching and training in the near future at ATISC for colleagues, students, and the public. I think it is so important that the general public understands the value of the work we do, where their tax money is going, and why conserving even non-commercial plant seeds is important for our future.

Leaving England was like leaving home for me and I left many lifelong friends behind that give me a good reason for future travel. Living in Europe and travelling abroad has been the single most influential experience on my social and political views. I strongly encourage everyone to step outside their comfort zone and look a little further than Mexico when they book a vacation next year. In my last 2 years I did a lot of travelling for work – Australia, Belgium, Republic of Georgia, and Germany – and this encouraged me to do some travelling alone. My best recommendations would be Tuscany, the Rhine Valley in Germany, and Paris. They are all just as amazing as their reputations and they are places that I will revisit.

I am finding a lot to love about Alberta and its culture – there is so much here that is new to me. I am trying to get involved and support community events and figure out which Ukrainian foods are my favourites (peteshke!). And I am desperately waiting for proper snow, so I can try out my new cross country skis and snowshoes!

If anyone would like to contact me, I would welcome an email or a phone call, even if it's just to introduce yourself.

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A NEW CONIFER SPECIES AFFECTS TAXONOMIC CLASSIFICATION IN THE Cupressaceae

The discovery of a new conifer species and its subsequent phylogenetic description has had significant and controversial impacts on classification within the Cupressaceae including yellow cypress (formerly known as *Chamaecyparis nootkatensis*). In this article I will outline the cause of this disagreement and how it is influencing the taxonomic classification of the new world cypresses including yellow cypress, for which I will refer to by various genera as it was then commonly accepted.

In the fall of 1999 a new conifer species was found in a moist forest on limestone karst ridges in northern Vietnam. This species had a morphological resemblance to others in the Cupressoideae subfamily of the Cupressaceae especially *Chamaecyparis* and *Cupressus*. However, after a thorough morphological description, the conifer was distinct enough to warrant a new genus and species, and was given the new scientific name *Xanthocyparis* (xantho=yellow, cyparis=cypress) *vietnamensis* Farjon & Hiep (Farjon et al. 2002) with the common name of Vietnamese golden cypress. The most distinct morphological feature of this species is the occurrence of juvenile, intermediate, and mature foliage in the upper crown (Farjon et al. 2002*)*. Upon closer examination, including molecular data, a number of authors placed *Chamaecyparis nootkatensis* as a sister taxa. Morphological similarities included seed cones with 4 (to 6) bract-scale complexes (Farjon et al. 2002), apically distributed ultimate branchlets, and externally dimorphic mature leaves (Farjon et al. 2002, Little et al. 2004). It was proposed initially by Farjon et al. (2002) that *Chamaecyparis nootkatensis* be renamed as *Xanthocyparis nootkatensis.* This genus name was later disputed by Little et al. (2004) in which they proposed the name *Callitropsis* for both species – more on this later.

Chamaecyparis nootkatensis has had an interesting taxonomic past being first placed in *Cupressus* in 1824 and later transferred to *Chamaecyparis* in 1842 (Little et al. 2004). To complicate matters further, Orsted created the monotypic genus *Callitropsis* in 1865 for *Chamaecyparis nootkatensis* because of the somewhat unusual ovulate cone configuration; however, this classification did not catch on (Little et al. 2004, Mill and Farjon 2006). Recently, new molecular evidence from Gadek et al. (2000) indicated that *Chamaecyparis nootkatensis* was closely related more to the genus *Cupressus* than to *Chamaecyparis*. There was also growing evidence that showed the species was unique within *Chamaecyparis* including duration of seed maturation, seed wing anatomy, wood anatomy and secondary chemistry, fertilization, and low cross-compatibility of microsatellite primers among others (citations in Little et al. 2004). *Chamaecyparis nootkatensis* also hybridizes with a number of *Cupressus* species (e.g., Leyland cypress); however, there are no documented hybrids with other *Chamaecyparis* species. We have been hybridizing *Chamaecyparis nootkatensis* with both *Chamaecyparis* and *Cupressus* species over the years at Cowichan Lake Research Station and in New Zealand and have had success only with the latter genus.

This leads us to the dilemma of naming *Chamaecyparis nootkatensis.* Compelling evidence has shown that this species is a sister taxa with *Xanthocyparis vietnamensis* (Farjon et al. 2002, Little et al. 2004, Little 2006, Mill and Farjon 2006) coupled with the above evidence that it is unique within *Chamaecyparis*. Farjon et al. (2002) correctly placed both species in a new genus since they were clearly distinct from those in *Cupressus* and *Chamaecyparis*. However it seems that taxonomic precedent favours *Callitropsis* under the rules of the International Code of Botanical Nomenclature, as the earlier-published name has priority over *Xanthocyparis* if that genus includes *Chamaecyparis nootkatensis* (Little et al. 2004).

A proposal was put forth by Farjon and others at the 2011 International Botanical Congress to use *Xanthocyparis* but it did not make it to the committee that decides on taxonomic conflicts. Essentially that leaves *Callitropsis* as the genus name we should now use. So, following Little (2006), the new scientific name for yellow cypress is *Callitropsis nootkatensis* (D. Don in Lambert) along with its sister taxa *Callitropsis vietnamensis (*Farjon&Hiep).

The Cupressaceae taxonomic controversy doesn't end here. Little (2006) states that "classifications within the Cupressoideae have been contradictory as a result of taxonomically incomplete intuitive analyses combined with an emphasis on characteristics of ovulate cones to the exclusion of vegetative, anatomical, and chemical characteristics". Little also presented exhaustive evidence supporting that the New World species of *Cupressus* are more closely related to *Callitropsis* than they are to the Old World *Cupressus* species. Little proposes to restrict *Cupressus* to the Old World species and to expand *Callitropsis* to include New World species currently classified as *Cupressus*. Species from *Juniperus* and *Chamaecyparis* would still be recognized separately. Although compelling, this reclassification is currently not universally accepted mainly because the relationship between *Callitropsis nootkatensis*, *Callitropsis vietnamensis*, and the New World species of *Cupressus* has not been resolved. Little (2006) states that this may change in the future based on research currently underway involving additional character data.

On a less significant note, the common name of *Callitropsis nootkatensis* is also being debated. The species has been known under a number of common names including yellow-cedar, Nootka cypress, Alaska-cedar, and yellow cypress. Given that it is now closely aligned with cypress species and that the description as a false cedar using a hyphen[1] is rather outdated, and yellow is an apt description for its heartwood colour, then yellow cypress seems appropriate. This common name has been used in British Columbia for quite some time along with yellow-cedar.

[1] True cedars (*Cedrus spp*) are in the Pinaceae and convention dictates that any common name referring to a false species should have a hyphen or be one word (e.g., Douglas-fir, western redcedar).

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NEW STRATIFICATION PROCEDURE FOR YELLOW CYPRESS

A new stratification procedure for yellow cypress (*Callitropsis nootkatensis*) has been introduced for testing and seed pretreatment at the Tree Seed Centre. The new pretreatment extends the soak duration from 48 to 72 hours and extends cold stratification from 56 to 77 days. New germination results are available on SPAR (seed planning and registry application on the Ministry's Tree Improvement Branch web site) with a G57 test code. The test specifics are presented below for lab testing and seed preparation (Table 1).

Table 1. An overview of pretreatment details for yellow cypress (YC)

	Testing	Seed Preparation
Soak	72-hour running water soak	72-hour running water soak
Warm Stratification	28 days at 20°C on Accelerated Aging trays in closed germination dishes Monitored 3X weekly for moisture levels with misting if any seed coat drying is observed	28 days at 20°C in plastic bags with an absorbent media covering Moisture content targeted to 44%, seed agitated and weight monitored 3X per week. Moisture misted onto seed if weight is reduced or drying is observed
Cold Stratification	77 days at 2–5°C on Accelerated Aging trays in closed germination dishes Monitored initially 3X weekly for moisture levels and then weekly after no seed coat drying is observed	77 days at 2–5°C in plastic bags with an absorbent media covering Moisture content targeted to 44%, seed agitated 2X per week and weighed every two weeks. Moisture misted onto seed if weight is reduced or drying is observed

The long soak and subsequent moisture content monitoring and maintenance at high levels are critical to successful pretreatment. That has been a consistent observation from everyone who has had success germinating the species. The long soak duration ensures adequate moisture is absorbed through the megaspore membrane and the cuticle of the megagametophyte which were found to be the most important structures restricting moisture uptake. The waxy seed coat itself was not a significant factor restricting water uptake (Tillman-Sutela and Kauppi 1998).

The extension of cold stratification is considered the major factor resulting in overcoming dormancy and increasing germination capacity. In previous trials, cold stratification was extended to 119 days, but this unfortunately resulted in large numbers of pre-germinated and unproductive seed. The extension of cold stratification to 77 days is considered beneficial in substantially increasing germination and not increasing the risk of pregermination even with an additional few weeks delay in sowing at the nursery.

Forty-one seedlots were tested with this new G57 pretreatment and an average gain in germination of 17.3% was obtained. Four of the seedlots were out of tolerance and require retesting and an additional 12 seedlots were not tested as they are quite small (all < 60 g and < 2000 potential seedlings). Seed owners can expect similar gains in germination for these small seedlots and owners are encouraged to review their seed inventories and use these small lots or consider donating them to the provincial seed bank for genetic conservation purposes. Please contact me directly if you wish to donate seed to the provincial seed bank.

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TREE SEED WORKING GROUP WORKSHOP AT THE CANADIAN FOREST GENETICS ASSOCIATION MEETING

The workshop was held August 15 at Lakehead University, Thunder Bay and consisted of four morning speakers and an afternoon field trip to the SeedTek seed upgrading facility and AbiBow Kakebeka Tree Improvement Property. The four speakers are listed below with a few tidbits from their presentations:

Michelle Fullarton New Brunswick DNR Seed Production
New Brunswick has advanced tree improvement programs for white spruce (*Picea glauca*), black spruce (*P. mariana*), and jack pine (*Pinus banksiana*). Orchards from selected parents are also available for tamarack (*Larix laricina*), but demand is currently very low for this species. There have been large changes in species preferences in the province (which is only one seed planning zone) with white spruce being the preferred species at present. Realized gain trials are currently 20 years old with 15-year measurements estimating a volume per ha gain of 25.6% for white spruce and 19.9% for jack pine. In terms of GA stem injections, four staff were able to inject 6000 white spruce trees over four days. The facility, which also has a nursery and seed centre, has also recently introduced a new temperature control system for their cone . Seed is stored at -8°C for the Maritime provinces.

Dave Kolotelo Challenges Processing Cones and Seeds from Mountain Pine Beetle Killed Stands
The presentation was aimed at informing others in Canada regarding seed collection and processing opportunities with mountain pine beetle killed stands of lodgepole pine (*Pinus contorta* var. *latifolia*) which produces serotinous cones. The benefits of a canopy seed bank was emphasized along with problems associated with the loss of cone serotiny. Serotiny loss results in seed loss and opening also allows the entry of fungal spores and moisture that could lead to wetting-drying cycles that could increase seed deterioration. Other problems have been increased debris levels and, in some cases, extremely dry cones that were more difficult to open. In order to maximize seed yields some seedlots needed to be rekilned and others divided into multiple seedlots at final cleaning.

Michael Stoehr Inter-Situ Conservation
Inter-situ conservation lies somewhere between *in situ* (protected areas that conserve the Biodiversity of all organisms in their 'natural' habitat) and *ex situ* (i.e., clone banks and seed banks) that conserve genotypes removed from their natural habitat. Some existing tests that meet certain criteria are considered to be part of this dynamic genetic conservation as the trees are subject to evolutionary forces in an environment appropriate to the material. Future challenges exist regarding management and potential "regeneration' of these conservation plantings.

Dale Simpson Genetic Conservation of *Fraxinus* spp.
Dale discussed the seed collection program to conserve *Fraxinus* species genotypes as a result of the current emerald ash borer "epidemic" that is currently occurring in central and eastern Canada and USA. This effort has been underway for several years and over 500 single tree collections are currently stored at -20°C at the National Tree Seed Centre. Some of these samples have also been sent to the National Centre for Genetic Resources Preservation, Fort Collins, CO and the Millenium Seed Bank in England. For a population collection, a minimum of 15 trees spaced 50–100 m apart are required. Of specific concern is blue ash and pumpkin ash that are rare in Canada, but this pest poses a threat to all ash species.

Field Tour
It was great that the CFGA was able to host a tour for this group as this has not been a normal occurrence. We visited SeedTek (http://www.seedtek.ca/) which is a seed upgrading lab specializing in PREVAC and IDS technologies. PREVAC is a method of removing mechanically damaged seeds by placing seeds into

a pressurized water chamber to remove air from damaged seeds. Once the pressure is released the damaged seeds take up water rapidly, sink, and can be separated from undamaged seed. The BC Tree Seed Centre (TSC) has been using PREVAC and it has been very useful with some collections from mountain pine beetle killed stands. The IDS technique has also been used at the TSC, but not currently. The IDS technique is a three part Incubate-Dry-Separate process that was originally popularized in Scandinavia. In BC various trials of the technique have been performed by Dr. George Edwards, Rob Scagel, Mishtu Banerjee, and the TSC. At SeedTek the seed is imbibed and then incubated in membrane tubes. The seed is then dried and monitored so that drying will result in a separation between viable and non-viable seeds. At this point drying is stopped and seed are fed into the 'sedimentation' flume based on the Swedish design. Seed will then flow and sink at different rates with the heaviest (viable seed retaining most water) sinking first through the flume with progressively lower quality seed following.

We also stopped at the Kakabeka Tree Improvement facility to view jack pine clonal archives, progeny tests, and a seedling seed orchard. Of specific interest was the solar powered electric fences used to protect the seed orchard from deer. The configuration is actually two fences as research has shown deer can leap incredible heights with a single fence, but due to depth perception issues are resistant to jump over lower fences that are approximately 1 m apart (Figs 1 and 2).

Figure 1. "Deer fencing" at a seed orchard.

Figure 2. "Deer fencing" at a seed orchard.

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UPDATE ON THE TREE SEED CONSERVATION PROJECT IN QUEBEC

A new research project has started in 2010 at the Direction de la Recherche Forestière (DRF Research Branch) in Quebec with two main objectives: 1) to develop a strategy for the conservation of forest genetic resources, based on gap analysis and 2) to identify optimal conditions for *ex situ* conservation of forest genetic resources in seed banks (see Colas et al. 2010 for more details).

The second objective was related to a problem we identified with some of our seeds (different species) stored for more than 15 years in a freezer. This problem was detected by water activity (WA) measures on many seedlots. In fact, we first established the optimal WA value for the selected species, which are black spruce (*Picea mariana*), white spruce (*P. glauca*), and jack pine (*Pinus* *banksiana*), and observed an increase of WA (sometimes twice the optimal value) for the seedlots stored for a long period of time in a freezer. Among possible causes, the most probable one is the entry of moisture into the containers due to their permeability and/or the poor seal of the lid. It is now well known that seed moisture content equilibrates with the relative humidity of the environment (Freire and Mumford 1986); in our case, the relative humidity of the freezer at -18 °C is around 70 %.

We are collaborating with the Center of Mineral Technology and Plastics Processing (CTMP), Thetford Mines, Quebec to develop a new formulation based on a polymer and clay which would limit the exchange of moisture between the seed and the atmosphere in the freezer during storage. The work is divided into four consecutive steps: 1) development of the new polymer formulation, 2) characterization of the formulation, 3) design of the new container and lid, and 4) production of the container (Fig 1). The objective is to complete the work by March 31, 2013.

Development of the Formulation

The HDPE (high density polyethylene) polymer was chosen and different clays were added to reduce water vapor permeability. In order to increase the barrier properties of the polymer, clays have to exfoliate and be well distributed in the polymer. This is a real challenge!

Figure 1. Equipment at CTMP used to extrude the polymer together with the clays. The compound is tested (permeability, mechanical properties, etc). Photo DRF.

Characterization of the formulation

Thanks to major investments by CTMP, new equipment allows specific measurements, especially for permeability. Measurements are not completed yet, but we plan to have the optimal formulation characterized by January 2012.

Next steps

While we have indications of the “ideal” number or volume of seeds to put in conservation for present and future needs (Tree Seed Working Group News Bulletin 52 was specifically dedicated to this matter), discussions between the research branch (DRF) and the seed and seedling production branch (DGPSP) have to be made regarding both the locations of the conservation banks and the volume of seeds to be kept at each location. In the meantime, we decided to develop a container with two different volumes. Thus we can adapt the container capacity to the seed volume so that the seedlot occupies the volume of the container. The subsequent step will then be to test the containers with seeds by exposing the containers to various levels of relative humidity in order to observe if there is any change in the WA of the seeds.

During these tests, CTMP will develop a new lid. Besides being hermetic, we want to have an indicator in the lid which will change if there is a change in relative humidity in the container. For the people in charge of the seed banks, it will then be very easy to check the reliability of the storage without opening the container. If the indicator changes, then the seedlot will be rapidly stabilized to its optimal water activity and then put back in the bank. With this tool, we will eliminate the repetitive openings of the containers which are very harmful for reliable conservation (Walters 2007).

If anyone is interested to discuss this topic with us or have ideas to improve our project, do not hesitate to contact us.

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ADVENTURES IN THE NEAR NORTH

I was fortunate enough to be able to go to the Canadian Forest Genetics Association conference this year, held in mid-August and hosted at Lakehead University in Thunder Bay. This was my first trip to Thunder Bay and my first time to attend this venue. I have been with the Ontario Ministry of Natural Resources (OMNR) for over 22 years and it was the first place the OMNR has ever flown me and some of my co-workers, not that we haven't been offered before. As I write about this in December it seems like yesterday we were there.

I started out early on a Sunday morning picking up my co-workers (who are also friends), Derk Sluiter, Sarah Drabble, and Charity Hendry. We then set out for our flight to Thunder Bay. We have all flown before, but never together for work, so it was something new and exciting. When we arrived, the first thing we noticed was how different it was for all of us to have the extra hour of light, as we were used to Simcoe County, Ontario. We put the extra daylight to good use by going to Montana's to taste the local food and beverages. First thing on Monday morning we were on the bus from our hotel to Lakehead University for what I thought might be boring subject matter and a long week of meetings. How wrong was I? It was so nice to be with people that spoke the same language; seed extraction, germination, seeds per gram, and cone receiving, to name a few.

When I'm out at a function and people ask where I work, I reply that I work for the OMNR, and they assume for Fish and Wildlife. When I tell them that I work with tree seed, they look confused and ask why, what do I do with tree seed? I try to explain, I say we extract tree seed for the province, and they look at me and just ask why. They have no clue that the Province of Ontario saw the need to collect and store tree seed at the Ontario Tree Seed Plant (OTSP) for future generations starting in 1923. It's amazing to think that we have seedlots that were stored in 1954, and that are still germinating at over 43%.

The amount of seed collected can change from year to year, depending on what Mother Nature gives us. It could be a year of insect infestation, late frost, or never ending rain, which contribute to the genetic make up of that year's seed (and in some cases the next year's as well). With the seed bank in place, we have the genetics of some conifer seeds of almost every year for over 65 years, as MasterCard says: priceless! We have over 4.5 billion viable seeds in our seed vaults; we process over 2.5 million viable seeds per year from over 390 seedlots.

In 1994 we began use of our very own computer program: Tree Seed Data base (TSD). Since that time we have processed over 6640 seedlots, done more than 14 000 germination tests and received seed from over 136 species (hardwood, conifer, shrubs, and herbaceous plants) not including our province-wide single-tree, tree improvement small lots numbering over 25 000, which are attended to by our OTSP trained technical staff.

This year we've had a near bumper crop of eastern white cedar (*Thuja occidentalis*), Ontario wide. We sent our seed collection targets to our certified seed collectors across the province. All our collectors have completed our "Certified Seed Collector Workshop", which was developed and is hosted by the OTSP and Forest Gene Conservation Association. We have received over 350 hl of cedar cones this year from across the province. We are just in the middle of extraction and are finding that the seed is fairly light, but the overall yield is good. We wonder why. This summer in Ontario we had a dry spell for the month of July, and that four week period had an adverse effect on the development of the seed. It made it very light, not only for cedar but also for American high-bush cranberry (*Viburnum trilobum*), tamarak (*Larix laricina*), and red-osier dogwood (*Cornus sericea*) to name a few. We have seen this before over the years and what we have found is that the seed still germinates well, but as it ages in the seed vaults (held at -23°C), it can lose its vigour quickly and germination can decline in as little as three years, especially for eastern white cedar. This just goes to show how much Mother Nature dictates how our field of work is affected and how every year the seed has a different make up.

With the commitment of the OMNR we have the genetic diversity from years of collections stored in our seed vaults. With all that said our trip to Thunder Bay was amazing. The Canadian Forest Genetics Association put on a great week. All the speakers and all the information was so much to take in. Their outings were so well planned; Kakabeka Falls, Terry Fox Lookout with a beautiful view of the Sleeping Giant, and our tour of Fort William ending with an amazing dinner. It was great to meet so many new associates from across Canada. I am looking forward to the next function that we all may meet at together!

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J.D. IRVING, LIMITED PARKINDALE SEED ORCHARD UPDATE

In 2011, a new second-generation red spruce (*Picea rubens*) orchard block was established. These grafts were made from selections by the Nova Scotia Tree Improvement Working Group. Eight hundred ninety-six grafts were transplanted to a previously established first-generation white spruce (*P. glauca*) block that was becoming obsolete.

Our cone harvest was very small this year: 9000 l of first-generation white pine (*Pinus strobus*) and 420 l of second-generation black spruce (*Picea mariana*). Seed extracting and seed cleaning is completed.

The first-generation Norway spruce (*Picea abies*) block was rogued in November 2011. This block was established from 1986 to 1991. Lift equipment with 12 m booms was used in 2009 to collect 38 000 l of cones. Instead of using the conventional power saw for roguing, a John Deere single-grip harvester was used (Fig 1). Six clones (430 ramets) were removed based on polycross test results. Another 204 ramets were cut to create more space for the remaining ramets from elite clones (Fig 2). The limbs will be chipped with a portable chipper and blown under the trees. Some of the trees were too large for the harvester to cut, over 60 cm in diameter.

Figure 1. John Deere single-grip harvester roguing a Norway spruce seed orchard.

Figure 2. Norway spruce seed orchard after roguing.

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TEN YEARS STORAGE OF WHITE SPRUCE SEED COLLECTED BY HAND AND MECHANICALLY

White spruce (*Picea glauca*) cones are normally collected by hand which is labour intensive. Many years ago, the Ontario Ministry of Natural Resources designed a machine for collecting jack pine (*Pinus banksiana*) and black spruce (*Picea mariana*) cones, although it has not been widely used. A project was conducted in Alberta in 2000 by a company that developed a machine to remove cones from branches. White spruce cones were harvested from two stands and the cones removed by hand and by machine. Germination tests were conducted at the provincial nursery and the seed was stored in plastic bags at -18°C for 10 years at the Alberta Tree Improvement and Seed Centre, Smoky Lake, AB. We conducted germination tests in order to evaluate the effect of cone collection method on seed germinability and dormancy.

Methods

In 2011, seed moisture content was determined by the oven-drying method and expressed as percentage of fresh weight. Moisture content was determined before moist chilling and for seed that had been moist chilled for 21 and 42 days. Seed germination was carried out by sowing 4 replicates of 50 seed each on top of moist VersaPak™ germination medium in Petawawa Germination Boxes (Wang and Ackerman 1983). Germination tests were performed on unchilled seed and seed that were moist chilled at 3–5°C for 21 or 42 days. The purpose of the extended chilling time of 42 days was to evaluate if it promoted more rapid germination and increased total germination by alleviating dormancy in more seed than the 21-day chilling treatment. Boxes containing unchilled, 21-, and 42-day chilled seed were placed in a germinator on the same day. Germination conditions consisted of 8 hours light at 30°C and 16 hour darkness at 20°C with a constant relative humidity of 85%. Germination was evaluated every second day beginning at day nine and continued to day 21. A seed was considered germinated when the cotyledons were visible and the radicle and hypocotyl were well developed. The degree of dormancy is expressed as the difference in germination between moist chilled seed and unchilled seed.

Results and Discussion

Seed moisture content after 10 years of storage had not changed (6.3–6.7%) except for the seed of seedlot ANC 49 from mechanically collected cones which lost 0.7%, declining from 7.4% to 6.7%. Moisture content of chilled seed was relatively uniform ranging from 36.7–38.3% for 21-day and 37.4–38.7% for 42-day moist chilled seed regardless of cone collection method.

Germination of chilled seed was significantly higher than for unchilled seed for both collection methods (Table 1). The longer chilling time had little positive impact on total germination except for the seed from mechanically-picked cones. Germination of seed from mechanically-picked cones was consistently lower than that from hand-picked cones.

Table 2 provides a summary of the germination test results showing the change in germinability and seed dormancy status after 10 years of storage. Germination of 21-day chilled seed declined 3 and 10% for hand-picked cones and 5 and 8% for mechanically-picked cones. After storage, seed dormancy increased 1–9% for hand-picked cones and declined 7% or increased 1% for mechanically-picked cones. Assuming all things being equal, seed dormancy of hand-picked cones should be similar to that of mechanically-picked cones. In fact, dormancy of seed from hand-picked cones (15–17% before storage and 18-24% after storage) is less than that of seed from mechanically-picked cones (24–43% before storage and 25–36% after storage) possibly indicating that the seed from mechanically-picked cones was damaged. The substantial improvement in germination of moist chilled seed from mechanically-picked cones could be attributed to a natural repair mechanism induced by moist chilling (Wang and Berjak 2000).

Mean germination time demonstrates that the beneficial effect of moist chilling was more pronounced in hand-picked than mechanically-picked cones indicating that seed from mechanically picked cones was damaged or stressed (Table 3).

Table 1. Analysis of variance for germination of white spruce seed collected by two methods and treated with three durations of moist chilling

Chilling period	Collection method: Hand	Collection method: Mechanical
0 days	62.3 a[1]	33.0 a
21 days	83.0 b	63.3 b
42 days	84.0 b	67.8 b

[1] means with different letters are significantly different at $P < 0.0001$

Table 2. Germination of white spruce seed, from cones collected by two methods, before and after 10 years in storage

Lot no.	Picking method	Treatment	Germination (%) 2001	Germination (%) 2011	Loss of germinability (%)
ANC 49	Hand	Unchilled	73	62	11
		21 d chill	90	80	10
		42 d chill	-	81	
		Dormancy (%)	17	18	
	Mechanical	Unchilled	32	31	1
		21 d chill	75	67	8
		42 d chill	-	68	
		Dormancy (%)	43	36	
ANC 50	Hand	Unchilled	75	63	12
		21 d chill	90	87	3
		42 d chill	-	86	
		Dormancy (%)	15	24	
	Mechanical	Unchilled	51	35	16
		21 d chill	75	60	5
		42 d chill	-	68	
		Dormancy (%)	24	25	

Table 3. Mean germination time (days) for white spruce seed collected by two methods with three durations of moist chilling and stored for 10 years

Seedlot	Hand-picked cones 0 d chill	Hand-picked cones 21 d chill	Hand-picked cones 42 d chill	Mechanically-picked cones 0 d chill	Mechanically-picked cones 21 d chill	Mechanically-picked cones 42 d chill
ANC 49	13.8	11.8	11.5	15.6	12.9	12.0
ANC 50	13.8	11.1	11.3	15.6	12.4	12.2

Conclusions

Our results show that dormancy in white spruce seed remained high for mechanically-picked cones after 10 years in storage. The reason(s) for the loss of germinability after 10 years in storage is/are unknown because seed moisture content has not changed and the seed were stored at a suitable temperature. There is an indication that moist chilling could induce a natural repair mechanism in seed. Seed quality of hand-picked cones is higher, thus maintaining seed germinability, (and perhaps vigour and genetic biodiversity) than that of seed from mechanically-picked cones.

Acknowledgement

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MEXICO TREE SEED CENTRE – ONE YEAR OLD

This facility is essential for successful reforestation in Michoacán State, and here is why. Nurseries produce over 150 million tree seedlings per year, however establishment success hovers around 35%. Various authors have identified the reasons for this poor success. The most definitive reasons are poorly conditioned seedlings and planting late in the wet season. This is driven by seed availability.

Pinus montezumae and *P. michoacana* cones ripen in October/November while *P. pseudostrobus* cones ripen in December/January. However, the wet season starts in early June, so seedlings have to undergo conditioning (hardening) in the month of May. The nurseries are outdoor compounds so irrigation cannot be controlled once the rains begin. Conditioning has to be scheduled before the rains.

The growing degree days required to produce suitable seedlings in nurseries was determined and the nutrition program used in nurseries was evaluated. When the growing degree day accumulation at the nursery sites was calculated, we realized that sowing has to be done in September/October in order to raise seedlings to meet specification by May 1, ready for hardening. Since this cannot be done with current-year seed it necessitated constructing a refrigerated seed storage facility (Fig 1).

Figure 1. Inside of seed storage unit at Mexico Tree Seed Centre.

Seed was purchased from Adolfo Soto for immediate sowing in December 2010/January 2011. Adolfo also agreed to supply "seed with wings" – for processing at the Seed Centre. Adolfo's de-winging process consists of immersing seed in water, stirring, and discarding all that floats. A mechanized de-winging machine was built and two Clipper screening machines were acquired with the kind assistance of IFA Nurseries (Canby, OR).

After screening, each seedlot is run through an air separator to remove the last of the debris producing two sublots – large and small seed. Seed for storage is then dried (< 10%) on a drying table. Storage is at 5°C and seedlots are sampled annually for viability testing.

This season cones were acquired to develop local expertise in kiln processing of *P. pseudostrobus* in a few days rather than the three weeks required for air drying. A small kiln has been built and the first trials have begun.

One immediate problem, as described last year by Adolfo, is that cone pickers tend to harvest too early if the crop is light, for fear of losing sales. We are looking for ways to avoid the harvest of immature cones and, perhaps next year, will be testing cone maturity with SAE 20 oil flotation. Any other suggestions are welcome.

An additional challenge is that *P. michoacana* tends to have resin particles mixed with the seed that have passed through screening and air separation. Warm water (40°C) separation results in too many seed sinking along with the resin. However, raising the density of the solution by adding sugar does minimize the seed-sinking fraction. Has anyone used this technique?

For the 2011/2012 season *P. montezumae* and *P. michoacana* seed have been sown with a good chance of meeting their required degree days by May 1, but only a limited quantity of *P. pseudostrobus* seed was available from last season.

The value of a forest seed processing and storage facility is now recognized and a new brand of technician is emerging in Mexico.

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IMPROVING THE QUALITY OF WHITE BIRCH (*Betula papyrifera*) SEED. I

White birch (*Betula papyrifera*) seed are winged and light making it very difficult to remove dead, damaged, and empty seed by air aspiration. The result is storing large quantities of seed, often with low germination. In 2000, average germination of 12 seedlots with winged seed was 35.8%. After de-winging, germination increased to 74.1% (Daigle and Simpson, 2001). After 10 years in storage at -20 C, germination of the de-winged seed averaged 67.7%. In 2001, another trial was set up to evaluate the improvement in germination as a result of de-winging the seed. Seventeen single-tree white birch seedlots were upgraded by de-winging. A sample of seed was left untreated in order to compare germination with treated seed. Germination at the time of treatment and after 10 years in storage at -20 C is reported below.

Methods

Seed were placed in a cotton bag and gently rubbed to break off the wings. The contents were then transferred onto a fine mesh sieve to separate the crushed wing debris from the seed. This process was repeated several times until seed were completely de-winged. The de-winged seed were carefully blown in an aspirator to remove light debris as well as some empty seed.

Germination tests were set up by sowing 4 replicates of 50 seed each on moistened Kimpak in Petawawa Germination Boxes placed in a germination cabinet for 21 days. Germination conditions were diurnal cycles of 20 C and darkness for 16 hours followed by 30 C and light for 8 hours. Humidity was maintained at 85%. Germination was assessed at 14 and 21 days. Germinants, classified as high vigor (cotyledons green and separated with a well developed radicle and hypocotyl), were removed at each assessment time. After 10 years of storage, germination tests were conducted following the same procedures as above.

Results and Discussion

De-winging had a positive impact on seed quality. On average, germination of de-winged seed improved by 20% over that for winged seed, at the time of treatment (Table 1). The degree of improvement varied by seedlot with the greatest improvement of 54% for seedlot 113 and the least improvement of 4% for seedlot 102. There is one instance (seedlot 092) where germination of treated seed was less than that for untreated seed.

After 10 years in storage at -20 C, de-winged seed maintained its 20% margin of improvement in germination (Table 1). The averages for winged and de-winged seed were slightly higher than at the time of treatment. Seedlot 113 still exhibited the greatest differential in germination between winged and de-winged seed. Germination of de-winged seed for seedlots 036, 042, 059 and 135 had declined after 10 years. There was little difference in the proportion of low vigor germinants after storage; 0.32% for winged vs. 0.44% for de-winged seed.

Conclusions

It was possible to improve the quality of white birch seed by de-winging and this treatment had no detrimental impact on germination after ten years in storage. Four seedlots did exhibit a decline in germination and it will remain to be seen if this trend continues to the next testing time, probably in 10 years.

Table 1. Germination of seventeen winged and de-winged white birch seed at the time of treatment and after ten years of storage at -20 C

	Germination (%) (time = 0)		Germination (%) (time = 10 years)	
Seedlot	Winged	De-winged	Winged	De-winged
035	28.0	80.0	30.5	80.5
036	61.0	97.0	55.5	89.5
039	50.0	61.0	55.0	62.5
041	29.0	44.0	31.0	62.5
042	65.0	78.0	72.5	53.0
043	4.0	21.0	18.5	34.5
054	36.0	61.0	45.0	61.5
059	32.5	44.5	41.0	35.5
091	42.0	72.0	39.5	81.5
092	53.0	49.0	53.0	72.0
102	49.0	53.0	62.0	56.0
110	12.0	42.0	25.5	43.5
113	31.0	85.0	31.5	88.0
134	40.0	53.0	40.0	72.5
135	39.0	49.0	42.0	33.0
140	44.0	54.0	44.0	59.0
143	19.0	48.0	27.0	69.0
Mean	37.3	58.3	42.0	62.0

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UPCOMING MEETINGS

AOSA Annual Meeting
May 20–24, 2012 Des Moines, IA, USA
www.aosaseed.com

IUFRO Seed Orchards and Breeding Theory Conference
May 21–25, 2012 Egirdir, Isparta, Turkey
http://atabeymyo.sdu.edu.tr/seedconference
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ISTA Annual Meeting
June 11–14, 2012 Venlo, Netherlands
www.seedtest.org/AM12

Forest Nursery Association of BC
Sep 24–27, 2012 Campbell River, BC
www.fnabc.com/
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Season's Greetings