



# **Forest Genetics Council of BC**

## **Tree Improvement Program**

### **Project Report**

### **2013-2014**

Front Cover: Saanich Seed Orchard, Orchard 175, western white pine with fog rolling in.  
(Chris Halldorson image)

Back cover: From standing Douglas-fir tree to 2x4s. Growth variables were assessed in the field and wood properties in the laboratory on sawn lumber. (Don Pigott images)

**2013/2014**

Forest Genetics Council of BC

**Tree Improvement Program  
Project Report**





## Introduction and Acknowledgements

During the period of this report the Forest Genetics Council of BC (FGC) continued to lead activities related to forest genetic resource management (GRM) in BC. Business planning was completed for the allocation of Land Base Investment Strategy (LBIS) funding from the Ministry of Forests, Lands and Natural Resource Operations (FLNR). This planning also guides investments from licensees and other collaborators to ensure delivery efficiency for all facets of the provincial program. Budgets and plans were published in an annual Business Plan and the previous year's results were summarized and reported in the FGC Annual Report (<http://www.fgcouncil.bc.ca/doc.html#FGCReport>).

Broad performance indicators linked to the use and genetic gain of select seed are largely being met, with the use of select seed jumping to 70% provincially for the 2014 sowing year. Although Council missed meeting its 2014 stretch objective of 75% in 2014, it is a remarkable achievement to reach 70% and to still be increasing seed production a decade after originally setting the 2014 75% objective (see Council Strategic Plans at <http://www.fgcouncil.bc.ca/StratPlan0914-Layout-Web-22Dec09.pdf>).

The primary impediment to meeting seed production objectives continue to be the difficulties with obtaining adequate amounts of seed from lodgepole pine orchards. However, a collective effort by orchard managers and researchers is narrowing the scope of the problem, and a solution is likely to be found soon. An additional challenge will be adjustments to seed orchard capacities to meet changing seed demands under a climate-based seed transfer system. These changes will require cooperation among orchard owners with guidance from the FGC to ensure long-term seed needs are met.

Further research developments in understanding the distribution of natural genetic diversity and the alignment of this diversity with climatic patterns will allow the implementation of climate-based seed transfer policy within the next several years. This policy will facilitate more assisted migration of tree genetic populations as a means of mitigating climate-change impacts. While implemented for the purpose of maintaining forest health and productivity to meet provincially-based objectives, this policy will be a leading-edge change that is largely unprecedented world-wide and is garnering a great deal of attention from other jurisdictions and from the popular media. Less recognized are the many years of investment in field-based long-term genecology research, in climate modeling to understand what climatic factors shape BC ecosystems, and in the expertise that allows pragmatic decision-making. BC is rich in all of these and we will continue to be watched as climate-based seed transfer policy is implemented. I'm confident that we will meet this challenge, as the people involved are highly capable, willing to work together, and amenable to change.

The contents of this report outline a broad array of projects, from the creative breeding of western redcedar to improve deer browse resistance, to the control of insects in seed orchards, and to the measurement of trees in progeny tests. These projects are implemented throughout BC's forests and provide a broad web of support for applied forest management. I'm proud to be part of this program and I would like to sincerely thank members of Council and members of all technical committees for their ongoing efforts, and to acknowledge and thank all of the people within the FLNR and industry who willingly collaborate and work to make this program a success.

Jack Woods  
Program Manager, Forest Genetics Council of BC  
CEO, SelectSeed Ltd

The Forest Genetics Council Co-chairs invite you to review the programs and projects described in this report and return any questions or comments to:

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Further Tree Improvement information can be found at our web sites:

Forest Genetics Council  
FLNR, Tree Improvement Branch

<http://www.fgcouncil.bc.ca>  
[www.gov.bc.ca/treeimprovement](http://www.gov.bc.ca/treeimprovement)  
<http://www.for.gov.bc.ca/hre/forgen/>



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## Tree Improvement in British Columbia

The Forest Genetics Council of British Columbia (FGC) is a multi-stakeholder group reporting to the provincial Chief Forester and representing the forest industry, Ministry of Forests, Lands and Natural Resource Operations (FLNR), the Canadian Forest Service, and universities. Council's mandate is to champion forest genetic resource management (GRM) in British Columbia, to oversee strategic and business planning for the provincial Land Based Investment Strategy (LBIS) Tree Improvement Program, and to advise the province's Chief Forester on forest genetic resource management policies. FGC Technical Advisory Committees (TACs) provide technical and policy information to Council and contribute to the development of FGC plans and associated budgets.

Council's vision is that BC's forest genetic resources are diverse, resilient, and managed to provide multiple values for the benefit of present and future generations. This vision is supported by six objectives that are set out in Council's Strategic Plan for the period 2009 to 2014\* and reported upon annually.

Annual business planning processes are designed to support achievement of the objectives, and the FGC Business Plan defines the annual set of activities and budgets needed to achieve objectives and realize the overall vision.

Forest genetic resource management is a co-operative effort in BC. In broad terms, FLNR leads tree breeding activities and private companies contribute with progeny test locations. Orchard seed production is a collaborative effort between FLNR and the private sector. The FLNR, universities and consultants carry out research supporting operational GRM programs.

Various technical advisory committees reporting to the FGC facilitate collaboration on a variety of support issues, including genetic conservation, climate-based seed transfer, orchard pest management, extension, and records management and decision support.

The Interior and Coastal Technical Advisory Committees (ITAC and CTAC) are the primary committees reporting to and informing the FGC. Members are drawn from people involved with GRM activities in BC including operational forestry staff from forest companies or government agencies. The Chairs of the ITAC and CTAC sit as members of Council to facilitate communication and input between the policy and management perspective of Council and the more applied perspective of the TACs. Other technical advisory committees also advise the FGC, but are not directly linked through Council membership. These TACs include Genetic Conservation (GCTAC), Seed Transfer (STTAC), Extension (ETAC), cone and seed Pest Management (PMTAC), and Decision Support (DSAC).

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\* <http://www.fgcouncil.bc.ca/StratPlan0914-Layout-Web-22Dec09.pdf>

## LBIS – FGC Tree Improvement Subprograms

The Land Based Investment Strategy, Tree Improvement Program (LBIS-TIP) is structured to deliver the provincial strategy for forest genetic resource management developed by the Forest Genetics Council.

There are eight subprograms:

- Genetic Conservation
- Tree Breeding
- Operational Tree Improvement Program (OTIP)
- Orchard Seed Supply (SelectSeed Co. Ltd.)
- Extension and Communication
- Genecology and Seed Transfer
- Genetic Resource Decision Support
- Seed Orchard Pest Management

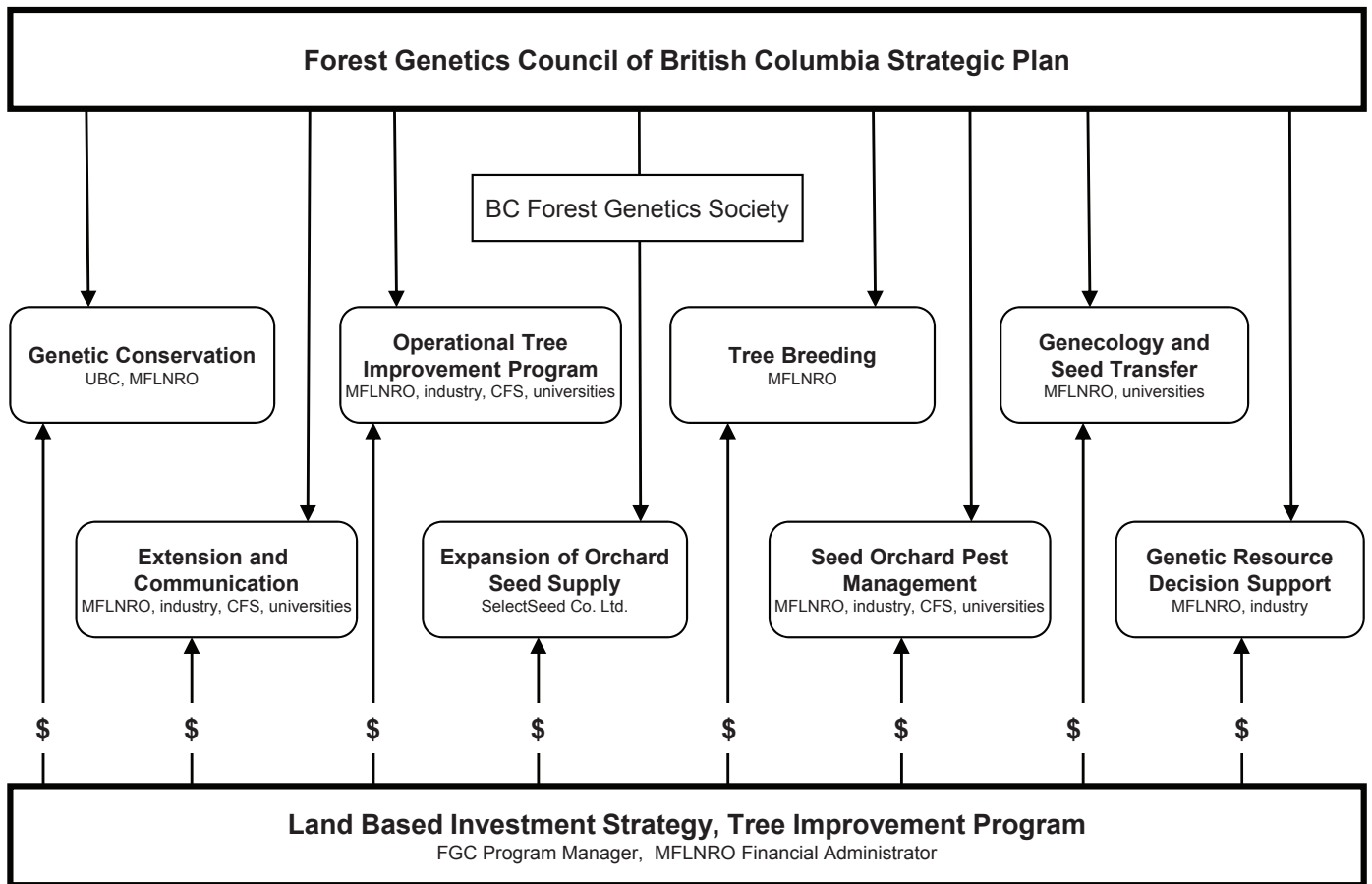


Figure 1. Relationship between FGC Strategic Plan, LBIS-TIP, and participants in various forest genetic resource management areas.

# 1.0 Expansion of Orchard Seed Supply Subprogram (SelectSeed Co. Ltd.)

Jack Woods

## Overview

SelectSeed Company Ltd. (SCL) is wholly owned by the Forest Genetics Council of BC (FGC) and mandated by Council to produce genetically selected tree seed for use on provincial Crown land in support of FGC objectives. SCL is also charged with providing management services to Council, including organizing meetings, developing business plans and annual reports, facilitating interactions, overseeing legal and accounting matters, and representing the FGC on issues related to genetic resource management in BC.

SCL is led by a five-member board of directors. The company generates revenue through seed sales that pay for seed orchard operations and for services provided to the FGC. All SelectSeed business follows an annual Business Plan that is prepared by management, reviewed and approved by the SelectSeed Board, and approved by the FGC.

## Seed Orchard Operations

Cone crops in 2013 were the largest to date for SelectSeed lodgepole pine orchards, moderate for interior Douglas-fir and nil for spruce (Table 1). Seed production, however, was lower for lodgepole pine than the previous year due to poorer average seed set. Figures 2 and 3 show cone and seed production by species and year since 2005.

Species	Total seed produced (kg)	SelectSeed share of seed produced (kg)
Lodgepole pine	102.2	76.3
Interior spruce	0	0
Douglas-fir	37.0	23.8

Table 1. 2013 seed production from SelectSeed orchards by species.

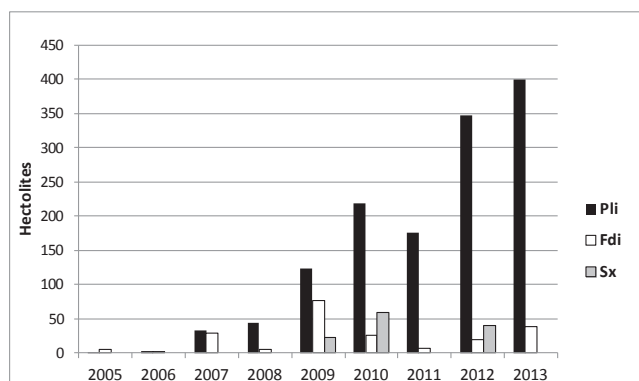


Figure 2. Cone production by species and year for all orchards (SelectSeed share of cones produced).

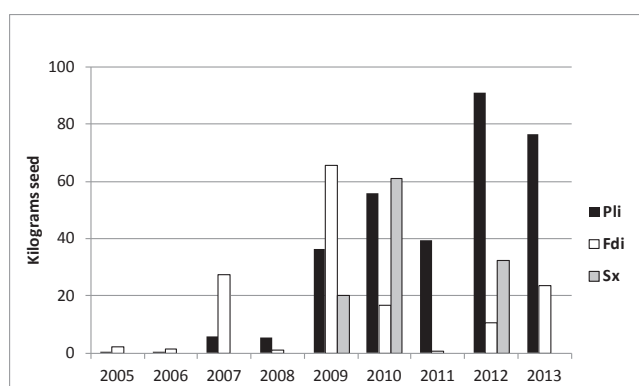


Figure 3. Seed production by species and year for all orchards (SelectSeed share of seed produced).

Seed set (filled seeds per cone) from lodgepole pine orchards continues to be the primary issue facing SelectSeed from both an operational and financial perspective. However, some orchards producing for southern seed planning units, and orchards located in the Kettle River Valley that produce for northerly seed planning units, are producing acceptable amounts of seed. Figure 4 illustrates average filled seed production across all seed orchards by species and year.



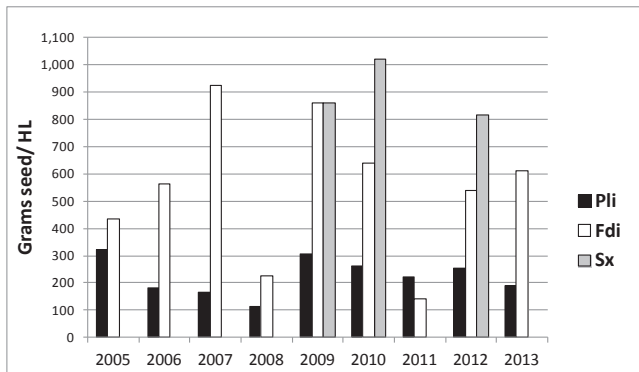


Figure 4. Average seed set by species and year across all SelectSeed orchards, measured as grams of seed per hectoliter of cones.

The total value of seed added to inventory in 2013 was \$709,309, based on SelectSeed seed prices (lower than FLNR prices), down from \$880,338 in 2012. This decrease is due primarily to a smaller lodgepole pine seed crop than the previous year. Seed sales, however, increased during the fiscal year to \$935,402 from \$595,324 the previous year. Business Plan estimated sales were \$900,000. Sales of unsold seed inventory following the 2012 sowing year allowed sales to exceed 2013 production value. At year end, SelectSeed held about \$129,000 worth of seed in inventory for sale in future years. During the sales year the SelectSeed customer base remained at over 70 clients, including BC Timber Sales, the FLNR, major licensees, first nations, woodlot owners, and community forests.

## FGC program management

FGC program management activities included the coordination of Business Plan development and reporting on progress indicators, as well as governance and organizational matters related to Council meetings and activities. The FGC was represented in numerous issues on seed, genetics, and policy matters. Reports and plans completed during the year on behalf of the FGC include:

FGC 2012/13 Annual Report

FGC 2013/14 Business Plan

Support was provided to Council's Technical Advisory Committees and species committees, and plans for seed planning units were maintained, updated, and made available for 54 provincial seed planning units. A website was also maintained on behalf of the FGC. A number of communication activities were completed, including formal and informal talks to various forest industry staff, conferences, and committees.

## SelectSeed management and administration

All SelectSeed financial and governance needs were completed. These include financial and legal matters on long-term seed orchard agreements, maintenance and audit of books of account, Company Act reporting requirements, Board of Director support, financial reporting on the SelectSeed Multi-Year Agreement, and reporting to the FGC.



Plate 1. SelectSeed board members with Kettle River seed orchard site manager Rick Hansinger (left) at the Kettle River lodgepole pine orchard producing for the Central Plateau seed planning unit. Board members are (right to left) Glen Dunsworth (chair), Henry Benskin, Reid Carter, and Russ Clinton (Jim Burbee not in photo). (J. Woods image)



## 2.0 Genetic Conservation Technical Advisory Committee (GCTAC)

Dave Kolotelo

The Genetic Conservation Technical Advisory Committee (GCTAC) is responsible for providing guidance, recommendations and reviews on projects and budgets related to genetic conservation activities for BC tree species. Several GCTAC members were able to attend a Whitebark Pine Recovery Planning meeting in Vancouver and contribute to the Federal strategy for the species. It was an interesting meeting and started with an evaluation of recovery feasibility and if we decided it was impossible then we would have all gone home. The evaluation indicated that reproduction and availability of critical habitat were not limiting factors (although future climate change challenges were acknowledged), but primary threat and recovery techniques both pointed towards the need for blister rust resistance screening.

The GCTAC has been actively funding collections of this endangered species and has initiated field trials for Whitebark Pine Screening for Blister Rust Resistance. We can fine tune our catalogue and talk about our lack of key indicators, but we have an endangered species in our province and we have the largest proportion of the species range. Unless the next FGC Strategic Plan isn't concerned with the maintenance of natural levels of genetic diversity for indigenous tree species, it is difficult to imagine that this priority species will not receive more attention within the context of a provincial action plan for whitebark pine species recovery.

I am quite pleased to be able to hand over the reins of chairing GCTAC to Dr. Pia Smets of UBC. I am very confident that Pia's technical knowledge, subject interest and excellent organizational skills will increase the capabilities of GCTAC to meet its challenges. I will continue to be actively involved in all aspects of GCTAC, but will focus my time on our seed bank and questions related to seed storage. Thank you for all your contributions and co-operation during my tenure.

## 2.1 Centre for Forest Conservation Genetics (CFCG) University of BC

Sally Aitken

In the 2013-14 fiscal year, we tested different approaches for developing genomic markers for population genetic studies, including targeted sequence capture and genotyping-by-sequencing. These methods have been developed as part of the AdapTree project, but we are evaluating their application to conservation questions as a substitute for more traditional allozyme and microsatellite markers. Targeted sequence capture works well, allows for separating targeted genes or genetic regions from the rest of the genome, and facilitates sequencing of these targeted regions (in our case, tens of thousands of genes). This method generates far more sequence data than necessary for most conservation questions, resulting in millions of single nucleotide polymorphisms (SNPs), and is very expensive (\$100 or more per sample). Genotyping-by-sequencing (GBS) is a much simpler approach, and generates data on variation throughout the genome at unknown locations for tens of thousands or hundreds of thousands of SNPs. We have also been testing optimal, efficient sampling designs for population genomic studies that allow for separation of adaptive patterns from neutral genetic diversity. Results will substantively change the way we assess population genetic architecture for conservation and management strategies.

BSc (honours) student Jonathan Degner conducted a pilot GBS study of a subset of trees in our Garry oak provenance trial for his undergraduate thesis, winning "Best Undergraduate Thesis" in the Faculty of Forestry. Sampling ~9 trees from each of 11 populations, he was able to elucidate patterns across the species range (from California to British Columbia) and determine that this species comprises two varieties, not three as has been previously described based on morphological traits. Genetic variation declines from south to north within var. *garryana*, the variety extending from the Oregon-California border to British Columbia. He is now analyzing additional individuals and populations to complete a manuscript on variation in Garry oak.

Two PhD students, Amanda De La Torre and Jill Hamilton, completed thorough investigations of the interior spruce (*Picea glauca* x *engelmannii*) and Sitka-white spruce (*P. sitchensis* x *P. glauca*) hybrid zones in BC for their PhD projects. While this research was mostly funded by NSERC and Genome BC, a small portion was supported

through the Genetic Conservation subprogram.

In 2013 and 2014, seven papers on the population genomics, phenotypic variation, and evolutionary history of these hybrid zones were completed or published. These hybrid zones are ancient, and the hybrid composition of populations closely reflects local ecological and climatic conditions. The high genetic diversity of hybrid populations has allowed for these species complexes to adapt across wide environmental and geographic ranges. The extent of hybridization in interior spruce extends farther north and east than previously thought. Breeding in interior spruce has favoured white spruce ancestry and decreased Engelmann spruce ancestry in these populations.

We continue to focus largely on capacity for evolutionary responses to climate change, and on the opportunities for forest management to facilitate adaptation to climate change. Tongli Wang continues to update ClimateBC and ClimateWNA (western North America) with new and higher-resolution climate data. These programs are now in wide use across western North America. Sally Aitken and population geneticist Michael Whitlock wrote a paper on the genetic advantages and risks of “assisted gene flow”, e.g., climate-based seed transfer. She co-authored three other review papers on experimental methodology and empirical evidence for genetic and plastic responses to climate change.

## Publications

Aitken, S.N. and M.C. Whitlock. 2013. Assisted gene flow to facilitate adaptation to climate. *Annual Review of Ecology, Evolution and Systematics* 44: 367-388.

Alberto, F.J., S.N. Aitken, R. Alia, S.C. Gonzalez-Martinez, H. Hanninen, A. Kremer, F. Lefevre, T. LeNormand, S. Yeaman, R. Whetten and O. Savolainen. 2013. Potential for evolutionary responses to climate change – evidence from tree populations. *Global Change Biology* 19: 1645-1661.

De La Torre AR, D.R. Roberts, S.N. Aitken. 2014. Genome-wide admixture and ecological niche modeling reveal the maintenance of species boundaries despite long history of interspecific gene flow. *Molecular Ecology* 23: 2046-2059.

De La Torre, A.R., T. Wang, B. Jaquish and S.N. Aitken. 2014. Adaptation and exogenous selection in a *Picea glauca* x *P. engelmannii* hybrid zone: Implications for forest management under climate change. *New Phytologist* 201: 687-699.

Franks, S.J., J. Weber, S.N. Aitken. 2014. Evolutionary and plastic responses to climate change in terrestrial plant populations. *Evolutionary Applications* 7: 123-139.

Hamilton, J.A. and S.N. Aitken. 2013. Genetic and morphological structure of a spruce (*Picea sitchensis* (Bong) Carr. x *P. glauca* Moench Voss) hybrid zone along a climatic gradient. *American Journal of Botany* 100:1651-1662.

Hamann, A. and S.N. Aitken. 2013. Conservation planning under climate change: accounting for adaptive potential and migration capacity in species distribution models. *Diversity and Distributions* 19: 268-280.

Hamilton, J.A., C. Lexer and S.N. Aitken. 2013. Genomic and phenotypic architecture of a spruce hybrid zone (*Picea sitchensis* and *P. glauca*) *Molecular Ecology* 22: 827-841.

Hamilton, J.A., C. Lexer and S.N. Aitken. 2013. Differential introgression reveals candidate genes for selection across a spruce (*Picea sitchensis* x *P. glauca*) hybrid zone. *New Phytologist* 197: 927-938.

Sork, V.L., S.N. Aitken, R.J. Dyer, A.J. Eckert, P. Legendre, D.B. Neale. 2013. Putting the landscape into the genomics of trees: approaches for understanding local adaptation and population responses to changing climate. *Tree Genetics and Genomes* 9: 901-911.

## 2.2 Tree Improvement Branch Conservation Activities

Charlie Cartwright

### Conservation Catalogue

Genetic conservation requires accurate inventories in order that risks can be recognized and the need for action evaluated. In the case of BC's forest trees, two recent catalogues have been produced. The first surveys *in situ* conservation status of indigenous species by biogeoclimatic zone and identifies environments in which particular species are threatened or their status is uncertain. The second document narrowed focus to the genetic conservation of operational species meaning those used most widely in commerce. Currently, a third catalogue is being put together to investigate the risks to minor indigenous tree species, the ones with less importance for commerce but still often utilized. This report covers the three native true fir species, mountain hemlock, ponderosa pine, and several broad leaves including red alder, bigleaf maple, paper birch, aspen and cottonwood. Completion is expected next year.

### Ex Situ Seed Collection

A reasonably good seed year for several species has allowed considerable progress in building the *ex situ* seed collections at the BC Tree Seed Centre. A considerable effort was put towards attaining whitebark pine due to its recent listing as endangered under the federal Species at Risk Act. In August cages were installed on 10 trees from each of 6 populations to protect against animal predation. Upon returning in mid-October (see Plate 2) a bounty of cones was collected for extraction and long-term storage of the seeds. Collections of a related species, limber pine, were much less successful, but progress was made with Rocky Mountain Juniper and subalpine fir. Our collection for Pacific crab apple was completed.



Plate 2. Remote stands of Whitebark pine in the Chilcotin.

## Whitebark Pine

After a disappointing delay of the proposed field tests of whitebark pine last year, it was a relief to receive permission to commence rust screening trials. Seed from 256 families derived from 43 provenances were put into stratification on November 16 (Plate 3), with scarification and sowing to occur in mid-March. As with some other high elevation species, this process is involved. Seed are taken from the freezer, put in mesh bags, then soaked for an hour to two in a peroxide solution. They are then moved to a bath with flowing tap water and additional aeration from an aquarium pump for several days. Following this treatment seeds are buried in sterilized sand and kept at room temperature for a month, then at 2 to 4° C for at least 3 months. Once out of the sand, the seed are scarified, (a razor blade is used to cut a small nick through the seed coat at the small end of the seed). With all the treatments completed we look forward to the notoriously uncertain germination of these valued plants.

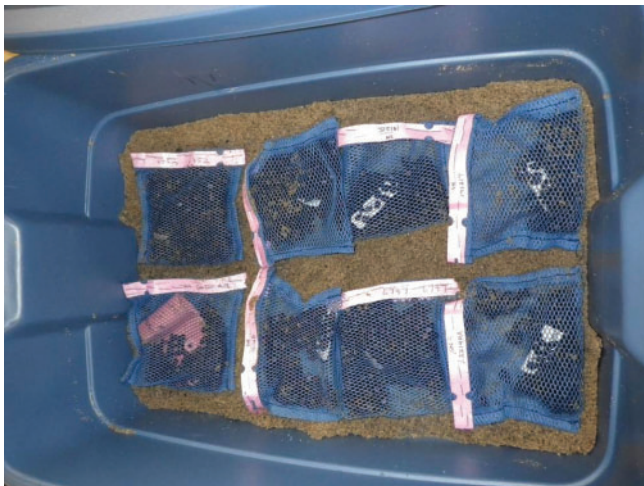


Plate 3. Mesh bags of seed are buried in sterilized sand for the 4 months in stratification of whitebark pine.

## Seed Bank Management Dave Kolotelo, Tree Seed Centre

This year we introduced the water activity ( $A_w$ ) technology for assessing whether our individual-tree genetic conservation samples could be freezer stored. Due to the value and size of these samples, destructive moisture content testing was deemed inappropriate and a backlog of samples had built up in cooler storage. Water activity measurement is non-destructive and provides a better indicator of seed longevity as it minimizes both biological and chemical deterioration agents. It basically narrows the range of acceptable moisture levels based on the thermodynamic properties of water. The technology is borrowed from the food industry and is been advanced with tree seed through work in England, France and Quebec. It is an exciting technology with a variety of applications. The Tree Seed Centre invested in a water activity meter and initiated testing the moisture content and drying them (if required) to moisture levels which would maximize seed longevity ( $A_w = 0.35 \pm 0.05$ ). Initially it was thought that calibration curves would need to be determined for each species and this would be difficult, time-consuming and counter-productive (consuming seed in our collections for calibration purposes), but there is a general consensus that the general figure of 0.35 will maximize seed longevity. During this fiscal year we were able to test dry-back and place in freezer storage a total of 542 individual tree samples from 45 populations. There is still a sizeable backlog of 774 samples which we hope to place in freezer storage next year.



## 2.3 University of Victoria Assessing range-wide genetic variation in subalpine larch (*Larix lyallii*)

### Marie Vance & Dr. Patrick von Aderkas

Climate change could negatively impact subalpine larch (*Larix lyallii*), a deciduous conifer found only at timberline in the Cascade and Rocky Mountains of the Pacific Northwest. For example, increases in the frequency or severity of late-summer drought events and/or increases in competitive interactions with other timberline conifers may impose strong directional selection on populations of subalpine larch. Since it is unlikely that this species will be able to migrate far or fast enough to escape the effects of climate change (Aitken *et al.* 2008; Gray and Hamann 2012) populations will be required to adapt *in situ* in order to avoid maladaptation and decline. Unfortunately subalpine larch may not be particularly adaptable. This species has a long generation time (average 500 years) and late arrival at sexual maturity (100 – 200 years), which will slow any potentially adaptive response to selection. In addition, a study of 19 populations in the northern part of the species range found low levels of genetic variation within populations (Khasa *et al.* 2006), suggesting that the magnitude of a potential response to selection could be limited. Low genetic variation in key traits could therefore prevent populations of subalpine larch from adapting quickly enough to avoid declining in response to relatively

rapid climate change.

To elucidate the genetic factors that may affect the long-term persistence in subalpine larch, we are working to assess genetic variation and genetic structure throughout the entire species' range. In the summer of 2013 we collected foliage samples from 44 populations of subalpine larch in the field and 17 populations of subalpine larch in the Kalamalka Seed Orchard in Vernon, BC, for a total of 1,478 trees from 61 populations distributed throughout the species range. Twenty western larch trees were also sampled to serve as an outgroup for phylogenetic analysis. DNA was extracted from dried foliage tissue using a modified version of the PL2 Extraction Protocol in the Machery-Nagel NucleoSpin 96 Plant II Core Kit. We had originally intended to use nuclear microsatellite markers (Khasa *et al.* 2000) and variable regions in the maternally-inherited mitochondrial DNA and paternally-inherited chloroplast DNA as our genetic markers but some issues with the microsatellites (null alleles, degenerate primers) and a lack of variation in tested organelle sequences led to the adoption of a new approach: restriction-site associated DNA sequencing (RAD-Seq). This will allow us to sequence thousands of short fragments (100 bp) associated with restriction enzyme cut sites throughout the genome in order to obtain single nucleotide polymorphism (SNP) data for hundreds to thousands of markers. This will allow us to assess genetic diversity in this species, providing information upon which future management and conservation decisions can be based. A subsample of individuals from each population have been selected. DNA is about to be shipped for analysis.



Plate 4. *Larix lyalli* images from BC. (Marie Vance images)

## 3.0 Tree Breeding

### 3.1 Coastal Douglas-fir Program

Michael Stoehr, Keith Bird, Lisa Hayton

#### EP708 Cassidy

To evaluate wood quality in various families of coastal Douglas-fir near their rotation age of 60 years, 12 families were destructively sampled to assess wood properties such as fibre characteristics of the wood, strength of the cut timber and its maximum dollar value that could be obtained. On Cassidy (ep708 Site #42), 170 families were planted in 1979 in a replicated test comprised of 2692 trees in total. The same set of families was also planted on 10 other test sites in coastal BC during the same year. The test trees growth (height, diameter and volume) was evaluated at age 12 years and 31 trees were selected based

on growth performance, juvenile wood density, stem form and branching characteristics in 1990 with some of these selections now growing in seed orchards. Now, at age 36 from seed, the trees here are approaching a harvestable size. We evaluated the wood, log and sawn timber properties of some of our selections, of some of the intermediate growing trees and of some slower growing trees on this test site. A total number of 100 trees were selectively harvested and assessed. Specifically, we measured log volume, density of the wood (specific gravity) and fiber characteristics important for wood strength. We sawed the logs into the most valuable structural lumber (6 by 6s, 4 by 4s, 2 by 4s) and tested their structural strength (modulus of elasticity, modulus of rupture). Data analysis is underway.

This study was spearheaded by the Forest Genetics section of the Tree Improvement Branch of the Ministry of Forests, Lands and Natural Resource Operations, the Canadian Forest Service and Yellow Point Propagation. Plate 5 shows the sequence of event.



Plate 5. From standing tree to 2x4s. Growth variables were assessed in the field, wood properties in the laboratory on sawn timber (D. Pigott images).



## Advanced Generation Breeding

Forward selections from the advanced generation breeding program Series 1, (selected in 2000) are producing pollen and seed cones. We took advantage of this and started making control crosses for the next round of breeding and testing (Plate 6, clone number 5017). Other selections were induced by girdling and GA applications. Series 2 final selections have been made using a combined spatial analysis of the GCA tests and the full-sib family tests. Final selections will have wood density estimates incorporated (reported in last year's issue). Breeding values for the best selections are listed in Table 2.



Plate 6. Insect bags on 5000 series tree pollinated to create next generation testing material.

### Series 2

Model 1			Model 3			Model 4		
Individual	BV	ACC	Individual	BV	ACC	Individual	BV	ACC
24286	33.1	0.747	24320	27	0.744	24320	26.8	0.748
24342	33.1	0.747	24261	24.9	0.744	24261	24.6	0.748
24151	32.6	0.751	23192	23.6	0.747	23192	23.5	0.751
24397	32.4	0.747	24317	22.7	0.744	23293	22.7	0.748
24320	32.3	0.739	24373	22.7	0.744	23690	22.7	0.751
23192	32.3	0.757	23293	22.6	0.744	23743	22.7	0.751
23690	31.9	0.757	23690	22.6	0.747	23744	22.7	0.751
23743	31.9	0.757	23743	22.6	0.747	24317	22.5	0.748
23744	31.9	0.757	23744	22.6	0.747	24373	22.5	0.748
23293	31.1	0.741	24286	22.5	0.746	25461	22.3	0.680
23907	30.5	0.757	24342	22.5	0.746	24286	22.2	0.751
25461	30.4	0.695	25461	22.1	0.676	24342	22.2	0.751
23566	30.3	0.749	24151	22.0	0.744	23580	21.7	0.750
23988	29.7	0.751	24397	22.0	0.746	24397	21.7	0.751
23568	29.6	0.749	24375	21.7	0.744	23907	21.7	0.751
Mean:	31.5	0.747		22.9	0.741		22.8	0.745

Table 2: Top-ranked breeding values (BV) and accuracies (ACC) of Series 2 forward selections derived from three models applied to complimentary progeny test of coastal Douglas-fir.

## Swiss Needle Cast Screening

Selected families from Series 3 are used for screening for Swiss needle cast. Three experimental populations are used: 1) seedlings growing in raised beds at Research Lab on Glyn Road for in-vitro screening, 2) seedlings planted under the full-sib Jordan River test (*in-situ* screening) and 3) seedlings growing in greenhouses at the Canadian Food Inspection Agency in North Saanich (in-vivo screening). In-vitro screening methods (Plate 7), were developed by David Noshad, our contract pathologist. From the raised bed seedlings, terpenes and phenolics have been extracted and analysed to explore potential associations with secondary metabolites and putative resistance. Single-tree heritabilities were found to be high and genetic correlations generally favourable (Table 3).

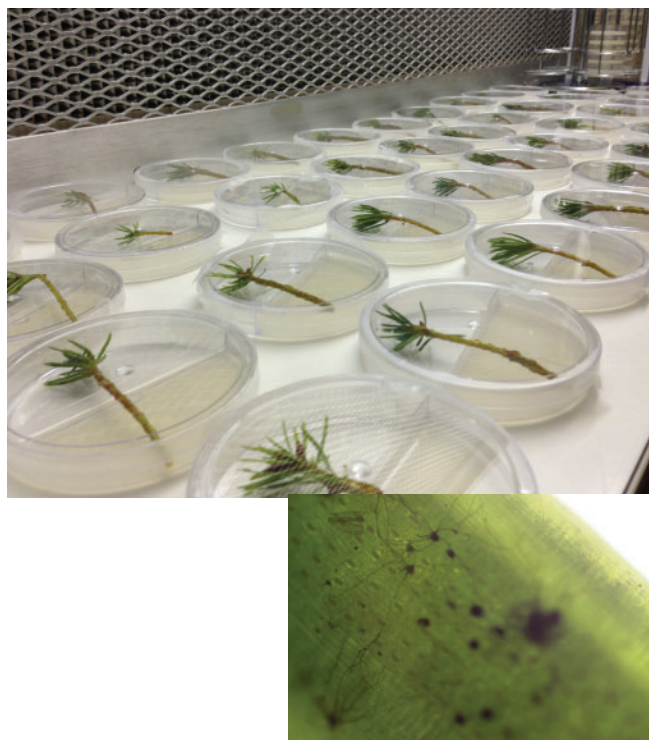


Plate 7. In-vitro testing set-up and Swiss needle cast fruiting bodies (*Pseudothecia*) with emerging hyphae (D. Noshad images).

Table 3: Heritabilities and genetic correlations of some terpenes and phenolics extracted from seedlings representing families from Series 4 of advanced generation coastal Douglas-fir program growing in raised beds in Victoria, BC.

	Taxifolin	3-Carene	$\alpha$ -Humulene	mono-Terpene	heritability
Trimethyl Gallic Acid	0.68	0.54	0.44	0.19	0.75
Taxifolin		0.10	-0.52	-0.76	0.74
3-Carene			0.93	0.76	0.81
$\alpha$ -Humulene				0.40	0.71
mono-Terpene					0.26

Table 3. Heritabilities and genetic correlations of some terpenes and phenolics extracted from seedlings representing families from Series 4 of advanced generation coastal Douglas-fir program growing in raised beds in Victoria, BC.



## 3.2 Western Hemlock Forest Genetics Program

### Charlie Cartwright

The forest genetics program for western hemlock in British Columbia is in the process of wrapping up. Attention is being focused on taking final measurements and putting trials into such condition that should information be required from them in the future, access will be guided by accurate notes, and test trees can be identified. To this end 8 low elevation trials (SPU 3) were attended to and a final measurement was made of the 3 full-sib clonal trials. For the progeny trials the data collected this year will be the last and breeding values for the program will soon be finalized. Maintenance was also undertaken for the 5 realized gain trials and 3 of those were also measured. Several more of the gain trials need to be wrapped up in the next several years and a report written to finalize that facet of the program. In contrast to this, provenance trials are part of a long term commitment to stewardship of the landbase, so will continue to receive attention. This year 12 installations were maintained and 3 were measured.

As with the low elevation breeding program, work for SPU 24, the high elevation seed planning zone, focused on concluding operations. To this end, maintenance was done on 4 test sites for which final measurements will be due next year. There are still several orchards for high elevation hemlock, so care is being taken to assure access to the progeny trials is well documented, and tree identities are clear in the tests.

## 3.3 True Fir Forest Genetics Program

### Charlie Cartwright

Though planting of true firs has declined over the last decades, they continue to be utilized in niches where their performance is strongest. The level at which they are used is not sufficient to justify intensive forest genetics programs, especially since they are largely unsuitable for seed production in orchards, but genecology trials are

a logical investment to set limits for seed transfer, and identify superior provenances for wild stand seed collection. As well, should interest in these species increase, perhaps for the purpose of enhancing species richness to improve stand resilience in face of climate change, necessary knowledge will be at hand.

Unfortunately the most frequently planted true firs are also the two for which our programs are least advanced. As many as half a million Pacific silver fir seedlings go out each year, but most of our installations are barely 10 years old. Three of these were measured this year, and maintenance was done on only 2 of them since most are well away and future attention will largely be on securing the identity of the individual trees through tagging them and removal of some large ingress. Reporting of results is expected next year.

For subalpine fir a million and a half are generally planted each year, but in contrast to the silver fir most of our trials are still at the establishment phase. Though there are three 10 year old nursery bed tests that need no attention, all 8 field trials required brushing. Since most of these young trees are entirely covered with snow all winter, full impacts of seed transfer are not apparent so analysis will be delayed until age 10 which will yield more meaningful results.

Trials for grand and noble firs were established in the 1980's and test sites are generally in great shape. In the case of grand fir, growth is very impressive and tours are frequently taken to the installations, both to witness the species potential and to see longer term consequences of long distance seed transfer. For the 4 installations of that species, checks were done of maintenance and labeling needs. (Our fishing lure like aluminum tags tend to attract the attention of elk). Though similar in age, the noble fir provenance tests are a different matter. They do show vigour exceeding that of other species in their area, but due to the elevation at which they occur, access to these 30 year old blocks is much more difficult and environmental damage considerably greater. Action taken with them this year was focused on the age 30 years measurement of height and DBH. Though data are not yet analyzed it was clear that most southerly seed sources were woefully mal-adapted. Current practice is to use only the Washington State seed sources, which is advisable given the results.

## 3.4 Western Redcedar Breeding Program

John Russell and Craig Ferguson

Our objective for the western redcedar breeding program is to develop a durable advanced generation population with potential cross resistance, or at least positive genetic correlations among resistance traits. Developing breeding populations that are resilient to multiple pests may not only give protection against the current target pests but potentially against future unknown ones. Growth and cedar leaf blight (CLB) severity are significantly correlated, and selections for increased volume production and CLB resistance are easily achieved. Genetic correlations between secondary extractives in the foliage and growth rate, and secondary extractives in heartwood and the foliage, although not strong, are positively low to moderate, allowing us some good potential for independent culling in future selections. Although this is certainly a simplistic measure and assessment of complex chemical pathways, it does give us a potential indication that there is minimal competition in chemical resources between foliage and heartwood extractives.

The first generation improvement program has been focussing mainly on selection for growth and cedar leaf blight resistance with some emphasis on heartwood durability through enhanced secondary extractives. The deer resistance program, which was developed after the other traits were already under selection, has been directed at elevating foliage monoterpene concentrations. This was achieved through rapid generation turnover, high selection intensity and early greenhouse testing. There was minimal information from the other populations to incorporate multiple trait selection. We currently have no information on CLB resistance mechanisms but as in many leaf disease studies, foliar monoterpenes have been implicated.

All seven series (48 test sites) of 1<sup>st</sup> generation polycross testing now have 10 year data and new breeding values will be released in 2015 and selections made for advanced generation breeding (Plate 8). Currently, F1 families using backward selections have been completed for the first four series of polycross testing (there are a total of seven). Seedlings from approximately 300 F1 families were established in 2 series of F1 seedlings trials (Plate 9).



Plate 8. A western redcedar 1<sup>st</sup> generation polycross test from the 7<sup>th</sup> series on Rutherford Main near Pemberton. Kirsten Mueller, a postdoc at SFU, examines a tree at this site in the CWHms1 (submaritime) at 800 m on a rocky slope with seepage water. There has been very good survival and growth up to 10 years after planting.



Plate 9. A western redcedar F1 trial on Big Tree Main near Sayward. Oldrich Hak, breeding consultant, and Tim Sexton, postdoc at UBC discuss breeding strategy at this zonal site in the CWHxm2 which has had excellent growth and survival after 2 years in the field.



Because of the partial confounding of breeding populations with selection objectives in the first generation population of screening, we are currently establishing clonal trials with a combined multi-trait objective in each of the growth/CLB and the deer resistance populations. For the growth/CLB traits, the above 300 F1 families were sampled for foliage monoterpenes in the greenhouse, and high monoterpene selections have been cloned through rooting. These clones have been bulked-up to produce enough cutting material for clonal testing in 2015 and 2016 for growth/CLB resistance across a number of environments.

For the deer resistance population, one-year-old seedlings from second generation full-sib breeding have already been tested for foliar monoterpenes and forward selections cloned for production hedges. Approximately 60 of these deer resistant clones were established in environments conducive to 1) good growth in the absence of CLB and deer browse (Powell River) and 2) cedar leaf blight and deer browse (north Vancouver Island). Another two sets of clonal studies will be established in 2015.

Selections for advanced generation breeding will be separated into partial diallels grouped by traits, and matings performed within and between groups with assortative mating within groups. In order to achieve an effective population size of around 150, the final population will be composed of: 1) 50 3<sup>rd</sup> generation forward selections from the deer resistant population further clonally selected for growth and CLB resistance; 2) 50 first generation parental selections based on clonal values for total heartwood extractive content with selections removed with poor parental volume breeding values and foliar monoterpene parental clonal values (independent selection, and; 3) 50 2<sup>nd</sup> generation forward selections for volume further selected for deer resistance. It will most likely not be necessary to include parents selected primarily for CLB resistance since all volume selections will be resistant to CLB and the deer population will be further tested for resistance. Currently, mechanisms for CLB resistance are unknown but being researched. It may be appropriate to make additional selections based on future potential resistance mechanisms for CLB.

Small-scale deployment trials of western redcedar with various levels of palatability have shown that deer prefer younger, smaller seedling stock with low terpenes compared to older, larger rooted cutting stock with higher concentrations of foliar monoterpenes (Plate 10). Deer resistant seedlots and veglots are currently



Plate 10. A western redcedar deer trial on Holt Creek Main near Duncan. The browsed trees in the foreground were one-year-old styro313 low terpene seedlings at the time of planting 4 years ago, and the larger trees in the background were three-year-old styro615 high terpene seedlings.

being made available to industry on a limited basis for operational deployment trials. Three annual sets of resistant material were planted out on central Vancouver Island in cooperation with TimberWest in 2011, Western Forest Products in 2012 and Interfor in 2013. These plantings will be monitored over the next few years to determine if browsing is reduced with mixtures of resistant seedlings and rooted cuttings. Preliminary results have shown that high terpene rooted cuttings are more resistant to browse than comparable seedlings.

A new western redcedar Genome BC project was funded focussing on genomic selection (GS) in the F1 breeding population to calculate Genomic Estimated Breeding Values (GEBV) for commercially important target traits, with heartwood durability being the main focus. The project has been structured into three phases. The first phase will run from April 2014 to September 2015. The study has partnered with Jörgen Bohlmann, UBC.

Older provenance trials have been maintained, pruned and permanent labels attached (Plate 11). Three new interior western redcedar provenance trials were established in the interior in spring 2014 in the southern ICH near Salmo, Revelstoke and Christina Lake bringing the total number of sites in the ICH to 16.



Plate 11. A western redcedar provenance trial near Jordan River. This trial was planted over 20 years ago on a very rich CWHxm2 site at 100m elevation. The trees in this trial and others in the same series expressed the first evidence of phenotypic variation in cold hardiness and cedar leaf blight resistance among populations in western redcedar.

### 3.5 Yellow Cypress Breeding Program

John Russell and Craig Ferguson

This program is currently focussing on maintenance and measurements of the clonal full-sib field trials. A select clonal population with a genetic gain of 20% volume has been established with serial propagated donors in greenhouses at CLRS. This elite veglot has the potential to be 35% greater in early height as compared to wildstand seedlots across a wide range of ecosystems within the yellow cypress maritime SPU. In addition, approximately 100 forward clonal selections based on growth and form will be established at CLRS for archiving and future advanced generation breeding.

## 3.6 Coastal Broadleaf Species Genetics Program

Chang-Yi Xie, Lisa Hayton and Keith Bird

### Red Alder

#### Range-wide provenance-progeny test

Nine provenance-progeny trials have been established (Plate 12 and Table 4) with the main objective of collecting ecological genetic information for developing seed transfer guidelines and selecting superior seed sources. About 10 families from each of 24 natural populations ranging from northern British Columbia to California were tested. Another test series of 4 sites is on the way and seedlings are growing at Cowichan Lake Research Station. Nineteen core populations will be included and the main objective of this test series is to estimate genetic parameters and make selections for future breeding and seed production.

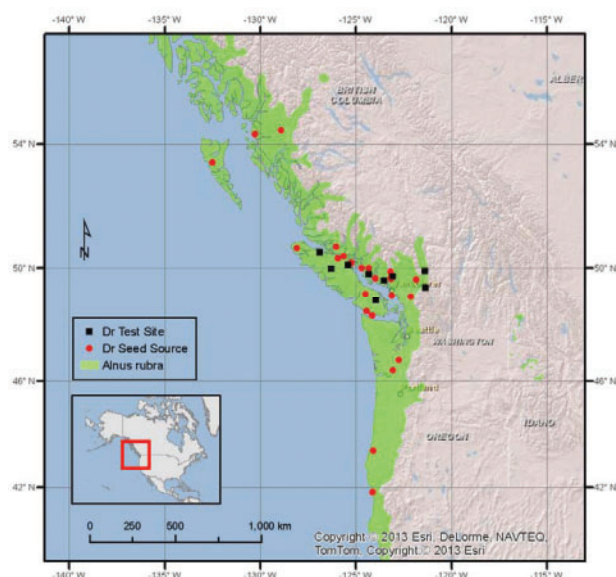


Plate 12. Locations of the 24 provenances and 9 test sites of red alder.

Site Name	District	Latitude	Longitude	Elev. (m)	BEC Zone
Mt. Brenton	S. Island	48 52 43	123 54 55	600	CWHxm1
Snowden Main	Campbell River	50 03 48	125 27 04	200	CWHxm1
Vernon Lake	N. Island-C. Coast	49 58 12	126 15 15	300	CWHmm1
Beaver Cove	N. Island-C. Coast	50 31 20	126 51 34	200	CWHvm1
Lois Lake	Sunshine Coast	49 47 58	124 18 04	180	CWHdm1
Rainy River	Sunshine Coast	49 33 57	123 30 14	200	CWHvm1
Mamquam	Squamish	49 42 57	123 02 12	480	CWH dm
Wrey Creek	Chilliwack	49 19 39	121 19 20	1200	CWH ms1
Styoma Creek	Chilliwack	49 54 29	121 21 58	1200	CWH ms1

Table 4. Red alder test site information.

### Bigleaf Maple

The second assessment (6 years) has been completed and data analysis is on-going.

Forward selections will be made by the spring of 2015 and growing techniques of bigleaf maple cuttings are currently under test.



Plate 13. Bigleaf maple cuttings.

### Black Cottonwood

A clone bank has been established at Saanich Seed Orchards. There are 106 clones and each has 5 copies. Those clones were selected based on 4-year performance at Harrison site and the cuttings were collected from UBC where no infection of *Septoria Musiva* has been noticed.



### 3.7 Coastal Western White Pine Tree Breeding Program

Nicholas Ukrainetz and Vicky Berger

The coastal western white pine breeding program has successfully incorporated local tested material with tested material from Oregon and Washington. After many years of collaboration with industry and the Canadian Forest Service, a formal F1 breeding program was initiated to take advantage of information gathered about local sources and families. Great effort was made to find the original parent tree selections in natural stands for breeding and pollen collection. The natural stand parent trees were ranked based on screening data and crossed to create a population of F1 progeny. Using collaborations with researchers in the United States, several control crossed families were acquired for testing in BC. The progeny of BC and US origins were deployed in 4 series of field trials on Vancouver Island, Texada Island and the Sunshine Coast; the seed for a fifth

series of F1 progeny tests is also available. These progeny tests will be a source of resistant and tolerant material for current and future seed orchards on the coast.

This year the white pine program was honoured by having one of its progeny selected as the 7 billionth tree planted in British Columbia. The ceremony took place at the UBC Malcolm Knapp Research Forest where the seedling was planted.

Height measurements were conducted on the Quinsam Lake and Duck Lake (series 3) sites and rust assessments were conducted at Duck Lake (series 3), Hallowell Road and Tom Brown North (series 4). This data will be analyzed and used to make additional selections for the coastal program. The seedlings for an additional testing series were sown for planting in the fall of 2014 and spring of 2015. This planting series will contain material from the coastal and interior programs and will be planted on two coastal sites (1 maritime and 1 sub-maritime) and 2 interior sites. A small demonstration site is also planned for Skimikin.

### 3.8 Interior Douglas-fir Tree Breeding Program

Barry Jaquish, Val Ashley, Gisele Phillips and Bonnie Hooge

The BC Interior Douglas-fir tree breeding program began in 1982 with the objective of producing improved and genetically diverse seed for planting stock on productive forest lands in south-central BC. Within this wide-ranging and ecologically diverse land base, six seed planning units (SPUs) were delineated for tree improvement. The first cycle of breeding in each SPU was based on: 1) phenotypic selection in wild stands, 2) establishment of grafted breeding orchards and clone banks, 3) progeny testing using open-pollinated (OP) seed collected from wild stand trees, 4) delayed clonal seed orchards established using backward selection based on early progeny test results, and 5) controlled mating to produce pedigree material for second-generation selection. The breeding goal is to improve traits related to tree size (height, diameter and volume) while maintaining wood relative density near old growth values. Moreover, the recent discovery of resistance to *Armillaria* root disease in Interior Douglas-fir suggests that resistance to *Armillaria* could become an important trait of interest. The first-generation progeny testing program includes 1,466 OP families and 31 test sites across the six SPUs.

Seed orchards were established for each SPU in the north Okanagan in the early 1990s and are starting to come into production. In spring 2013, approximately 4 million of the 12.8 million Interior Douglas-fir seedlings planted in BC came from seed orchards. In 2010, selected parent trees from southern SPUs were identified to establish new 1.5 generation seed orchards for the Thompson Okanagan high and low elevation SPUs. The Thompson Okanagan region was excluded in the program's early stages because of low site productivity; however, recent increases in planting numbers combined with severe seed shortages for the area necessitates the establishment of orchards for these lands.

The second-generation crossing program focuses on the Nelson SPU and includes selected parents from the old West Kootenay, Shuswap Adams and Mica regions. Moreover, since inter-varietal (coastal x interior Douglas-fir) hybrids have shown to be hardy and fast growing in the Nelson low elevation zone, the Nelson second-generation breeding population has been augmented with 16 high breeding value parents from the BC coastal breeding program and 16 forward selections from superior Submaritime provenances in the Trinity Valley range-wide

Interior Douglas-fir provenance test.

In spring 2013, 187 controlled crosses were completed in all of the Douglas-fir SPUs and 246 pollen lots were collected, processed and stored for future breeding. Controlled crossing for the Nelson SPU is about 90 percent complete and second-generation full-sib progeny tests should be established within three years. In fall 2013, maintenance and diameter at breast height (dbh) measurements were completed on four test sites in two seed planning zones: 1) Nelson low elevation SPU (25-years-old), and 2) Cariboo Transition SPU (30-years-old). Data analyses are in progress. New breeding values for parents from the Prince George, Mt Robson and Cariboo Transition/Quesnel Lakes SPUs were released in spring 2013.

In spring 2013, seedlings were grown to expand the joint BC FLNR/Canadian Forest Service Douglas-fir *Armillaria* resistance screening program. This second phase of screening includes 130 high breeding value parents from five SPZ's (East Kootenay, Cariboo Transition, Quesnel Lake, Prince George and Mica), 23 inter-varietal hybrids, 10 parents from phase I screening, and 15 wild stand control seedlots from the respective SPZ's. In spring 2014, 3,418 seedlings from the 178 seedlots were planted in 1 gallon pots and located in a greenhouse at Kalamalka Forestry Centre (Plate 14). The potted seedlings will grow for one year and then be inoculated with *Armillaria* transfer blocks currently in storage at Pacific Forestry Centre, Victoria, BC. The inoculation period is expected to last 2-3 years.

#### Publications

- Cruikshank, M.G. and B. Jaquish. 2014. Resistance and tolerance in juvenile interior Douglas-fir trees (*Pseudotsuga menziesii* var. *glauca*) artificially inoculated with *Armillaria ostoyae*. Forest Pathology. Doi: 10.1111/efp.12107.
- Rehfeldt, G.E., B.C. Jaquish, J. López-Upton, C. Sáenz-Romero, J.B. St Clair, L.P. Leites and D. G. Joyce. 2014. Comparative genetic responses to climate for the varieties of *Pinus ponderosa* and *Pseudotsuga menziesii*: Realized climate niches. Forest Ecology and Management. pp. 126-137.  
<http://dx.doi.org/10.1016/j.foreco.2014.02.035>

G.E. Rehfeldt, L.P. Leites, J. B. St Clair, B.C. Jaquish, C. Sáenz-Romero, J. López-Upton and D. G. Joyce. 2014. Comparative genetic responses to climate in the varieties of *Pinus ponderosa* and *Pseudotsuga menziesii*: Clines in growth potential. Forest Ecology and Management. pp. 138-146. <http://dx.doi.org/10.1016/j.foreco.2014.02.041>

G.E. Rehfeldt, B.C. Jaquish, C. Saenz-Romero, D.G. Joyce, L.P. Leites, J.B. St Clair, J. Lopez-Upton. 2014. Comparative genetic responses to climate in the varieties of *Pinus ponderosa* and *Pseudotsuga menziesii*: Reforestation. Forest Ecology and Management. pp. 147-157. <http://dx.doi.org/10.1016/j.foreco.2014.02.040>



Plate 14. One-year-old Interior Douglas-fir seedlings from 178 seedlots (open-pollinated seed orchard parents and inter-specific Douglas-fir hybrids) planted for screening for resistance to *Armillaria*. Note the white PVC plug included in each pot. In spring 2015, plugs will be removed and replaced with an *Armillaria* inoculated transfer unit.

### 3.9 Western Larch Tree Breeding Program

Barry Jaquish, Val Ashley, Gisele Phillips and Bonnie Hooge

In 2013, 5.2 million western larch seedlings were planted in B.C, 86 percent of which originated from seed orchards. In the second-generation crossing program, 32 crosses were completed and 47 pollen lots were collected and stored for future crossing. Second-generation crossing in the East Kootenay and Nelson SPUs is now about 80% and 92% complete, respectively. Ten year maintenance and measurement was completed on the East Kootenay realized gain genetic tests. Data analyses are ongoing.

#### Publications

Ratcliffe, B, F.J. Hart, J. Klapste, B. Jaquish, S.D. Mansfield and Y. A. El-Kassaby. 2014. Genetics of wood quality attributes in Western Larch. *Annals of Forest Science*. Vol. 71(3): 415-424. Doi: 10.1007/s13595-013-0349-4.



### 3.10 Interior Spruce Tree Breeding Program

Barry Jaquish, Val Ashley, Gisele Phillips and Bonnie Hooge

Interior spruce is the oldest tree improvement program in the BC Interior and is structured in two phases. Phase one began in the late-1960s with parent tree selection, grafting and progeny test establishment in three ecologically and geographically unique regions: Prince George, Bulkley Valley and East Kootenay. Phase two began in the mid-1970s and centred on the remaining geographic regions in BC where Interior spruce was commercially and ecologically important. The program has progressed to the point where 97 percent of the 77.5 million spruce seedlings planted in 2013 came from improved first-generation seed orchards. Phase 1 second-generation full-sib progeny tests were established in 1996, 1997 and 1999 for the Prince George, East Kootenay and Bulkley Valley SPUs, respectively. In the Prince George Series 1 program, 65 second-generation forward selections have been grafted and established in clone banks and breeding orchards. In the East Kootenay SPU, 75 second-generation parents were selected in fall 2009 based on 10-year tests measurements. Grafting was completed in spring 2010 and a new breeding orchard was planted at Kalamalka in spring 2012. Forward selections within the Bulkley Valley tests will be made in fall of 2014 based on 15-year measurements. In spring 2006, second-generation tests for the Prince George Phase 2 program were established on four sites (Skimikin, PGTIS, Burns Lake and Parsnip, north of Prince George). Controlled crossing is

now focussing on the Nelson low and mid-elevation SPUs. Unfortunately, the 2013 spruce flower crop was very poor and no crosses were completed.

Over this reporting period, maintenance and measurements were completed on nine progeny test sites in three SPZs: 1) Peace River – 4 first-generation open-pollinated (o.p.) sites (15-years-old), 2) Bulkley Valley – 3 second-generation full-sib sites (15-years-old), and, 3) Bulkley Valley – 2 first-generation open-pollinated (o.p.) sites (25-years-old). Seedlings were also grown to establish new 2nd-generation full-sib progeny tests for the Thompson Okanagan SPZ.

Approximately 12,000 terminal weevils (*Pissodes strobi* Peck.) were raised at the Kalamalka Forestry Centre (Plate 15) for release in genetic tests to: 1) screen o.p. families from parents in the Vernon Seed Orchard Company orchard 211 for resistance to terminal weevils on two sites (Plates 16 and 17), and 2) assist in the development of molecular markers for weevil resistance. Preliminary results from the screening of orchard 211 parents look very promising (Plate 18 and 19). At Kalamalka, 44 percent of the trees presented weevil top kill. Family top kill ranged from 9 percent (family 1843) to 76 percent (family 3792). Assessment of the second site (Skimikin Seed Orchard) is ongoing.

#### Publications

O'Neill, G.A., M. Stoehr and B. Jaquish. 2014. Quantifying safe seed transfer distance and impacts of tree breeding on adaptation. *Forest Ecology and Management*. 328 (2014) 122–130.



Plate 15. Weevil populations are reared by collecting infested interior spruce leaders in the spring and capturing emerging adults in cages where they are reared until release in fall the same year. Adult weevils overwinter in the duff and attack trees the following spring.



Plate 16. Weevil release into raised beds at Kalamalka in early October.



Plate 17. Three adult weevils were placed on each of the 3,920 two-year-old seedlings from 70 seedlots at two sites: 1) raised beds at Kalamalka Forestry Centre, and 2) nursery beds at Skimikin Seed Orchard.



Plate 18. Overview of 2014 weevil kill in raised beds at Kalamalka Forestry Centre.



Plate 19. Variation among families for weevil kill. Families located on the left and right side of the image are putatively resistant, while families in the centre are susceptible. Each family is represented by 7-tree row plots in four replicate blocks (28 seedlings per seedlot on each site).



### 3.11 Lodgepole Pine Tree Breeding Program

Nicholas Ukrainetz and Vicky Berger

The lodgepole pine breeding program has advanced to the second generation of progeny testing in five seed planning zones (BV, CP, PG, TO and NE). Within each seed planning zone (SPZ), 50 parents were selected for good growth, and 50 parents were selected for high wood density. Breeding was conducted among the parents within each group to create 65 controlled cross families. The families, along with several operational control and seed orchard (A-class) seed-lots, were deployed on 3 test sites within each SPZ. These tests will become a supply of high gain material which will be available to current and future seed orchards. The first generation, open pollinated progeny tests continue to provide information for updating seed orchard parent tree breeding values and opportunities to assess pest and disease resistance, while the provenance tests remain a wealth of information for modelling impacts of climate change and seed transfer regulations.

After substantial data analysis, several trees were selected from two of the three second generation progeny tests in the Prince George breeding zone. The analysis was complicated by significant genotype by environment interaction among the second generation test sites in this zone suggesting the need for refining zone boundaries. The top parents at the high productivity southern site planted in the ICH (Abbot Creek) were different than those in the lower productivity sites in the northern part of the zone. The final strategy was to select a complement of around 100 trees at the Abbot Creek site and at one of the two northern sites (Chief Lake). After data analysis, the selected trees were field verified and marked for scion collection (Plate 20). Scion was then harvested in the winter for grafting in the spring. A total of 92 trees were selected at the northern site (Chief Lake) and 109 were selected at the southern site (Abbot Creek). This material will be used for future breeding for the Prince George and Nelson zone, and will be incorporated into new seed orchards.



Plate 20. An example of the quality of the 12 year old forward selections that were made in the Prince George seed planning zone at the Abbot Creek site.

To further our understanding of the major pathogens affecting lodgepole pine, Nicholas has been in contact with Alex Woods, regional pathologist in Smithers, and Richard Reich, regional pathologist in Prince George. Nicholas and Alex visited the 1200A progeny test near Kispiox which was assessed for dothistroma resistance. The two visited several resistant and susceptible trees (Plate 21) and

21 A



21 B



Plate 21. Example of a tree that is resistant (A) and susceptible (B) to *Dothistroma* needle blight.

determined that the data collection and analysis procedure was appropriate for finding resistant trees. Richard and Nicholas visited three comandra resistant progeny tests and observed patterns of infection on the sites in relation to microsite variation and the location of the alternate host. Although there are few moderately resistant families in this population, there is an opportunity to find resistant trees (Plate 22).



Plate 22. A tree located in a *Comandra* blister rust resistance progeny test that appears to be resistant. It is located in an area of high mortality but appears healthy.

We are continuing to move the program forward by maintaining and measuring the second generation progeny tests. This year we measured and assessed the three sites in the Bulkley Valley. This data will be analyzed and used to select trees that will form the next breeding population and future orchard populations. We also had the Realized Genetic Gain (RGG) southern trials measured and assessed (4 sites; age 15), as well as a series of family provenance trials in the Big Bar and Chilcotin region (4 sites; age 10), and another family provenance trial established at high elevation in the Thompson Okanagan (5 sites; age 10). The two series of family provenance trials are growing on high elevation or low productivity sites so selections will likely be postponed until age 15. A third progeny test site in the East Kootenays was maintained and measured (age 9) after access was re-established. This site will help to calculate more accurate breeding values for parent trees in orchard 340 and can be used for obtaining new material for future orchards and breeding if desired.

Maintenance was conducted on second generation progeny test sites from two breeding zones (Nelson and Thompson Okanagan). These sites were brushed and



prepared for measurements which will occur next year. The northern series of the RGG trials were also maintained and prepared for 15-year measurements. We also directed resources to re-tag six open pollinated test sites in the Nelson and Thompson Okanagan zones. The integrity of the OP sites will be compromised if re-tagging does not occur in the next 3 to 4 years

Breeding has continued to focus on insect and pathogen resistance. The mountain pine beetle (MPB) resistance crossing program for the Prince George (PG) breeding population is nearly complete and breeding will shift to resistance against *Dothistroma* in the Nass-Skeena. The generous people at Skimikin have continued to clear and prepare a 3ha piece of land for our lodgepole pine clone bank and breeding arboretum. Planting will occur in the fall of 2014. We have continued to graft material from seed orchards and our old clone banks for archiving at Skimikin.

One of the recommendations at the lodgepole pine program review in 2011 was to investigate the potential of incorporating biotechnology and genomics into the breeding program. We will conduct a pilot study in lodgepole pine to create a genomic selection model for growth, wood density, microfibril angle, branch angle and western gall rust resistance. This model will allow us to rank trees for selection based on its molecular profile which will vastly decrease the amount of time required for testing. This project is in the planning stages and is expected to continue for the next 4 to 5 years.

### 3.12 Interior Western White Pine Tree Breeding Program

Nicholas Ukrainetz and Vicky Berger

The supply of blister rust resistant western white pine seed in the interior of BC is produced at the Bailey Seed Orchard (orchard 335) in Vernon. The seed orchard is composed of a combination of parent trees imported from Moscow, Idaho, and local seedlings from BC. The parent trees from Idaho were selected from a 17 year old, full-sib family screening trial growing at the Priest River Experimental Station. The full-sib families were created by inter-crossing tested and selected first generation parents. Seed from BC parent trees was screened for rust resistance at the Cowichan Lake Research Station. Surviving trees were selected for having good resistance reactions to white pine blister rust. Scion was eventually collected from selected trees, grafted and planted in the seed orchard. The

genetic material now located in the Bailey Seed Orchard will form the breeding population for future breeding activities. A second seed orchard was established at the Skimikin Seed Orchard site with a component of clones selected from the Bailey Seed Orchard.

The interior western white pine breeding program has focused on the inoculation of seedlings with white pine blister rust by placing seedlings beneath infected *Ribes* spp. plants at the Skimikin Seed Orchard site. Many controlled crosses were made among Idaho parent trees but few among parents originating in BC. We have developed a crossing scheme for interior white pine to cross Idaho and BC parents. BC parents were selected based on previous information from inoculation studies and orchard information about cone and pollen production. Crossing continued this year in the Bailey Seed Orchard. We have also expanded the clone bank and breeding orchard at the Kalamaka Forestry Centre which now includes all Idaho parents and all BC parents used in the current crossing program. We will continue to archive genetic material in the clone bank.

This year we completed maintenance on the realized gain trials which included brushing and checking tags. We have also taken responsibility for a series of progeny tests established by pathologist Dr. Michelle Cleary before she moved to Sweden. These sites include 33 families from the old Burton Seed Orchard site in Nakusp. The test includes 3 sites in the interior and 1 site on the coast (Plate 23). The trials were maintained this year and will be assessed in the near future.



Plate 23. An example of the "Cleary" sites which are a series of progeny test sites established by Dr. Michelle Cleary to assess resistance to white pine blister rust in 33 families.

### 3.13 Ponderosa Pine Genetics Program

Nicholas Ukrainetz and Vicky Berger

The Ponderosa pine genetics program consists of one well designed provenance test established in 1992 on two sites in the north Okanagan. The test includes provenances from throughout the range of Ponderosa pine in BC and the northern United States. After several years of measurements and data analysis, the provenance test site at the Skimikin Seed Orchard was converted to a seed orchard for operational seed production. Little seed has been produced and a component of the orchard was moved to the Bailey Seed Orchard site in Vernon. The data collected from these provenance tests will help us to assign pseudo-breeding values for parent trees in the orchards and develop seed zones. They can also serve as a source of material for future progeny tests.

Data from the ponderosa provenance tests and inventory data were used to generate a draft seed zone for review at ITAC. The zone was created by overlaying inventory data onto BEC7 spatial data to determine the BEC subzones in which ponderosa grows historically. An elevation limit of 1,000m was proposed based on the elevation of ponderosa populations in the provenance tests and seed orchards (Plate 24). This zone will be used to restrict planting of seed from seed orchards to ensure it is well adapted to the planting site.

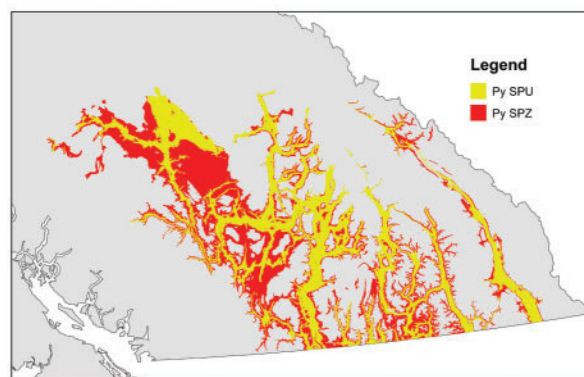


Plate 24. The proposed interior seed planning zone (Py SPZ) for ponderosa pine orchard seed with an elevation restriction of <1,000m (Py SPU).

## 4.0 Seed Transfer Technical Advisory Committee

### Lee Charleson

The Seed Transfer Technical Advisory Committee (STTAC) continues its work developing priorities in genecology research, vetting project proposals and providing budget recommendations to FGC for genecology and seed transfer research.

Funding of ministry genecology projects is done in accordance to the Genetics Section STTAC Strategy, 2011-2016, dated January 31, 2012. For genecology research outside of the ministry Forest Genetics Section, a call-for-proposals is administered each year. The objectives of the call supports FGC's strategic objectives and provincial seed transfer policy development.

Genecology and seed transfer projects are reported in this section and sections in the Centre for Forest Genetics Conservation and Breeding Reports.

Seed Transfer TAC will be a key advisory body in the development of climate-based seed transfer. The commencement of phase 2, policy development, is delayed until next year when a report is expected from the scientific analysis. The report will be a foundation piece for preparing evidence-based provincial policy.

## 4.1 Assisted Migration Adaptation Trial (AMAT)

### Greg O'Neill, Vicky Berger, Nick Ukrainetz and Michael Carlson

The AMAT (<http://www.for.gov.bc.ca/hre/forgen/interior/AMAT.htm>) is a long-term multi-species field trial intended to provide a better understanding of tree species' climate adaptation. The trial involves 48 seed sources (mostly orchard seedlots) from 15 species native to western North America planted at 48 test sites in BC, Yukon, and neighbouring US states. Growth and health are being assessed every 5 years. The first assessments began in fall 2013 on the 12 sites planted in 2009. Relationships of seedlot growth and health with plantation climate will be developed enabling identification of the seed sources most likely to be best adapted to current and future climates. The information will be used to revise BC's species and seed source selection guidelines, helping to ensure maximum health and productivity of BC's planted forests well into the future.

## 4.2 Interior spruce genecology/ climate change trial

Barry Jaquish and Greg O'Neill

A large interior spruce climate change/genecology field trial established in 2005 is beginning to provide valuable information that will guide seedlot selection for interior spruce. The trial consists of 128 seedlots (99 wildstand; 29 orchard) from BC, AB and neighbouring states and territories tested at 17 locations in BC, AB and YK. Age-6 growth was assessed in 2009 and is being used in the climate based seed transfer project (see Figure 5, below) to quantify safe seed transfer distance for spruce and to compare transferability of orchard and wildstand seed sources.

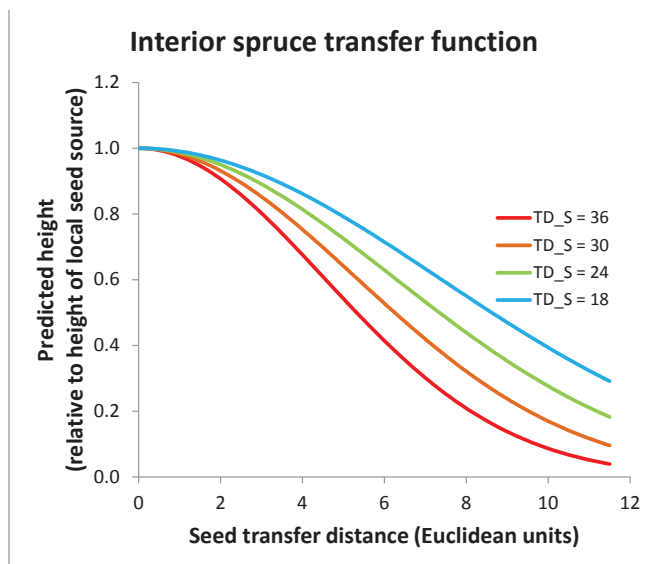


Figure 5. Site-specific transfer function predicting impacts of seed transfer on height of Interior spruce. Note: TD\_S is the temperature difference (between maximum warm month temperature and minimum cold month temperature) at the planting site. Euclidean transfer distance is a multivariate measure of distance that includes latitude and 5 climate variables.

## 4.3 Identifying critical soil moisture thresholds to support assisted migration practices and climate-based seed transfer in central British Columbia

College of New Caledonia  
Hardy Griesbauer

This research project seeks to provide scientific information and guidance with respect to selecting ecologically suitable sites when planting western larch and western redcedar as part of assisted migration in central BC. In this project, we will plant multiple seedlots of western larch, Douglas-fir and western redcedar across a range of site series in four biogeoclimatic variants in central BC, and monitor their survival and early performance as a function of actual soil moisture regime. This research will help identify species- and seedlot-specific critical soil moisture thresholds associated with seedling survival and early performance, and help guide species and seedlot selection for tree species planted outside their natural ranges.

In 2013, we located six research installation locations in the SBS wk1 (1 site), SBS dw3 (2 sites), SBS mc2 (1 site) and SBS dk (2 sites) biogeoclimatic variants. One of the research installations is located in the Wetzin'Kwa Community Forest near Smithers and one installation is located in the College of New Caledonia Research Forest north of Prince George. We installed four climate and soil moisture monitoring stations (one in each BEC variant) to collect site specific information. We are also using climate and soil data from the other research projects conducted by FLNR researchers Stephane Dube and Marty Kranabetter. In each installation, we identified five plots that spanned xeric to subhygric relative soil moisture regimes.

BC Timber Sales and Forests For Tomorrow silviculture foresters donated seedlings from eight seedlots to the project consisting of Douglas-fir (three seedlots), western redcedar (three seedlots) and western larch (two seedlots). We used the climateWNA model to select seedlots from a wide range of seedlots. We planted a total of 17 plots in four research installations in spring 2014. In three of the four installations, we did not plant the xeric plots because they were excessively rocky. Seedling survival and early growth will be measured in 2015. We note that there has been extremely low precipitation in this region over spring and summer of 2014, which may result in abnormally high seedling mortality.



Analysis of the effects of soil moisture, provenance climate, and annual climate on seedling survival and early growth will occur in late 2015 and will continue for at least five years. This research will provide scientific information regarding soil moisture thresholds on western larch, Douglas-fir and western redcedar seedling survival and early performance where planted outside of their natural ranges. This information will then be used to identify ecosystems (i.e., BEC variant and site series combinations) that may be ecologically suitable for planting these species as part of assisted migration strategies in central BC. Students from the College of New Caledonia and Northwest Community College will be involved in this research. Thus, this research project will help foster interest and knowledge about genecology in young forest professionals.



Plate 25. Hardy Griesbauer planting seedlings in a subhygric site in the Wet'zinkwa Community Forest near Smithers. (Amanda Follett/Wetzin'Kwa Community Forest image).



Plate 26. Western redcedar seedling. (Amanda Follett/Wetzin'Kwa Community Forest image).



Plate 27. Melissa Mjolsness preparing a subxeric plot near Prince George. (Leigh Anne Dutton/College of New Caledonia image).

## 4.4 UVic Summary report on amalgamated grant on:

### I. Seed shortfall in lodgepole pine and cone induction in lodgepole pine and Douglas-fir,

### II. Assessing range-wide genetic variation and genetic structure in subalpine larch, and

### III. Estimating female cone production of lodgepole pine in Southern B.C.

#### Project Ia. Seed loss in Pli seed orchards.

Lisheng Kong, Will Hintz and Patrick von Aderkas

A series of trials in recent years revealed a sudden drop in seed production in BC seed orchards. Seed orchards in the North Okanagan, where summer temperatures are high, have spectacular seed losses compared with seed orchards in Prince George, where the average summer temperature is much lower. It was thus supposed that trees originating from low temperature areas may not be adapted to high temperature. In 2013/14, samples were collected from seed orchards in the Okanagan and Prince George. Seed development was closely monitored during the period of seed shortfall. Initial investigation with fungus cultures and molecular identification of Pli seed borne fungi were also carried out. The results (2013/14) showed a dramatic seed shortfall in six out of the seven seed orchards under investigation, *i.e.* O-218 (Vernon Seed Orchard Company-VSOC), O-238 (Kettle River Seed Orchard-KRSO), O-241 (Sorrento Seed Orchard-SORR), O-307 (Kalamalka Seed Orchard-KSO), O-313 (GDV) and O-339 (Eagle Rock Seed Orchard-EGLR) in Okanagan, with the exception of seed orchard O-223 (Prince George Tree Improvement Station-PGTIS) at Prince George. The major shortfall had occurred before July 15 with low number of filled seed in three orchards, VSOC (O-218), KSO (O-307) and EGLR (O-339). In other orchards, the percentage of filled seed per cone (FSPC) diminished progressively to <50 % FSPC over the course of investigation. Based on average number of seed per cone, there was no direct correlation between seed shortfall and origin. Histological study revealed that substantial degeneration occurred in seed from all orchards, except PGTIS seed, which had low percentage of failed seed. A preliminary investigation of seed borne fungi was completed using sequence analysis of the internal

transcribed spacer (ITS region) of the ribosomal DNA (rDNA) repeat region. As a result, several fungi were found in the infected seeds in lodgepole pine, including *Sydowia polyspora/Rhizosphaera kalkhoffii*, and possibly *Alternaria arborescens*, and *Gibberella avenacea*.

#### Project Ib. Cone induction in lodgepole pine and Douglas-fir.

Lisheng Kong and Patrick von Aderkas

For Douglas-fir genotypes historically characterized by having no or low female cone yield, stem-injection of a combination of GA<sub>4+7</sub> and ethephon gave the best results for female cone induction. In lodgepole pine, among the treatments applied in 2012/13, stem-injection with GA either alone or in combination with cytokinin significantly increased female cone yield. For the first time, we were also able to induce female cone clusters with a single stem-injection. Genotype effects were more noticeable with bud treatments, *i.e.* bud paste or spray, than with stem-injection. Our results prove that with optimized protocols both stem-injection and bud spray could be industrially practical. With that aim in mind we developed refined protocols and applied them to ramets of both Douglas-fir and lodgepole pine in spring/summer 2013.

#### Project II: Assessing range-wide genetic variation and genetic structure in subalpine larch.

Marie Vance & Patrick von Aderkas

In 2013 we made significant progress towards our goal of assessing range-wide genetic variation and genetic structure in subalpine larch (*Larix lyallii*). First, we collected tissue samples from populations distributed throughout the species' natural range. In the summer of 2013 PhD candidate Marie Vance and UVic Co-op student Genoa Alger sampled 44 populations at sites in the Cascade and Rocky Mountains. Another 17 populations were sampled at the Kalamalka Seed Orchard in Vernon for a total of 61 populations. Second, DNA was extracted from all samples using a modified version of the Machery-Nagel PL2 DNA extraction protocol. Although we had originally planned to use PCR to amplify nuclear microsatellite markers and variable regions in the mitochondrial and chloroplast genomes to assess genetic variation, we are now pursuing a next-generation sequencing approach. DNA will be sent to Floragenex for library preparation and restriction site-associated DNA sequencing (RAD-seq). This approach will produce single nucleotide polymorphism (SNP) data for the analysis of genetic diversity and structure in subalpine larch.



### Project III: Estimating female cone production of lodgepole pine in southern BC.

Anne Berland & Patrick von Aderkas

The results of the study support our hypothesis that climate parameters impact the number of cones produced on lodgepole pine trees in provenances ranging from across the species' range. We made use of sixteen sites from the lodgepole pine provenance trial established by Illingworth in 1974. The effects of site and provenance on cone yield rankings were significant. Using predictive models for cone yield that included both temperature and moisture terms, we were able to identify values of climate variables associated with peak cone yield. These ranges in values were used to produce maps depicting areas of high suitability of cone production for the included provenances.

## 5.0 Decision Support Advisory Committee

Susan Zedel

The Decision Support Advisory Committee (DSAC) is responsible for identifying the needs for clients, exploring decision support options, developing proposals and submitting the proposals and budget to the FGC for approval and support. The committee works closely with Tree Improvement Branch (TIB) on project management and development of decision support tools.

### Development of ArcGIS Online Mapping Tool for Climate Based Seed Transfer

A Request for Proposal process for GIS (Geographic Information System) work for TIB was completed in the summer of 2013 and a contract awarded to Forsite Consultants Ltd. (Salmon Arm) in late August. The focus in 2013-14 was the development of decision support mapping tools and map products to support the scientific analysis for the Climate-Based Seed Transfer (CBST) project. Forsite has successfully taken data provided by Greg O'Neill and developed an ArcGIS Online mapping application that is currently being used by the CBST science working group to review multiple species, risk levels and options for seed transfer. Forsite created the spatial data for the Ponderosa Pine (Py) South Interior (SI) seed planning zone and seed planning unit. As a result of this project in 2014, Forsite and TIB were the recipients

of ESRI Canada's Pacific Region Award of Excellence in recognition of innovative use of web GIS tools for collaboration and decision support.

### GIS Analysis and Map Products for various Tree Improvement activities

Using the GIS contract with Forsite Consultants Ltd, TIB has been able to request the production of ArcGIS maps to assist with the analysis of the *Chief Forester's Standards for Seed Use* seed transfer limits in relation to new climate based stocking standards developed by Resource Practices Branch. Maps were also produced for the TIB forest genetics section as required.

In early 2014, the TIB Policy and Planning section and Barry Jaquish conducted a review of the western larch (Lw) seed planning zones developed in 2010 for the range and population expansion of that species. Clients found the original 2010 zones to be too limiting in area and difficult to use due to the blocky geometry of the zones. Forsite produced ArcGIS Online maps of the Lw zones using the original spatial data for both one and two climate model concurrences. This enabled TIB to review the data again, and merge LW1 and LW3, which use the same Lw seed source, into the LW1 zone. Forsite also used algorithms to smooth the spatial geometry into more usable polygons. This work was critical in enabling the review of the seed planning zones and policy for Lw within a short time-frame. The 2014 version of the LW1 and LW2 zones and the Py SI zone were loaded into the BC Geographic Warehouse in September 2014.

### Seedlot Spatial Area of Use

The goal of this project is to create spatial area of use data for all seedlots registered in SPAR (Seed Planning and Registry system). This project was started at the beginning of the 2012-13 fiscal year with the contractor Vivid Solutions (Victoria) and was to be completed by December 2013. There has been a series of delays in the development work and information systems infrastructure over the duration of the project. The project was not completed in 2013-14 with DSAC funding, but is continuing with funding from the SPAR maintenance budget. The spatial data will be loaded into the BC Geographic Warehouse in 2015 once the Corporate Services for the Natural Resource Sector infrastructure and server capacity can accommodate the data.

## 6.0 Operational Tree Improvement Program

### Darrell Wood

The objective of the Operational Tree Improvement Program (OTIP) is to increase the quality and quantity of select seed produced from existing industry and Ministry of Forests, Lands and Natural Resource Operations seed orchards.

To meet this objective, a Call for Proposal process is administered each year in support of FGC objectives and based on priorities developed by the Interior and Coastal Technical Advisory Committees. FGC committees review and rank these proposals based on technical merit, impact, value and costs.

Funding for this work comes from the Land Based Investment Strategy Tree Improvement Program and this investment assists in:

- Boosting genetic gain in seed orchards through grafting, ramet removal and replacement, and pollen management
- Boosting seed production through induction, pest management and supplemental pollination, and
- Supporting technical projects that address issues preventing orchards from meeting production objectives.

OTIP uses a performance measurement system to monitor progress and set reasonable targets for project success. This year, as in past years, orchardists and researchers have responded to this approach and have achieved and exceeded planned targets.

For additional information regarding the budget and key performance indicators, please refer to the FGC Annual Report 2013/2014 at <http://www.fgcouncil.bc.ca/FGC-AnnualReport-1213-04Nov2013-web.pdf>

## 6.1 Orchard Projects

### 6.1.1 Saanich Forestry Centre (WFP)

#### Annette van Niejenhuis

Western Forest Products manages orchards and hedges for the Maritime Zone at the Saanich Forestry Centre on the Saanich Peninsula. Our orchards and hedges cover seven Seed Planning Zones: low and high elevation Douglas-fir, low elevation western redcedar, low and high elevation western hemlock, low elevation Sitka spruce seed orchards, and yellow cypress hedges. Our orchards primarily deliver volume gain to reforestation programs; the low elevation Sitka spruce orchard delivers pest resistance. Upon recommendation of the Forest Genetics Council, WFP receives OTIP funds to implement incremental orchard management techniques to deliver better quality seed in greater quantity where best seed is in short supply.

#### Low Elevation Coastal Douglas-fir Crop and Orchard Enhancement

Orchard development continues in response to additional new and available selections from the breeding program, and to orchard mortalities. We acquired and held 245 replacement grafts, and we replaced, filled or replanted 137 orchard positions in the low elevation orchards.

Cultural management continued with fertilization of young replacement stock for good crown development (Plate 28). Our low elevation orchards have average breeding values of 17% (Fdc Orchard166 includes mature stock) and 19% (Fdc Orchard 405) for volume; however, newer high-gain ramets do not yet contribute fully to the crops. Phenological and reproductive bud surveys together with pollen monitoring were used to determine the male parental contribution to the seed crop. Early and late clones, together with small trees in the young orchard, received supplemental mass pollination. Insect pests were low in the small 2013 crop.



Plate 28. Straw is used to mulch Douglas-fir replacement stock in orchard 166.

#### High Elevation Douglas-fir Orchard Enhancement

In response to revised scores, we replaced 54 ramets in the high elevation Douglas-fir orchard (removed 4 selections and added 5 new selections). This raised the breeding value to 13% for volume. Nutrient management for the promotion of good crown development continued.

#### Western Redcedar Crop and Orchard Enhancement

Pollen management of the 2013 crop was completed in the 4<sup>th</sup> quarter of 2012-13. An infestation of western redcedar cone midge (*Mayetiola thujae*) required treatment in the 1<sup>st</sup> quarter of this year. Treatment was successful, but damage may have been done; the small crop had a light yield for 0.6 mil plantables. This crop, with a Genetic Worth 17, represents the continued improvement of this orchard. Nutrient management of the replacement stock continued. Induction of 128 trees led to estimates of a good crop for 2014. Pollen monitoring and management was undertaken in the 4<sup>th</sup> quarter, and midge monitoring indicated no concerns.



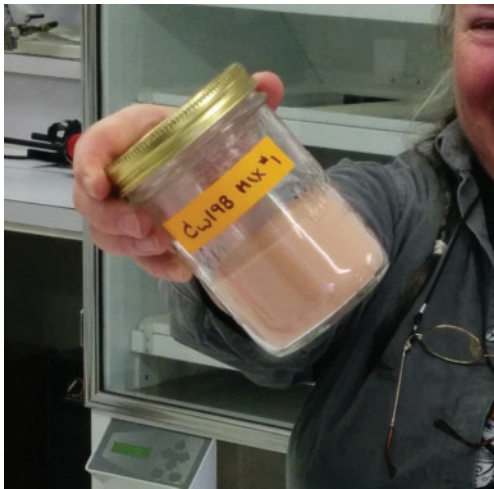


Plate 29. Western redcedar pollen is harvested from a number of clones, measured for clonal contribution estimates, and blended, prior to its application to receptive female flowers. This effort reduces pollination from outside-orchard sources, and may reduce self-pollination rates in western redcedar.

### Sitka Spruce Orchard Enhancement

As the current seed supply of weevil resistant Sitka spruce seed is adequate for a number of years, and we are awaiting scores for growth as a secondary trait, only orchard maintenance and foliar pest management activities were approved for OTIP funding. Green aphid surveys indicated population levels below treatment thresholds, and no other foliar insects showed significant presence; no treatments were required.

### Yellow Cypress Production Hedges Enhancement

Potted donor stock and ground based donor stock identified for growth and rooting success are used to deliver 20% volume to our yellow cypress regeneration program. Continual renewal of the hedges is required to maintain high nursery recoveries of stocklings. Maintenance of the stock (Plate 30) included fertilization and tip-pinching to promote more cuttings production. Cuttings were supplied for 300k plantables (Plates 31 & 32). Nematodes were deployed to combat root weevils with good success, and surveys for *Trisetacus* mites were completed; treatment for mites will be undertaken. Clones showing plagiotropism or reduced rooting were removed from the production population, and 25 new top selections from the clonal trials have been added to the production population.



Plate 30. Periodic replacement of yellow cypress donor stock is needed to maintain rooting in the selected clones.



Plate 31. Cuttings – shoot tips – are harvested from the yellow cypress hedges and shipped to nurseries where they are grown in an environment to promote rooting. These hedges deliver stocklings with a volume gain of 20%.



Plate 32. These yellow cypress potted hedges have been harvested; the cuttings will be rooted and shipped to plantations. This stock will outperform wild stock by 20% volume at rotation.

## 6.1.2 Mt. Newton Seed Orchard (TimberWest Forest Company)

Tim Crowder and Bevin Wigmore

### SPU0106 Enhancing Productivity and Gain in Coastal Douglas-fir Orchards

This project covers activities in four Douglas-fir Maritime low elevation orchards: 134, 154, 183, and 197. These orchards are similar in composition and cone collections from the younger orchards are typically blended together. The management objective for these orchards is to produce enough seed for at least 5 million seedlings annually, while increasing the genetic gain from the current 12% to approximately 20% by 2020.

Supplemental mass pollination was used this year to augment the insufficient pollen cloud on 480 trees of the early- and late-flowering clones and crop trees in the young orchards. Six litres of pollen was collected and extracted.

GA<sub>4/7</sub> and girdling was used to induce a 2014 crop on 680 orchard trees with high breeding values.

Insect pests have proved to be a serious impediment in achieving the full potential of the cones produced in these orchards. A monitoring and control program was implemented and approximately 860 crop trees were

sprayed with an air-blast sprayer for control of *Contarinia* midge.

The 2013 cone crop yielded a total of 120 kg of seed with a weighted average genetic worth of 13%.

107 large 25+ year old trees were removed from the orchards, and 350 new trees were established in vacant spaces. 864 young ramets were pruned and fertilized in the orchards, and an additional 595 were maintained in the holding beds.

500 additional trees were grafted at the orchard in the fourth quarter.

### SPU0205 Enhancing Productivity and Gain in Western Redcedar Orchards

This project covers activities in two western redcedar Maritime low elevation orchards: 140 and 152. These two orchards are similar in composition and are typically induced in alternating years to provide a steady seed supply. The management objective for these orchards is to produce enough seed for 2 million seedlings annually with a genetic volume gain of 20%.

A total of 224 orchard trees were maintained and managed. 173 non-crop trees in orchard 140 were induced with gibberellic acid for a crop in 2014. Holding beds containing 325 grafts were maintained, in order to accelerate delivery of gain when progeny test results are made available.

The 2013 crop was harvested in the third quarter and yielded 4400 grams of seed sufficient for 1.07 million potential seedlings.

In the fourth quarter, the developing 2014 crop was surveyed for midge. The infestation was light so chemical control was not required.



Plate 33



Plate 34

Plates 33 and 34 . March 2014, Tim Crowder and Terry Simpson moving large cedar trees from the holding bed into orchard 140.



## 6.1.3 Saanich Seed Orchards

Lisa Meyer

Saanich Seed Orchard on southern Vancouver Island manages 5 seed orchards covering 5 different SPUs. Our orchards include maritime and sub-maritime Douglas-fir (Fdc), maritime western redcedar (Cw), rust resistant maritime western white pine (Pw) and maritime western hemlock (Hw). In the 2013/14 season 4 of the 5 SPUs received OTIP funding for activities to manage and increase seed production and the overall genetic gain in the orchards. Table 5 shows the projects.

### Genetic Enhancement and Production of Seed Crops from Coastal Douglas-fir Seed Orchard 199 (SPU 0114)

The management objective of this project is to improve the genetic quality and the quantity of seed crops produced from the Douglas-fir Seed Orchard #199 at the FLNR Saanich Seed Orchard site. This moving front orchard design allows early seed production, and accommodates for changes in the orchard because of revised genetic gain scores and addition of new parents to the orchard. The interim orchard will gradually give way to the final orchard. All ramets that were less than 6 years of age were managed by out planting of ramets from holding beds, roguing, crown and basal pruning, fertilization and foliar sampling. Insect monitoring was performed throughout the year in the entire orchard. The funded project deliverables are identified in Table 6.



Plate 35. Supplemental mass pollination in orchard 199.



Plate 36. Douglas-fir conelets and pollen buds in orchard 199.

### Orchard Management Operations to Maintain Productivity and Increase Gain in Seed Orchard 181 SM Fdc – Coastal Douglas-fir (SPU 1902)

Objectives for Orchard #181 are to enhance the seed yield and genetic worth and quality of seedlots produced.

The management is based on progeny test results, utilizing orchard management techniques to optimize growing stock vigour and crop health, and monitor for and apply pest control if necessary. Grafts were not added to the orchard this year because no new genetic gain information was available. Future test scores will result in parents being added or removed from the orchard. All ramets that were less than 6 years of age were managed by out planting of ramets from holding beds, roguing, crown and basal pruning, fertilization and foliar samples. Insect monitoring was performed throughout the year in the entire orchard. Refer to Table 6 for specific SPU activities.



Plate 37. Douglas-fir conelets Orchard 181.

All images by Chris Halldorson.





Plate 38. Douglas-fir pollen buds in orchard 181.

### Orchard Management and Seed Crop Production 184 Cw - Western Redcedar Seed Orchard (SPU 0218)

Orchard #184, western redcedar orchard, a greenhouse based orchard, relies on supplemental mass pollination (SMP) for seed production. Stored pollen from past year collections was applied to the early and late receptive females. In addition pollen from ramets with a genetic gain or BV of 15 or greater was collected in the spring of 2014, mixed and applied to the mid-season receptive females. The remaining pollen was processed and stored for future applications. To induce a crop for next year, gibberellin (GA3) foliar spray was applied to ramets that did not carry a crop. This year there was a noticeable increase in the presence of tussock moth, black vine weevil and Cw cone midge. Monitoring and control with BTK, Pounce and Met 52 was effective to reduce the population of these insects. Table 6 highlights the specific SPU activities.



Plate 39. Western redcedar pollen collection in orchard 184.



Plate 40. Western redcedar conelets and pollen buds in orchard 184.

### Orchard Management and Seed Crop Production 175 - Rust Resistance Western White Pine (SPU 0804)

FLNR Saanich Seed Orchard is working with the western white pine breeding program to development a rust resistant white pine orchard. Supplemental mass pollination techniques are being employed to apply Major Gene Resistant (MGR) pollen to the Slow Canker Growth (SCG) and Difficult to Infect (DI) white pine ramets. The deliverables from this project is a vigorous orchard that is capable of producing SCG and DI seedlots of rust resistant seedlings. The advancement of this orchard is based on results of the progeny tests that are currently being conducted through the genetics program. The OTIP proposal provided funding for replacement ramets, but because the tests results were not completed and no new information was available, monies for those replacement ramets were returned. See Table 6 for specific activities.



Plate 41. Western white pine 2 year cones orchard 175.



Plate 42. Orchard 175, western white pine with fog rolling in.

PROJECT	SPECIES	ORCHARD #	IN ORCHARD	IN HOLDING
SPU 0114	Fdc	199	1417	548
SPU 1902	Fdc	181	874	417
SPU 0218	Cw	184	405	50
SPU 0804	Pw (R)	175	589	80

Table 5. Saanich seed orchard projects.

PROJECT	GRAFTS PURCHASED	POLLLEN COLLECTED (litres)	SMP APPLICATION	GA APPLICATION	RAMETS ROGUED	RAMET REPLACEMENT	HOLDING	PEST MANAGENT
	321	325	325	326	324	323	322	341/343
SPU 0114	250				20	1417	548	1417
SPU 1902		1.0	874			874	417	874
SPU 0218		0.4	405	250				405
SPU 0804		1.0	452			452	80	452

Table 6. Saanich seed orchard funded project activities.

## 6.1.4 Kalamalka Seed Orchards

Gary Giampa

In 2013/2014, Kalamalka Seed Orchards received OTIP approval for 10 orchard enhancement projects under the operational production sub-program. The funding allowed for a significant enhancement of the effectiveness of our orchards in delivering improved seed. Activities included:

- Improving orchard composition through roguing, grafting higher-breeding-value ramets, maintaining recently grafted high-value ramets destined for orchards, and transplanting the older higher-value ramets to the orchards;
- Improving orchard seed quantity and quality through cone induction and pollen management, including collecting high-breeding-value pollen from clone banks and applying supplemental mass pollination; and
- Improving orchard productivity through pest management and other management activities.

Project	Species	SPZ	Orchard	Grafts Made	Rogue	Transplants	Induction
SPU0401	Sx	NE	305		301		
SPU0502	Sx	NE	306		330		
SPU0701	Pli	NE	347 / 307		250	55	
SPU1302	Lw	NE	332				
SPU1501	Pw	KQ	335				
SPU1708	Pli	BV	230	100		131	
SPU2201	Fdi	NE	324				600
SPU3201	Pli	EK	340		115	40	
SPU3901	Fdi	EK	336				300
SPU5101	Py	SI	346				
<b>Totals</b>				100	996	171	900

Table 7. Orchard Composition Activities by Project.

Project	Species	SPZ	Orchard	Pollen Collected (litres, dry)	Trees Pollinated
SPU0701	Pli	NE	347 / 307	3	1249
SPU1501	Pw	KQ	335		1300
SPU1708	Pli	BV	230	2	822
SPU2201	Fdi	NE	324	2	1300
SPU3201	Pli	EK	340	3	1701
SPU3901	Fdi	EK	336	1	500
SPU5101	Py	SI	346		100
<b>Totals</b>				11	6972

Table 8. Pollen Management Activities by Project.



Pest management activities included:

- monitoring pest levels to make informed decisions regarding control,
- using Safer's Soap sprays to control adelgids in Sx and Fdi,
- removing weevil-infested spruce leaders to reduce weevil populations,
- removing pine pitch moths damaging orchard tree stems,
- hand removal of European pine shoot moth from young Pli grafts,
- baiting for control of rodents feeding on tree roots,
- sanitation picking of cones in orchards with non-collectible crops to reduce pest populations,
- spraying to control *Dioryctria* in Fdi,
- spraying to control mites in Fdi,
- applying dormant oil to control larch adelgids.

The OTIP funding was instrumental in increasing both the quantity and quality of seed produced. At Kalamalka in 2013, we produced approximately 90 kg of western larch, interior Douglas-fir, Ponderosa pine, lodgepole pine, and western white pine seed equivalent to over 10 million seedlings with an average Genetic Worth of +15. Large areas of the interior of the province are using Kalamalka seed.



Moving day for the 2014 Kalamalka cone harvest.

Plate 43. Moving day for the 2014 Kalamalka cone harvest.



## 6.1.5 Vernon Seed Orchard Company (VSOC)

### Dan Gaudet

Operational Tree Improvement Program (OTIP) annually provides funding for seed orchard production companies to successfully produce improved seed for Industry and the Province of British Columbia (BC). With OTIP funds, orchards are able to collect scion and graft, control pests, collect pollen for breeding, as well as a multitude of other vital tasks that are required to produce the best seed available.

As we consistently achieve the Forest Genetics Council (FGC) objectives, we also continue to increase the gain, health, production levels and standards of the tree improvement programs of BC. Vernon Seed Orchard Company has been diligent in fulfilling contract obligations through required reporting mechanisms. This has been very successful in assuring continued growth and accountability for this important provincial program.

### Key Program Activities for VSOC in 2013/14

#### SPU 1202 Enhancing the Effectiveness of Prince George Orchard 222 - Pli

Lodgepole pine is a crucial species for northern BC and seed production levels need to increase through ramet development and increased future production levels. VSOC has begun to replace and increase production trees to reach target level demands set out by the FGC and will continue to develop and pursue silviculture demands through grafting strategies and forward selection parent trees.

To reach these goals:

- 500 ramets rogued in the mature orchard 222
- 1063 ramets were planted in the replacement orchard
- 1600 ramets are being maintained in a holdbed for future planting

#### SPU 1208 Pollination and Pest Management in Prince George Orchard 236 - Pli

Supplemental mass pollination was completed for this maturing orchard to increase the total seeds per cone produced.

Insect control, particularly *Zellaria* and *Sequoia* pitch moth control monitoring and spraying were completed for 4500 trees.

#### SPU 1403 Enhancing Production in Prince George Weevil Tolerant Orchard 211 - White Spruce

Routine pest management and monitoring such as Adelgid control is essential in maintaining and increasing ramet health and vigour.

#### SPU 1706 Pollination and Pest Management for Bulkley Valley Orchard 234 - Pli

Two litres of pollen were collected for future use to improve seed set and orchard gain. Insect control, particularly *Zellaria* and *Sequoia* pitch moth control, monitoring and spraying were completed for 3000 trees.

#### SPU 1801 Enhancing the Effectiveness of Central Plateau Orchard 218 - Pli

*Dioryctria* has begun to infect orchard cones. Timing and spray strategies are now in place to effectively prevent further seed loss. Monitoring for this and other pests is ongoing in routine orchard care.

#### SPU 3702, 3703, 4102, 4103, 4301 Increasing Seed Production in Interior Douglas-fir Orchards 231, 232, 233, 225, 226

Douglas-fir orchard seed production has started to flourish in the interior. Pollen strategies, pest monitoring and inducing crops through GA<sub>4/7</sub> are part of the success. Pest control and good orchard management strategies play an important role in success.

Main activities include:

- Pollen collection
- *Dioryctria* and Adelgids control
- Cone induction using giberellic acid
- Weekly pest monitoring

#### SPU 4202 Pest Management in Prince George High Elevation Orchard 239 - Sx

Spider mites, adelgids and other pests can seriously affect crop potential. Funding allows orchards to control potential losses efficiently through sprays and monitoring.

## 6.1.6 PRT - Armstrong Seed Orchards

Mike Brown

### Projects 0702, 0721, 1001, 1002A, 1007, 2101, 0728

With the awarding of funding from the Operational Tree Improvement Program, PRT has been able to effectively maintain the supply of class A seed to the BC forest industry.

Activities carried out during the 2013 season include:

- Removal of Orchard 308
- Pollen collection
- Supplemental mass pollination (SMP) for young Fdi ramets which produce insufficient pollen
- Use of GA<sub>4/7</sub> hormone to promote flowering in Douglas-fir
- Introduction of girdling to promote flowering in Douglas-fir
- Improving the overall genetic value of individual orchards by selectively removing lower breeding value ramets and replacing them with higher breeding value ramets
- Regular thorough pest monitoring to ensure all ramets remain safe from disease and insects
- Removal of Western Gall Rust from infected Pli trees
- Crown management to maintain height and width and encourage promotion of lateral branching thereby increasing cone sites

PRT Armstrong has 4 lodgepole pine orchards and one Douglas-fir orchard. These orchards collectively contain approximately 10,000 ramets which produce seed for the Thompson Okanagan low elevation (TO Low) and the Nelson Low elevation (NE low). Three of the orchards (337, 338 and 321) are jointly owned by PRT and SelectSeed Company Ltd. The other two orchards (311 and 313) are jointly owned by PRT and FLNR.

In the spring of 2013 the decision was made to remove orchard 308. This 25 year old orchard had been in production decline over the past 4 years, was heavily infected with Western Gall Rust and over the winter of 2012/13 sustained heavy winter damage with substantial loss of limbs. An independent contractor was brought in to remove and chip the trees; these chips were then reincorporated into the old orchard site (Plate 44).



Plate 44. Seed Orchard 308 removal at PRT.



Our mature lodgepole pine orchards receive an assist to their pollen distribution through the use of an air blast sprayer. On calm days, the air blast sprayer was driven up and down the orchard rows. With the use of the powerful sprayer fan, pollen would be blown out of the buds to be dispersed to receptive flowers (Plate 45).



Plate 45. Bev Hulley spraying pollen in Seed Orchard 337.

While SMP is taking place, we collect pollen from clones which have an abundant supply. This collection is done with the use of a backpack vacuum, whereby pollen is collected into vacuum bags then dried down, and stored in the freezer for the following years' use in SMP. In the spring of 2013 we collected 2 litres of lodgepole pine pollen for future use in the TO low orchards and 2 litres of lodgepole pine pollen in the NE Low orchards.

In the spring we take soil samples which are sent away to be analyzed for nutrient availability. The results of the analysis allow us to create an effective fertilizer mix for a spring application. This application is timed to be in place prior to bud elongation so the tree can take advantage of the nutrient availability.

Pest monitoring is carried out on a weekly basis. The orchards are monitored for insect, disease and rodent activity. We use insect pheromone traps located on the perimeter of the property, and at strategic locations within the orchards. These traps alert us to insect flight periods during the growing season. Using these tools allows us to streamline our monitoring and apply control sprays in a timely fashion to ensure the trees stay as healthy as possible throughout their growing season.

With the removal of Orchard 308 the level of Western Gall Rust (WGR) inoculum was significantly reduced. With funding from OTIP, we were able to commence a program designed to gain control over the spread of WGR into adjacent orchards. Using electrocoup pruners, Seed Orchard Technicians systematically went through one half of each orchard pruning out all visible galls. The hope being that by addressing these galls now, we will be able to slow down the spread of WGR through the remaining lodgepole pine orchards.

As part of a collaborative project with other orchards in the Okanagan, PRT applied an "Attract and Kill" formulation to its Pli orchard trees in a study to attempt to eradicate Pitch Moth. This black tar like substance is placed on trees branches and contains the pitch moth pheromone as well as insecticide. The male, being attracted to the substance, is then killed. Pheromone traps were placed in the orchard to detect the presence of the insect pest in both the control and test block. The experiment will hopefully show a reduction in the population of pitch moth which could lead to less larval damage to the trees.

Crown management for height control and lateral branch control was carried out on orchard 338. The objective was to bring the overall tree height down to a manageable level and facilitate ease of movement through

the orchard with lateral pruning. This pruning will promote lateral branching on the remaining portions of the tree and increase cone sights in future years.

Monitoring for Red Turpentine Beetle (RTB) continued in 2013. Pheromone traps set out around the site showed a flight in early May. There was a notable decrease in the number of trees being killed through insect attacks which clearly shows the positive effect the past preventative sprays have had. Over the course of the season, the trees were monitored 2 additional times to ensure no fresh attacks were found. Dead trees were regularly removed to be certain any eggs which may have been laid were destroyed.

*Sequoia* Pitch moth larva were once again removed manually from the lodgepole pine orchards with a concerted effort to address all of the Pli ramets twice during the season.

### Fdi Orchard 321

SMP in the fir orchard this year targeted the early and late clones. Due to the random sparse nature of the pollinating trees, younger trees were also a focus for our SMP program. Two litres of pollen were collected for use with SMP in subsequent years.

The Douglas-fir orchard was monitored weekly for insect, disease and rodent activity. With the fir, particular attention was paid to the presence of *Dioryctria abietivorella* whose larval stage feeds on Douglas-fir cones and will do large amounts of damage to the seed crop if left unchecked.

We applied two well timed insect sprays for *Dioryctria* which kept the 2012 crop clean from insect damage.

Once again this year we applied GA4/7, a natural tree hormone used to promote cone production. The GA 4/7 is used in conjunction with a stress signal to encourage the tree into flower production instead of being purely vegetative. In the past, we have relied on drought as a stress factor however the weather over the past few years has seen wetter than average springs. This year we decided to implement girdling as a stress factor, using saw or knife cuts around the circumference of the trunk at breast height to send a stress signal to the tree. The hope is that the girdling is a much more defined stress for the tree to respond to. Assessments in the spring of 2014 will let us know if this new method had any effect.

The cone harvest for 2013 had a yield of 49 kg of lodgepole pine seed while the Douglas-fir crop produced 30.9 kg of seed. OTIP funding has continued to help us produce increasing amounts of A-class seed thereby supporting the FGC's goal of making larger volumes of genetically improved seed available for use in the forest industry.



Plate 46. GA in Douglas-fir. A saw cut being performed by Laura Whitney.



Plate 47. GA in Douglas-fir. A close up of the saw cut.



Plate 48. GA in Douglas-fir. Jane McLean and Laura Whitney (with drill) applying GA and wrapping trunk with vet wrap to protect against *Dioryctria*.



## 6.1.7 Eagle Rock Seed Orchards (Tolko Industries)

### Rod Massey

Four orchards are managed by Tolko Industries for the Thompson Okanagan seed planning zone. Three orchards, two interior spruce 342 and 343, and one lodgepole pine 339 (Plate 49) are SelectSeed Ltd. partnership orchards. The fourth orchard, lodgepole pine 310 is managed fully by Tolko Industries. The three projects funded by the Operational Tree Improvement Program aid in improving the quality and quantity of seed produced for the Thompson Okanagan forest community. In 2013, Eagle Rock produced seed for 1.1 million lodgepole seedlings for reforestation in the Thompson Okanagan seed planning zone.

### SPU 16 Thompson Okanagan Pli High, Orchards 310 (Tolko) and 339 (SelectSeed), Project 1601

- When pollen and flower surveys indicated optimal receptivity, supplemental mass pollination was completed with the aid of an air blast sprayer and helicopter.
- Pocket gophers were controlled by administering bait in the spring.
- Pest monitoring was completed for *Eucosma*, *Dioryctria auranticella*, *Rhyacionia buoliana* and *Lepidoglossus occidentalis*. Although all pest populations had increased from 2012, no control was required.

- *Synanthedon sequoiae* and *Dioryctria* spp. were manually removed from the base of young ramets to prevent girdling and full or partial ramet loss.
- Grafts (900) of high breeding value and performing clones from orchard 339 for the orchard 310 replacement were maintained at Skimikin for planting in 2014.
- Weekly cone collection was completed for OTIP0722 (see page 51). Information and samples were sent to Kalamalka Seed Orchard for processing.

### SPU 28 and 30 Thompson Okanagan Sx Low, Orchard 342 and Sx High, Orchard 343 (SelectSeed)

- There was no spruce crop in 2013, therefore no SMP or pollen collection was required.
- Monitoring for pests such as *Adelgids*, *Pissodes strobi*, and *Oligonychus ununguis*, *Dioryctria abietivorella*, *Choristoneura occidentalis* was completed.
  - Leaders containing *Pissodes* spp. were removed in June to decrease the population within the orchard.
  - To increase health in small ramets, *Oligonychus ununguis* was controlled in March in both orchards.
- Pocket gophers were controlled by administering bait in the spring.



Plate 49. Orchard 339 provides seed for the Thompson Okanagan higher elevation seed planning unit. This orchard is operated by Tolko Industries under agreement with SelectSeed Ltd. (J. Woods image).

## 6.1.8 Prince George Tree Improvement Station (PGTIS)

Rita Wagner

SPU 1203, 1802, 1702

Activities are aimed at increasing the quantity and quality of lodgepole pine seed from Orchard 220 (Prince George low planning zone), Orchard 223 (Central Plateau low planning zone) and Orchard 228 (Bulkley Valley low planning zone).

Three Operational Tree Improvement Projects were conducted at the Prince George Tree Improvement Station in 2013-2014.

Surveys for western gall rust, *Elytroderma* needle cast, *Lophodermella* pine needle cast, *Zelleria* pine needle-sheath miner, *Cecidomyia* pitch midge, and various other insects were completed.

Lindgren traps were set up throughout the site to

monitor secondary bark beetle flights (mainly *Ips*). Since 2009, mountain pine beetle activity in the Prince George area continues to drop.

Since 2005 all three orchards had three years of very high production, two years of medium high production and two years of medium-low production. However, even the medium-low production still exceeded the FGC target forecast for each orchard. In 2013, the three provenance orchards yielded 18.7 kg of seed, the equivalent of approx. 3.8 million potential seedlings with a genetic worth of 6%.

Wet spring/summer weather in 2011/12 caused an explosion of fungal infection on shoots/flowers resulting in 35-40% seed loss in our 2013 crop. The fungus attacked flowers resulting in abortion as well as partial damage to flowers. These flowers developed normally, but quite a few ovules were damaged. This reduced our seed yield/HI by about 15-20%.

Squirrel trapping was carried out to prevent seed loss and loss of potential cone sites.



Plate 50. Bulkley # 228 with double rainbow.

Plate 51. A visitor to the holding area.



## 6.1.9 Skimikin Seed Orchards

Hilary Graham

### Summary for Projects 0404, 3502, and 4002.

Skimikin Seed orchards are comprised of 13 orchards covering 9 SPU's and 4 conifer species – interior spruce (Sx), western white pine (Pw), lodgepole pine (Pli), and Ponderosa pine (Py). There are also research plantations covering a wide variety of species and projects. Seven Sx orchards produce seed for the Bulkley Valley, Peace River, and Nelson seed planning units. One Pw orchard produces rust resistant seed for the Kootenay Quesnel SPU and a 2<sup>nd</sup> Pw orchard for the same SPU is nearing production. The three young Pli orchards will produce seed for the Thompson Okanagan low, Nelson high, and Prince George low SPUs.

In 2013/14, 3 projects received OTIP funding for activities to increase the yield and genetic gain of seed produced in the spruce orchards. These activities included holding grafts, planting grafts, cone induction, roguing, insect and disease monitoring and control, and rodent control.

### Interior spruce – orchards 301 (0404), 207/208/229 (3502), and 212/213 (4002)

In the Sx West Kootenay low elevation orchard 301 (project 0404), the orchard was monitored for pest populations and treatments were applied as necessary. A spray for spruce budworm was not required. Damage due to rodent feeding was minimized by the application of rodenticide in affected areas.

For the Bulkley Valley low orchards 207, 208, and 229 (project 3502), 364 high breeding value grafts were maintained in holding beds for future planting. All orchards were monitored for pest populations and treatments were applied as necessary.

Spider mite populations were controlled by a miticide spray in June and leaders affected with the spruce leader weevil were removed by hand. A spray for spruce budworm was not required and the dry spring conditions made a spray for cone rust unnecessary. Damage due to rodent feeding was minimized by the application of rodenticide in affected areas.

To increase genetic gain, a total of 300 low breeding value ramets were rogued. These orchards produced a combined seedlot (63439) of 36.4 kg of seed with a genetic gain of 23%.

In the spruce orchards for the Peace River low and mid elevation zones (orchards 212 & 213 – project 4002) 52 grafts were planted in the spring of 2013. Three hundred and thirty-three ramets were induced for cone production with Giberellic acid (GA) to induce a crop for 2014.

Both orchards were monitored for pest populations and treatments were applied as necessary. Leaders attacked by the spruce leader weevil were removed by hand and damage due to rodent feeding was minimized by the application of rodenticide in affected areas. Sprays were not required for spider mites or spruce budworm in these orchards. Also, the dry spring conditions made a spray for cone rust unnecessary.

Orchard 212 produced 7.23 kg of seed with a genetic gain of 23%, and orchard 213 produced 1.76 kg of seed with a genetic gain of 4%.





Plate 52. Skimikin crew.



Plate 53. Cone harvest 2013.



Plate 54. Spruce cones 2013.



### 6.1.10 Kettle River Seed Orchard Company (KRSO)

Rick Hansinger

#### Pollination and Pest Management in Central Plateau (CP) Orchard 237 – Lodgepole Pine (SPU 1210)

##### Objectives

Collect and store 3.0 litres of pollen for SMP in young Pli Orchard 237 to increase the production of Class A seed to 305,000 potential trees for sowing year 2013/2014.

Minimize filled seed losses from predation by *Leptoglossus* through pesticide applications.

##### Results

Three litres of pollen was vacuum collected in the Pli PG low orchard, the pollen was cleaned, dried and stored for application in spring (2014 Genetic Worth G+17 Class A). Approximately 3 litres of pollen was applied to 4500 ramets during the early/late receptivity period from May 20 to June 10. The Pli PG orchard is now producing sufficient pollen to meet SMP and open pollination requirements. Two SMP passes were completed with compressed air in order to ensure early and late receptive clones received sufficient pollen to fertilize female conelets. Pollen was air blasted with a turbo fan sprayer on calm days to encourage pollen flow over receptive conelets. Three litres of pollen was stored for SMP of future crops.

Developing cones were inspected for the presence of *Leptoglossus* and the risk to the seed crop was deemed negligible, pesticides were not applied.

##### Output and Deliverables

SMP and open pollination combined have resulted in a yield of 241 seeds per gram and approximately 137 seedlings per gram at 98% germination. Seedling yield for harvest year 2013 is approximately 305,000.

#### Pollination and Pest Management in Central Plateau Orchard 238 – Lodgepole Pine

##### Objectives

Collect and store 3.0 litres of pollen for SMP in young Pli Orchard 238 to increase the production of Class A seed to 572,000 potential trees for sowing year 2013.

Minimize filled seed losses from predation by *Leptoglossus* through pesticide applications.

##### Results

Three litres of pollen was vacuum collected in the Pli CP low orchard, the pollen was cleaned, dried and stored for application in spring 2013 (Genetic Worth G+20 Class A). Approximately 4.5 litres of pollen was applied to 3,000 ramets during the receptivity period from May 20 to June 10. The Central Plateau Pli orchard is now producing sufficient pollen to meet SMP and open pollination needs. Two SMP applications were completed in order to ensure early and late receptive clones received sufficient pollen to fertilize female conelets. Remaining pollen was air blasted with the turbo fan sprayer. Six litres of pollen remains in storage for spring 2014 SMP.

Developing cones were inspected for the presence of *Leptoglossus* and the risk to the seed crop was deemed negligible, pesticides were not applied.

##### Output and Deliverables

SMP and open pollination combined have resulted in a yield of 230 seeds per gram and approximately 131 seedlings per gram at 98% germination. Seedling yield for harvest year 2013 is approximately 572,500.

## 6.1.11 Sorrento Seed Orchards

### Harry Hamilton and Jack Woods

Sorrento Nurseries Ltd. manages two lodgepole pine orchards in partnership with SelectSeed Company Ltd. These orchards total about 5000 ramets and supply seed for the Bulkley Valley (BV) and Central Plateau (CP) low-elevation seed planning units from orchards 240 and 241, respectively.

Projects were carried out under OTIP contracts 1707 for orchard 240 (BV) and 1803 for orchard 241 (CP). Activities were similar for both orchards and are described together in this report.

### Supplemental pollination

Approximately three liters of pollen was collected using a backpack vacuum in each orchard and distributed to emerging receptive flowers. In addition, the air-blast sprayer was used to enhance the distribution of pollen when conditions were suitable (dry afternoons during pollen shed).

### Pest control

Pitch-moth larva (*Synanthedon sequoia*) were manually removed from ramets in both orchards. No other pests were considered to be a threat to tree health or to the crop.

### Non-OTIP projects

Ongoing management in the orchards included irrigation, fertilization, crown pruning on larger ramets, tag maintenance, mowing, and the replacement of dead ramets. Crop harvest was undertaken in early August, yielding 65.8 and 54.7 hl of cones for the BV and CP orchards, respectively. Seed production for these orchards was 5.6 and 6.2 kg.



Plate 55. Sorrento orchard 240 producing seed for the Bulkley Valley.

## 6.2 Technical Support Programs

### 6.2.1 Increasing Quality, Genetic Gain, and Quantity of Yellow Cypress Cuttings (SPU 1113)

Mark Griffin, John Ogg, Craig Ferguson and John Russell

#### Introduction

This project involves increasing the quantity and quality of high-value yellow cedar cuttings for the coastal program.

Objectives include:

1. provide the cultural treatments required to improve hedge production, and
2. enhance hedge composition by replacing lower-genetic-value families and clones with newly tested, improved clones.

#### 2013/2014 Highlights

Pruning of hedges occurred in April 2013. This year the pots were top dressed with Nutricote 18-6-8 type 180, augmented with periodic applications of hi-sol. For pest control, predaceous nematodes were applied regularly to control root weevils.

During 2013, a large amount of roguing was done and the number of contributing ramets in the hedge has been reduced to 71 clones with 5755 ramets. The roguing was based on the results of rootability studies where samplings of every clone were previously set and each clone's propensity to root was assessed. Poor rooting clones have therefore been culled from the operational hedge.

With the removal of the poorly rooting clones, it is now estimated that the hedge is made up of stock where at least 60% of the cuttings supplied to the nurseries will yield a plantable "seedling" at the end of 10 months of growth in the greenhouse. We expect the 5755 ramets currently in the hedge will be able to supply an estimated 152,000 cuttings.

To replace the rogued material and to reinvigorate the hedge, material from series 3 selections have been set and additional clones are being added based on field performance data. These additions are currently of a size where they are to be transferred to styroblock 615As.

In late 2013, some 151,500 cuttings from this operational hedge were supplied for production to reforestation nurseries.

## 6.2.2 Estimating Pollen Contamination in Coastal Douglas-fir Seed Orchards SPU0113

### Final Report

Joe Webber

The 2013 contamination levels in four coastal Douglas-fir orchards were as follows: 66.4% at Western Forest Products (WFP-166); 18.8% at TimberWest (TW-183), 100% at FLNR (181) and 54.4% at FLNR (199). These values were estimated using the mean pollen load (PL) from three regional monitors and two orchard monitors and calculated as  $\%Contamination = (Regional\ PL / Orchard\ PL) * 100$ . These estimates do not include the orchard adjustment factor (OAF). Applying the OAF substantially reduced the contamination estimate at WFP, increased the contamination estimate at TW and had little effect on the two ministry orchards.

Contamination values at WFP have been estimated for the period 2005 to 2013 (Table 9) and regional pollen load (grains per millimetre squared per day summed over the orchard receptivity period) ranged from a low of 4.6 (2005) to a high of 39.1 (2009). The range of orchard pollen load for the same period was a high of 114.6 (2007) to a low of 11.6 (2013). For the last six years, the mean regional and orchard pollen loads were 13.2 and 41.5, respectively. The mean contamination value for this period was 38.4%.

During the last four years, the mean orchard pollen loads at WFP have dropped substantially from about 85 (2009) to 29 (2010 to 2013). This is the principal reason contamination values at WFP have remained high over the last four years.

Contamination values at TW have been estimated for the period of 2008 to 2013 (Table 9). Since receptivity periods from TW were similar to WFP, the range of regional pollen loads were also similar (3.2 in 2008 to 37.6 in 2009). The range of orchard pollen loads during the same period was 31.3 in 2013 to 80.3 in 2009. Over this six year period the mean regional pollen load was

similar to WFP (13.2) and orchard pollen loads were 48.0. The mean contamination value at TW for the last six years was 24.1%. Orchard pollen loads at TW have also dropped in the last four years.

In general, orchard and regional pollen loads trends follow each other suggesting that pollen production in both orchard and non-orchard trees is the result of the same environmental induction conditions. For the last four years, regional pollen loads have been relatively low (7-15) but orchard pollen loads have also been low leading to generally high contamination levels. In general, the lower contamination levels at TW was largely attributed to higher orchard pollen loads arising from substantially more mature, pollen producing trees (about 3000 at TW and 400 at WFP).

The spring receptivity period for the last four years (2010-2013) was slow enough to allow the Orchard Adjustment Factor (OAF) to be calculated. For all four years, adjusting the regional pollen data with the OAF using only early pollen capture data decreased the regional pollen load by an average factor of 0.28 and 0.64 at WFP and TW, respectively. The four year mean for contamination without the OAF was 42.4% and 21.7% for WFP and TW, respectively. Applying the OAF factor to adjust the regional pollen load decreased the four year (2010 to 2013) mean contamination values to 8.8% and 18.0% for WFP and TW, respectively.

The orchard adjustment factor is very sensitive to a few pollen grains capture by either the orchard or regional monitors. While the OAF has reduced regional pollen load values for the last four years at WFP and three of the last four years at TW, the question becomes, do we accept those years where the OAF increase regional load (and therefore contamination). Since calculation of the OAF varies substantially by dates within and between seasons, it must be used carefully. While the simpler ratio of regional to orchard pollen loads is recommended, the error of estimating contamination will increase with increasing pollen loads. The extent of this error will be determined by the more robust DNA technique being applied to those seed crops where high contamination values have been estimated.



	Douglas-fir Orchard Receptivity 7-day Monitor Pollen Load (grains/mm <sup>2</sup> /day) and %Contamination															
	WFP										TW					
	2005	2006	2007	2008	2009	2010	2011	2012	2013		2008	2009	2010	2011	2012	2013
REG PL	4.6	6.7	24.6	7.3	39.1	12.7	7.3	14.8	7.7		3.2	37.6	12.7	8.0	12.0	5.9
ORCH PL	96.3	54.6	114.6	48.9	85.4	27.5	29.0	46.7	11.6		28.6	80.3	63.7	43.2	40.6	31.3
%Cont																
PM	4.8	12.5	21.5	14.9	45.8	46.1	25.2	31.7	66.4		11.2	46.8	19.9	18.5	29.6	18.8
DNA																
(MS)	9.7	11.7	19.3	na	na	na	na	na			na	na	na	na	na	
DNA																
(ELK)	10.5		na	na	(15)	no crop	na	na								

Table 9. Regional (REG) and Orchard (ORCH) pollen load (PL) values (2005-2013) and estimates of contamination for WFP-166 and TW-183 Douglas-fir orchards using pollen monitoring (PM) and DNA paternity analyses.

### 6.2.3 Interior Lodgepole Pine Orchard Seed Set SPU 0722 Summary of Field Trials 2000-2012

#### Joe Webber

We have been aware of the low seed set problem in north Okanagan lodgepole pine orchards for over 15 years but numerous attempts to determine the underlying cause and improve seed yields have eluded us. This report summarizes many studies done over the past 15 years and relates their findings to the problem of poor seed set in lodgepole pine.

The only consistent effect is insect protected cones produce more seed. However, assuming that the difference in yields between protected and unprotected cones is attributed to insects alone, we still do not equal seed yields consistently obtained in Prince George. On average, we should see 32 total seed per cone (empty + filled) and 24 filled seed per cone. We see these numbers at Prince George but we do not see them in the north Okanagan. Some of the losses can be attributed to insects but not all.

The most cited cause of this short fall is the hot, dry weather of the north Okanagan leads to reproductive failure but attempts to improve irrigation and cool crowns had no affect on yields. More recent work shows fungal activity is

associated with dying embryos during the later stages of seed development but whether this is a cause or side effect is not clear. In fact most of the recent work has focused on the late July/August collapse of seed yields which could be attributed to insect predation since bagged cones have better seed yields.

We have not looked carefully at the post-dormant pre-fertilization period of rapid cone expansion of the second-year cones, especially regarding loss of fertile ovules. We assume from bagging experiments that insect bags are put on before insect predation begins but if not; insect damage would reduce the number of total seed per cone. If insects do not cause the loss of potential seed (total seed per cone), then what does? It seems reasonable to consider a systematic sampling of cones from early April to mid June when fertilization and early embryo development occurs.

A principal challenge for north Okanagan orchards will be managing insect populations. All orchards show signs of declining seed yields as they age suggesting tree vigour is declining. For older orchards, improved seed yields may be available if management activities focus on smaller crowns, fewer trees, and improving soils. If these activities can be successfully applied to north Okanagan orchards, then improved seed production is possible.

The report details results for pollination biology, cultural effects, bagging effects, seed biology, crown management and cone production.

## 6.2.4 Pilot-scale operational application of yellow cypress seed production across climatically contrasting sites using methods developed under previous OTIP projects and existing clonal trials. SPU1118

### Oldrich Hak

Yellow cypress (*Callitropsis nootkatensis* (D. Don) Oersted) grows naturally in cold and humid climates in the Pacific Northwest, occurring at high elevations in the southern areas of its range and from low to high elevations in the central to northern extent of its range. It is the first of its associates to free itself of snow and to undergo pollination when heavy snow load on the ground and severe temperature fluctuations are common early in the spring.

Yellow cypress seed production in natural stands can be minimal and sporadic; consequently, seed is in short supply for reforestation. Seed orchards were established at low elevations with the idea that the influence of warmer climate will promote earlier and increased quantity of cones and higher quantity and quality of filled seed. To the contrary, seed orchards produced lower quantities of viable seed than wild stands. Climatic differences between high elevation natural stands and low elevation seed orchards could be one of the main causes for reduced viable seed. Results from previous yellow cypress studies under OTIP indicate that site location has a considerable influence on viable pollen and seed production. Study results point out that the lack of filled seed at low elevation sites, most likely because of both poor quality pollen and poor female flower and embryo development, may be a consequence of prevailing climate. For example, significant differences in pollen viability were observed between populations characterized by distinct climatic conditions.

The objective of this project is to determine the effect of climatic site differences on yellow cypress operational seed production by applying methods developed under previous OTIP projects. The project was initiated at four climatically variable field sites during 2008/09. These sites were selected from existing Western Forest Products' clonal trials in the field near Port McNeill and Jordan River. There are two replications per site. Each replication was thinned to approximately 100 trees to mimic actual seed orchard conditions and enable viable seed production.

One replication per test site was induced in May 2010 with GA<sub>3</sub> according to protocols developed by an earlier OTIP project. Second replication per test site was induced in May 2011. This involved two-time spray applications in two week intervals using mechanical sprayers. All 100 trees in the thinned replications were induced to ensure an adequate pollen cloud during pollination. Pollen was collected for viability testing from ten trees at each of four sites during the following spring after each induction. Pollen viability at three sites at Port McNeill and at Jordan River high was high: 80%, 89%, 84% and 76% respectively in 2011 (Rep. 1), and 75%, 87%, 82% and 78% in 2012 (Rep. 2).

Assessments of cone production for each test site were done during the summers of 2012 (Rep. 1) and 2013 (Rep. 2). Average number of cones per tree for each test site was calculated. All three sites at Port McNeill produced higher average number of cones per tree than the Jordan River site in 2012: 1,845 cones, 1,154 cones, 2,930 cones, and 697 cones respectively, and 236 cones, 1,673 cones, 1,651 cones, 1,599 cones, and 570 cones respectively in 2013. Cones from 10 randomly selected trees at each site were collected during the fall 2012 (Rep. 1) and 2013 (Rep. 2). Seed was extracted from 100 cones for each test site, total seed counted and the number of seed (full and empty) per cone calculated. Extracted seed was germinated from December 2012 until March 2013 (Rep. 1), and from December 2013 until March 2014 (Rep. 2). Germination testing was based on 4 replicates of 100 seeds per test site. Results for cone and seed production by test site for 2012 and 2013 are documented in Table 10 and Table 11.

Final report with statistical analyses, discussion, and recommendations will be completed in September 2014.

	Average	Average	Germination	# Germinated	# Germinated
Site	# cones/tree	# seed/100 cones	Capacity %	Seed / cone	Seed / Tree
N.I. 5	1,845	1,045	72	8	14,760
N.I. 6	1,154	904	64	6	6,924
N.I. 8	2,930	1,042	76	8	23,440
J. R. High	697	1,077	68	7	4,879

Table 10. Test Site Productivity 2012 (based on % germination capacity).

	Average	Average	Average	Germination	# Germinated	# Germinated
Site	# cones/tree	# seed/100 cones	# seed/cone	Capacity %	Seed/cone	Seed/Tree
N.I. 5	236	749	8	47	4	944
N.I. 6	1,673	825	8	66	5	8,365
N.I. 8	1,651	867	9	73	7	11,557
J. R. High	570	819	8	60	5	2,850

Table 11. Test Site Productivity 2013 (based on % germination capacity).



## 6.2.5 Kalamalka Seed Orchards Develop Harvest Timing Profiles for Major Clones in Pli orchards 307 and 340

### Gary Giampa

In 2013/2014, Kalamalka Seed Orchards received OTIP approval to develop harvest timing profiles for 47 clones in Bailey EK Pli orchard 340 (SPU3202) and 23 clones in Reservoir Ne Pli orchard 307 (SPU0729).

### Background

North Okanagan Pli orchards have consistently struggled to meet the expected annual average seedling production per ramet figures predicted in the Forest Genetics Council Business Plan. Results from earlier OTIP funded Pli harvest timing trials indicate that there is a point in time when the number of filled seed in each cone declines dramatically (we refer to this as the “crash” in seed set). This event can be tied to growing degree days and is different for each clone. If we can capture (harvest) the seed from each clone before its “crash” occurs we can increase overall seed delivery from our orchards by an estimated 25% to 30%.

### Objectives

In order to maximize seed yield we have to know when seed loss occurs for each major producing clone in each orchard. Developing these profiles will allow us to organize our harvest to ensure that the cones from each clone are collected and in storage before seed decline starts to occur.

### Procedure

We already have harvest timing profiles for ten clones in orchard 307 (this information was developed using observations from previous OTIP funded Pli harvest timing trials run in this orchard in 2011 and 2012). We wanted to develop harvest timing profiles for another 23 orchard 307 clones. We had no information for orchard 340 so we needed to develop profiles for 47 clones in this orchard.

The following process was used to generate these profiles:

- We selected three ramets from each clone. These ramets came from different parts of the orchard.
- Ten random cones were collected from each clone on a weekly basis starting July 10. The last collection was August 28. This worked out to

eight collection periods. Our objective was to time our collection periods to bracket the “crash” in seed set.

- The sample cones were bulked by clone and collection period for a total of 184 samples. The seed from each sample was extracted from the cones, x-rayed and the filled and empty seed were counted and recorded. These data were used to create a graph showing each clone’s seed decline profile. Please see figure 6 below as an example of the type of profiles generated. Note how % seed set for clone 4023 is fairly good until it suddenly declines at about 1334 growing degree days. For this particular clone we would want to make sure that it is harvested and stored before we reach 1334 GDDs.
- Similar graphs were created for each major producing clone in orchards 340 and 307. We now have permanent records that allow us to plan the “right” time to collect each clone. In the future, as long as we monitor growing degree days, we can refer to these records and plan our harvest schedule to maximize seed set and increase overall seed yields from each orchard.
- Seed decline started in some clones as early as 1126 growing degree days. Other clones would not start to “crash” until 1866 growing degree days. This wide distribution in seed decline periods provides us with the opportunity to move our picking crews through the orchards focusing only on clones that need to be harvested at that time.

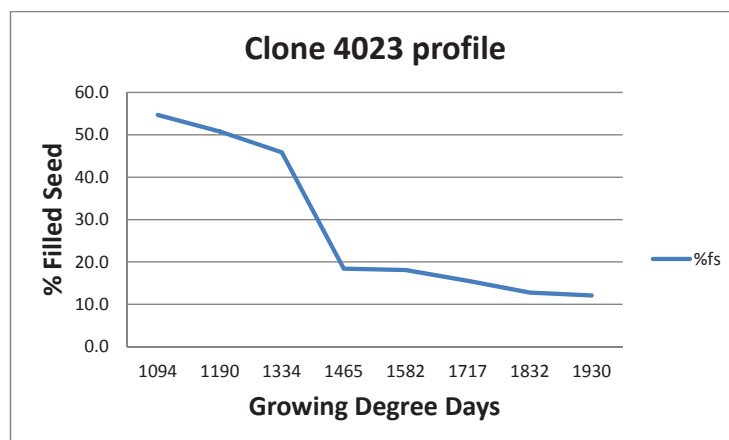


Figure 6. Example of a typical harvest timing profile.

## 6.2.6 Seed Shortfall in Lodgepole Pine and Cone Induction in Lodgepole pine and Douglas-fir

Lisheng Kong and Patrick von Aderkas  
University of Victoria

### Summary

A dramatic seed shortfall occurred in six out of the seven seed orchards under investigation, i.e. O-218 Vernon Seed Orchard Company (VSOC), O-238 Kettle River Seed Orchard (KRSO), O-241 Sorrento Seed Orchard (SORR/SRR), O-307 Kalamalka Seed Orchard (KSO), Pacific Regeneration Technologies (PRT) - Armstrong/Grandview O-313 (GDV) and O-339 Eagle Rock Seed Orchard (EGLR), with the exception of seed orchard O-223 Prince George Tree Improvement Station (PGTIS) at Prince George. The major shortfall had occurred before July 15 with low numbers of filled seed in three orchards, VSOC (O-218), KSO (O-307) and EGLR (O-339).

In other orchards, the percentage of filled seed per cone (FSPC) diminished progressively to <50 % FSPC over the course of investigation. Based on average number of seed per cone, there was no direct correlation between seed shortfall and genotype origin, i.e. from locations in British Columbia characterized by relatively lower or higher temperatures. Histological study revealed that degeneration existed in samples from all of the seven seed orchards. However, in samples from PGTIS, the degree of degeneration was generally low. In particular, type III degeneration commonly found in other orchards was rarely found in samples from PGTIS. In 2013/14, a preliminary investigation of seed borne fungi was completed with molecular tools, using sequence analysis of the internal transcribed spacer (ITS region) of the ribosomal DNA (rDNA) repeat region. As a result, several fungi were found in the infected seeds in lodgepole pine, including *Sydowia polyspora*/Rhizosphaera kalkhoffii, and possibly *Alternaria arborescens*, and *Gibberella avenacea*.

For Douglas-fir genotypes historically characterized by having zero or low female cone yield, stem-injection of a combination of GA<sub>4+7</sub> and ethephon showed the best result for female cone induction. In lodgepole pine, among the treatments applied in 2012/13, stem-injection with either GA or GA and cytokinin significantly increased female cone yield. Female cone clusters were induced by one-time stem-injection treatments. Genotype effects were more noticeable with bud treatments, i.e. bud paste or spray, than

with stem-injection. Our results prove that with optimized protocols both stem-injection and bud spray could be industrially practical. With that aim in mind we developed refined protocols and applied them to ramets of both Douglas-fir and lodgepole pine in spring/summer 2013.

Our 2013/14 research program included two parts. The first part was to investigate causes of seed loss in lodgepole pine seed orchards. The second part was targeted improvement of cone yield and, by extension, seed production in lodgepole pine (Pli) and Douglas-fir (Fd) seed orchards.

### Project I. Study on causes of Pli seed loss in BC seed orchards

#### Introduction

A series of trials in recent years by researchers at Kalamalka Research Station revealed a sudden drop in seed production. Numbers of filled seed per cone dropped off dramatically throughout the month of August. Losses during conifer seed development are generally associated with insect predation, ovule abortion and selfing, as well as failure to pollinate or to fertilize. Previous studies of lodgepole pine had ruled out limitations due to pollination and fertilization in this orchard. Although losses due to selfing could occur at all stages of embryogenesis as well as during early seedling growth, losses during embryo development are most prevalent during globular stage in pines (Koski 1971, Williams et al. 2008). In a normal year, this stage occurs in late June for lodgepole pine at Kalamalka (Owens et al. 2005). However, the recently observed losses occurred much later in the season, which ruled out inbreeding depression due to selfing. Insect seed predators were also intensively studied. Historically, the most important predator has been *Leptoglossus occidentalis*. Insects may have played a role in predation, since controls that were bagged all season and were inaccessible to insects had significantly higher seed yield than unbagged cones. Unfortunately, studies that monitored *Leptoglossus* in the orchard showed that their populations were very low during the particular period of sudden seed shortfall.

It had been suggested that seed shortfall may have been a consequence of locally elevated temperatures. Seed orchards in the North Okanagan, where summer temperature are high, have spectacular seed losses compared with seed orchards in Prince George, where the average summer temperature is much lower. It was thus supposed that trees originating from low temperature areas may not be adapted to high temperature.

In the previous years (2012/13), we showed that there were three types of seed tissue degeneration during seed shortfall. Type I tissue degeneration began with appearance of tiny intercellular spaces. These spaces increased gradually in size and then the tissue developed large holes. Filamentous structures were frequently observed in intercellular spaces as well as in cells. These filamentous structures were reminiscent of fungal hyphae. Type II tissue degeneration was characterized with cell liquidization, dissolution of cell walls and amorphous coagulation of cell contents. Yellow particulate structures were frequently observed with this type of degeneration. Type III degeneration was progressive loss of cell contents until only cell walls remained. Protein body breakdown was followed by vacuolation and nuclear disintegration. Tissue integrity failed with cells showing signs of cytoplasmic collapse and cell wall rupture.

In 2013, samples were collected from seed orchards in the Okanagan and Prince George. Seed development was closely monitored during the period of seed shortfall. Initial investigation with fungus cultures and molecular identification of PlI seed borne fungi were also carried out in 2013/14.

## Material and methods

Samples were collected weekly from seven seed orchards including O-218 (VSOC), O-223 (PGTIS), O-238 (KRSO), O-241 (SORR), O-307 (KSO), O-313 (GDV) and O-339 (EGLR). Orchard managers and staff at these orchards provided us with 20 cones from ten genotypes (two cones per genotype). The schedule was organized by Dr. Michael Carlson of Kalamalka Research Station. Samples were sent to us weekly from the middle of July to middle of September. All seed were dissected from the cones. We measured total seed, filled seed, and empty seed.

A subsample of megagametophytes and embryos were removed and fixed. We sectioned samples from all orchards at four important time points, e.g. July 15 (week 1), Aug 5 (week 4), Aug 19 (week 6), and September 9 (week 9). An additional set of samples from week 2 were also assessed from two orchards, SORR (O-241) and GDV (O-313). Fixed samples were dehydrated in an ethanol series, then infiltrated in glycol-methacrylate and embedded. Sections (5 µm) were stained with dyes, *i.e.* Amido Black10B, Toluidine Blue O (TBO), or lactophenol blue. In order to check seed conditions, sections were stained mainly with Amido Black10B to show details for tissue degeneration.

Fungal identification was based on sequence analysis of the internal transcribed spacer (ITS region) of the ribosomal DNA (rDNA) repeat region. The ITS region is the most widely sequenced DNA region in fungi and has been recommended as the universal fungal barcode sequence. DNA was extracted from either potentially infected seed tissues or from fungi isolated in pure culture from seed stock. We extracted total DNA, amplified the fungal ITS region using polymerase chain reaction (PCR) and fungal-specific DNA primers, after which we determined the sequence of the amplicons. Basic Local Alignment Search Tool (BLAST) scores were then used to provide measurements of homology between a query sequence and the nearest “hit” in the database or similar sequences.

Isolated fungal cultures from seed stock were cultured from either unsterilized or surface-sterilized seed. Samples were cultured on PDA (potato dextrose agar) plates and/or YPD (or YEPD, Yeast Extract Peptone Dextrose) plates. This part of research was completed in close collaboration with two mycologists at UVic, Jon LeBlanc (MSc) and Dr. Will Hintz.

## Results and discussion

### Dissection of cones and seeds

A dramatic seed shortfall occurred in six out of the seven seed orchards (Figures 7 and 8). The only exception was seed orchard O-223 at Prince George (Figures 9 to 12). Within the normal operational harvest window from August 5 to August 19, the number of filled seed per cone was especially low (Figure 11). During these two weeks, the lowest number of seed per cone (Figure 12) occurred in SORR (O-241) and EGLR (O-339). The onset of decline in seed numbers varied among seed orchards. In three orchards VSOC (O-218), KSO (O-307) and EGLR (O-339)) it would appear that significant shortfall (< 50% of the total seed per cone) had occurred even before July 15, which was the first sampling date (Figure 7). In other orchards, the percentage of filled seed per cone (FSPC) diminished progressively over the sampling period (Figure 8). Comparison of the average number of seeds per cone late in the growing season (Table 12) reveals that there was no direct correlation between seed origins, *i.e.* low or high temperature origin, and seed shortfall. This year, 2013, had a notably advanced seed decline: at KSO (O-307), seed shortfall began much earlier than it occurred in 2012 (not shown).



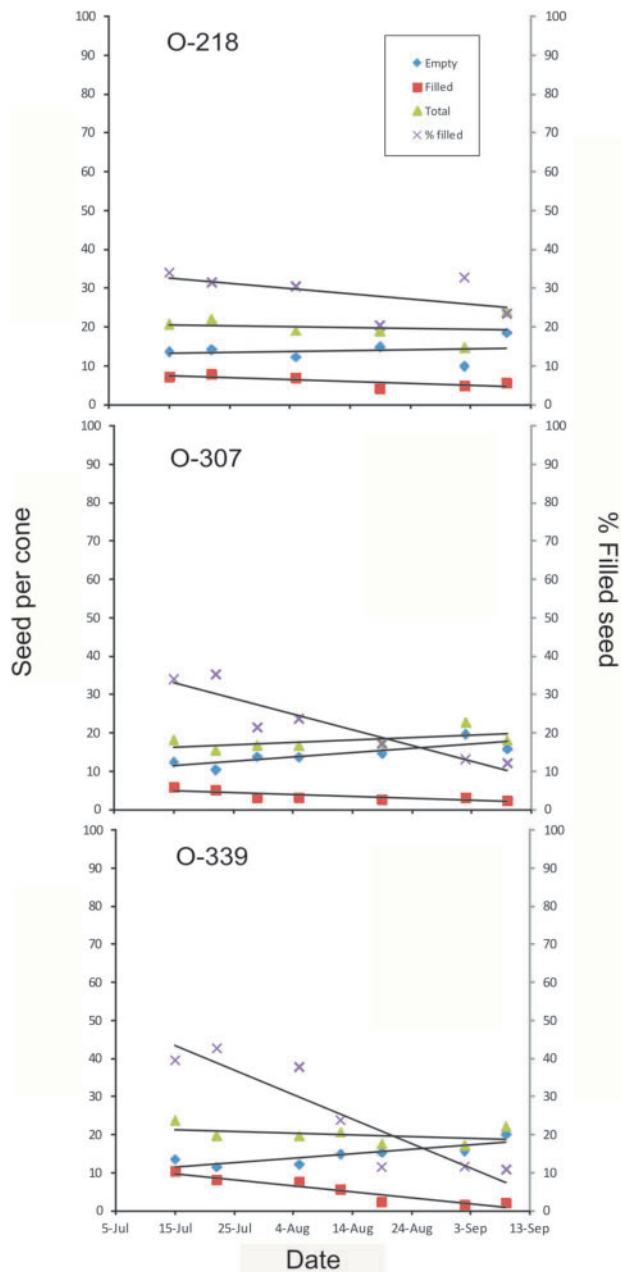


Figure 7. Seeds per cone versus sampling date for VSOC (O-218), KSO (O-307) and EGLR (O-339). N=20.

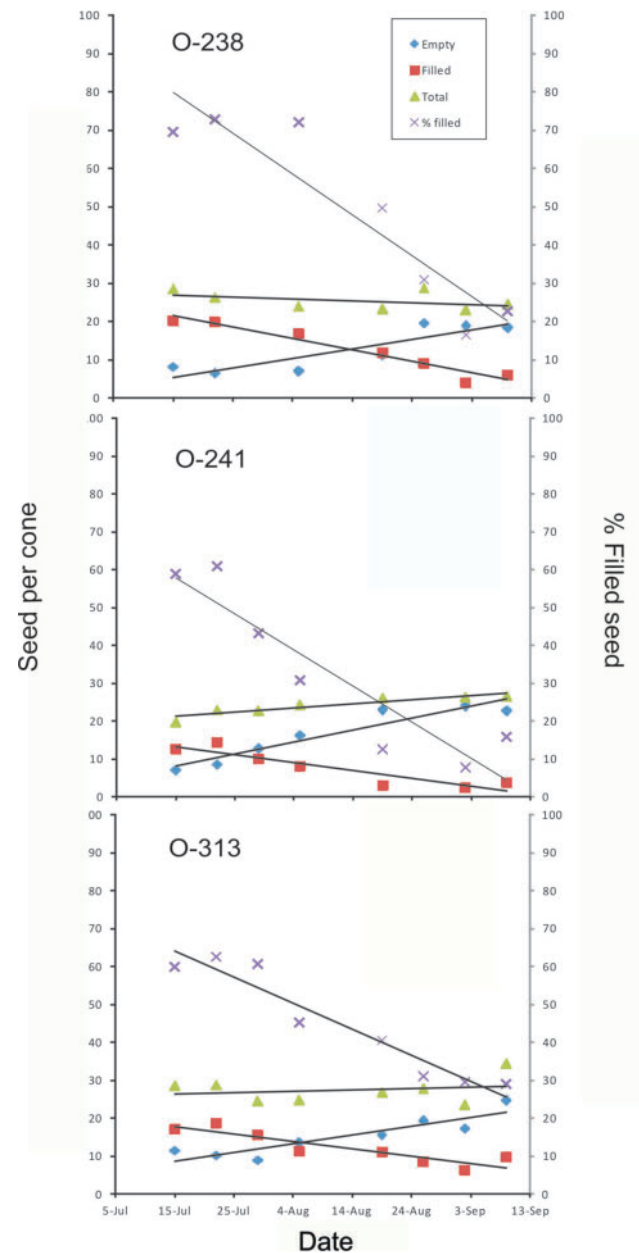


Figure 8. Seeds per cone versus sampling date for KRSO (O-238), SORR (O-241) and GDV (O-313). N=20.

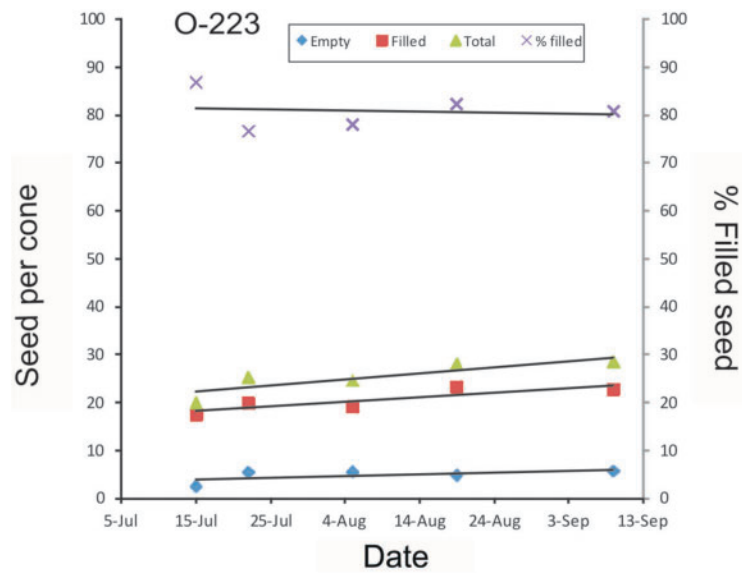


Figure 9. Seeds per cone versus sampling date for PGTIS (O-223). N=20.

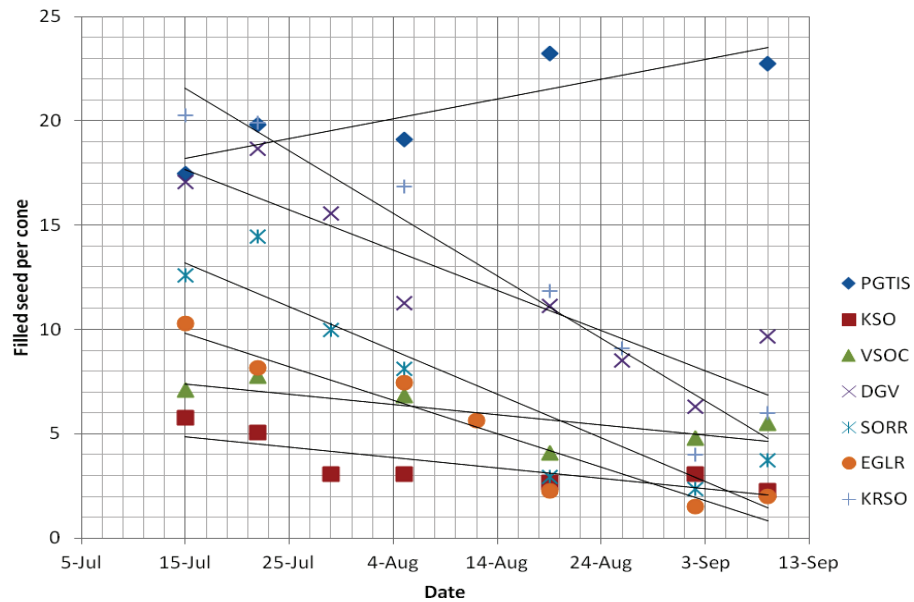


Figure 10. Filled seeds per cone versus sampling date in seven BC seed orchards. N=20.

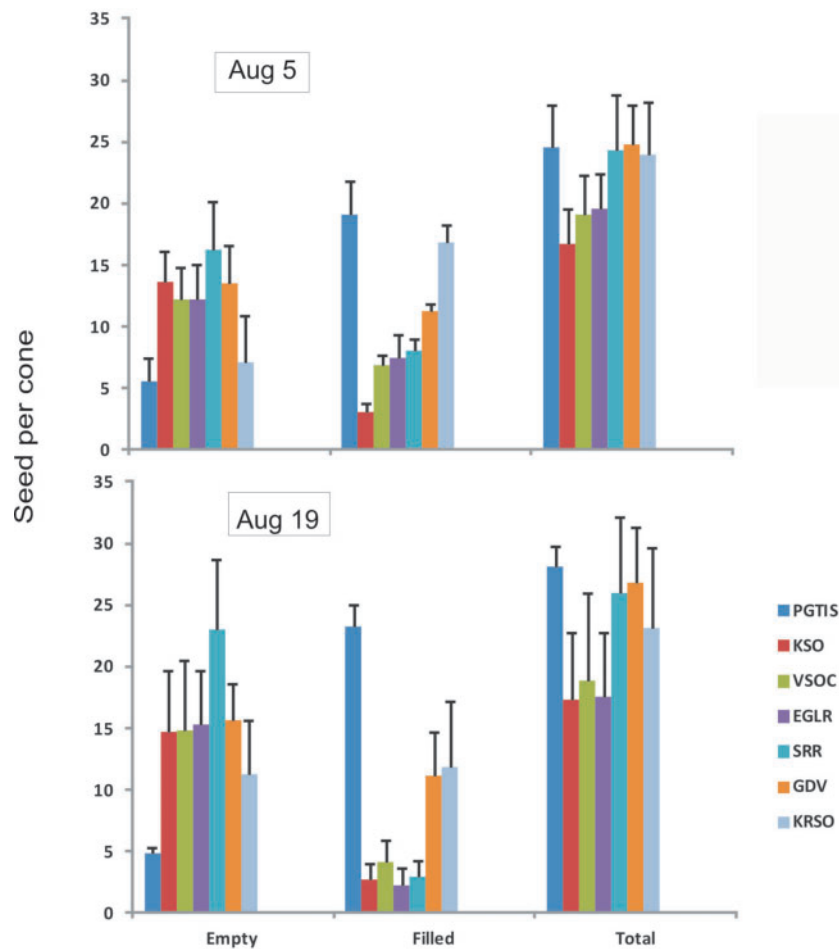


Figure 11. Comparison of seeds per cone in seven BC seed orchards. Samples were collected August 5 and August 19 respectively in 2013. Mean  $\pm$  SE, N=20.

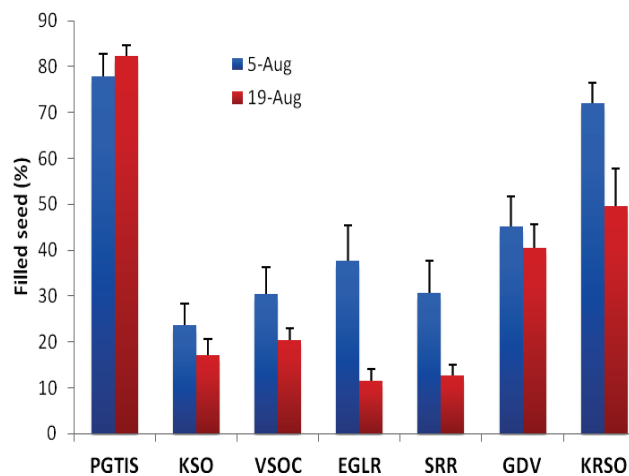


Figure 12. Comparison of percentage filled seed in seven BC seed orchards sampled August 5 and August 19, 2013. Mean  $\pm$  SE, N=20.



<b><u>PGTIS (223)</u></b>		<b><u>KSO (307)</u></b>		<b><u>VSOC (218)</u></b>		<b><u>GDV (313)</u></b>	
<u>Genotype</u>	<u>Seed/cone</u>	<u>Genotype</u>	<u>Seed/cone</u>	<u>Genotype</u>	<u>Seed/cone</u>	<u>Genotype</u>	<u>Seed/cone</u>
C1532	39.5	C1524	7	C2006	10.5	C846	20.0
C1503	31	C1528	5.5	C124	8	C1142	19.0
C1563	29	C1523	4	C1829	7	C1789	11.5
C1474	23.5	C946	2	C1813	6.5	C845	11.0
C1490	22.5	C1526	1	C1833	6	C1783	8.5
C1582	21	C1527	1	C1616	4.5	C1366	8.0
C1473	18.5	C1515	1	C1632	4.5	C1779	7.5
C1471	18	C1536	0.5	C391	3.5	C1202	5.0
C1480	14	C1529	0.5	C2008	3.5	C1450	4.0
C1476	10.5	C1531	0	C190	1	C1689	2.0
<b><u>SORR (241)</u></b>		<b><u>EGLR (339)</u></b>		<b><u>KRSO (238)</u></b>			
<u>Genotype</u>	<u>Seed/cone</u>	<u>Genotype</u>	<u>Seed/cone</u>	<u>Genotype</u>	<u>Seed/cone</u>		
C1821	9.5	C1125	5	C4128	14.5		
C4346	6.5	C1086	4.5	C1616	9.5		
C1831	5.5	C1126	3	C391	8.5		
C2003	4.5	C4350	3	C4421	7		
C4157	3	C4436	2	C1833	6.5		
C4151	2.5	C1124	1	C1821	6.5		
C4421	2	C1127	0.5	C1831	4.5		
C2006	2	C4353	0.5	C4153	3		
C4152	1.5	C4435	0.5	C4144	0		
C2008	0.5	C1083	0	C2008	0		




 - Low temperature origin; 
  - High temperature origin; 
  - Unknown origin.

Table 12. Average seed per cone from various genotypes in seven seed orchards. Genotypes that originate from low or high temperatures are marked in green or yellow, respectively. For each seed orchard, the number of filled seed per cone are ordered from highest to lowest. Samples were collected on September 9, 2013.

## Histology

Our results revealed that intercellular spaces, yellow particles with liquidized tissue, which were types I and II tissue degeneration, as previously described, were found in samples from all of the seven seed orchards. The percentage of seeds displaying each type of tissue degeneration varied between orchards (Tables 13 and 14) and over the growing season. Samples from the PGTIS seed orchard were the most healthy in appearance of all the samples. Even when seeds showed evidence of degeneration, it was less extensive than in samples from all other orchards (Plates 56 and 57, Table 13). Filamentous structures were commonly found in megagametophytes (Table 13) but these were seen much more frequently in samples from the six poor seed orchards. Although the filamentous structures were reminiscent of fungal hyphae, molecular evidence is necessary for confirmation of the hyphae and identification of fungal species. Mottled seed coats and damaged seed coats (Plate 58) were found in many samples. Occasionally, fungal growth within infected seeds could be observed directly with a dissecting microscope (Plate 59A to C).

The major difference in seed tissue degeneration between samples from the PGTIS seed orchard and the other orchards is that the former showed little type III degeneration in seed. Type III tissue degeneration always resulted in death (Plate 57 H) and empty seeds. It was one of the major causes of a sharp drop in filled seed per cone. In addition, the degree of type I degeneration in the samples of Prince George seed orchard was usually lower than that of other orchards: intercellular spaces were smaller in size. Filamentous structures were not as prevalent in the Prince George samples. With only limited testing of seeds for the identification of pathogen(s), such as seed borne

fungi, it is too early to state with certainty that there is a causal relationship between seed shortfall and the presence of pathogenic fungi. To resolve this, a proper survey over the course of the entire disease period would be in order. In our study, some particles that were stained in dark blue color with Amido Black 10B or lactophenol blue were found frequently in intercellular gaps in seed (Plate 57 C) during the season of seed shortfall.

Seed shortfall was so widespread that it does not appear to be linked to the origins of particular tree genotypes. Genotypes that originated from locations that have different summer temperatures, whether higher or lower, all did poorly at the six poor seed orchards. Type I and II tissue degeneration were found in samples from all of the seven seed orchards. Type III degeneration was rarely observed in samples from O-223 (PGTIS), but was frequently seen in samples from other orchards. Occurrence of earlier high temperature in 2013 (Figure 13) and earlier seed shortfall at seed orchard O-307 (at Vernon, BC) was observed when compared with year 2012. The higher temperatures in early summer of 2013 compared with 2012 may have accelerated the process of seed shortfall. Higher temperatures may have contributed to promotion of fungal growth as well as other factors that may be contributing to shortfall.

NOTE: We tested the samples from the design that spanned all seven orchards. We did not sample a related experiment carried out on bagged cones at KSO, as these were not sent to us. Sectioning and histological study of the seeds from bagged cones may generate useful information for exploring causes of seed shortfall, since bagging is known to reduce seed shortfall in Okanagan (Strong 2014). In addition to excluding insects, bagging may also prevent pathogen infection or create a micro-climate better suited to seed maturation in the North Okanagan.

Date	Tissue decline	O-218 (VSOC)	O-223 (PGTIS)	O-238 (KRSO)	O-241 (SORR)	O-307 (KSO)	O-313 (GDV)	O-339 (EGLR)
WK1 (Jul 15)	Type I	54.2%	40%	62.5%	50%	62.5%	66.7%	55.2%
	Type II	33.3%	15%	15%	31.3%	29.2%	29.2%	27.5%
	Type III	0%	2.5%	5%	9.4%	4.2%	4.2%	5.0%
WK4 (Aug 5)	Type I	66.7%	53.1%	75%	90.6%	70.8%	83.3%	83.3%
	Type II	41.7%	25%	50%	37.5%	29.2%	33.3%	33.3%
	Type III	8.3%	0%	8.3%	12.5%	8.3%	8.4%	8.3%
WK6 (Aug 19)	Type I	79.2%	62.5%	80%	68.2%	80%	95.8%	87.5%
	Type II	45.8%	40.6%	56.7%	58.3%	27.5%	45.8%	37.5%
	Type III	12.5%	0%	12.5%	9%	0%	8.4%	4.2%
WK9 (Sept 9)	Type I	79.2%	75%	91.7%	81.3%	75%	91.7%	91.7%
	Type II	45.8%	34.4%	58.3%	28.1%	37.5%	29.2%	45.8%
	Type III	8.3%	0%	4.2%	6.3%	0%	0%	4.2%

Table 13. Tissue degeneration in Pli seed (percentage of the total seed sectioned). Seed cones were collected from seven BC seed orchards in 2013 from July to September.

NB: Type I tissue degeneration began with appearance of intercellular spaces that increased gradually in size until the tissue developed large holes, in which filamentous structures were frequently observed. Type II degeneration had the appearance of cell liquidization. Cell walls were dissolved and cell contents were amorphously coagulated. Yellow particulate structures were frequently observed. Type III degeneration displayed progressive loss of cell contents until only cell walls remained. Protein body breakdown was followed by vacuolation and nuclear disintegration. Tissue integrity failed with cells showing signs of cytoplasmic collapse and cell wall rupture. N= 56 seeds (O-223, O-238, O-241) or 24 to 32 seeds (O-218, O-307, O-313, O-339) at each time point.

Date	O-218 (VSOC)	O-223 (PGTIS)	O-238 (KRSO)	O-241 (SORR)	O-307 (KSO)	O-313 (GDV)	O-339 (EGLR)
WK1 (Jul 15)	41.7%	7.5%	30%	56.3%	45.8%	41.7%	56.3%
WK2 (Jul 22)	50.0%	43.8%	58.3%	71.9%	45.8%	54.2%	70.8%
WK4 (Aug 5)	45.8%	37.5%	40%	55%	57.5%	54.2%	62.5%
WK9 (Sept 9)	45.8%	46.9%	66.7%	71.9%	54.2%	62.5%	83.3%

Table 14. Percentage of filamentous structures in Pli seeds (percentage of the total sample seed). Seed cones were collected from seven BC seed orchards in 2013 from July to September. N= 56 seeds (O-223, O-238, O-241) or 24 to 32 seeds (O-218, O-307, O-313, O-339) at each time point.

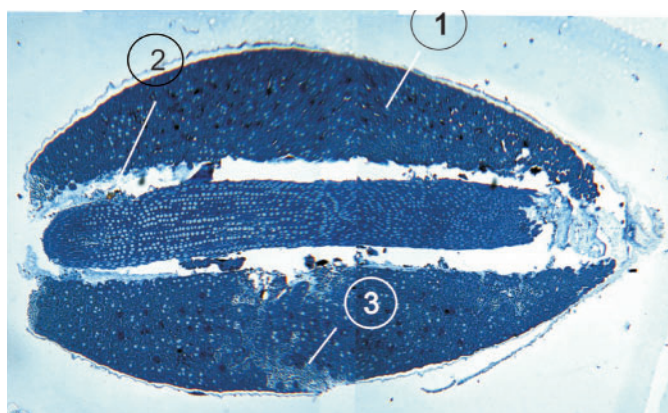


Plate 56. A longitudinal section of a megagametophyte with an embryo in the centre to show locations and types of tissue degeneration. Samples of Pli cones were collected at Prince George seed orchard at Aug 19, 2013. 1) Tiny intercellular spaces were visible in some area of megagametophyte tissue, which was type I tissue degeneration at its early stage; 2) Yellow particle structures; 3) Type II degeneration showing appearance of cell liquidization at a small area of megagametophyte tissue. The embryo was in good condition.

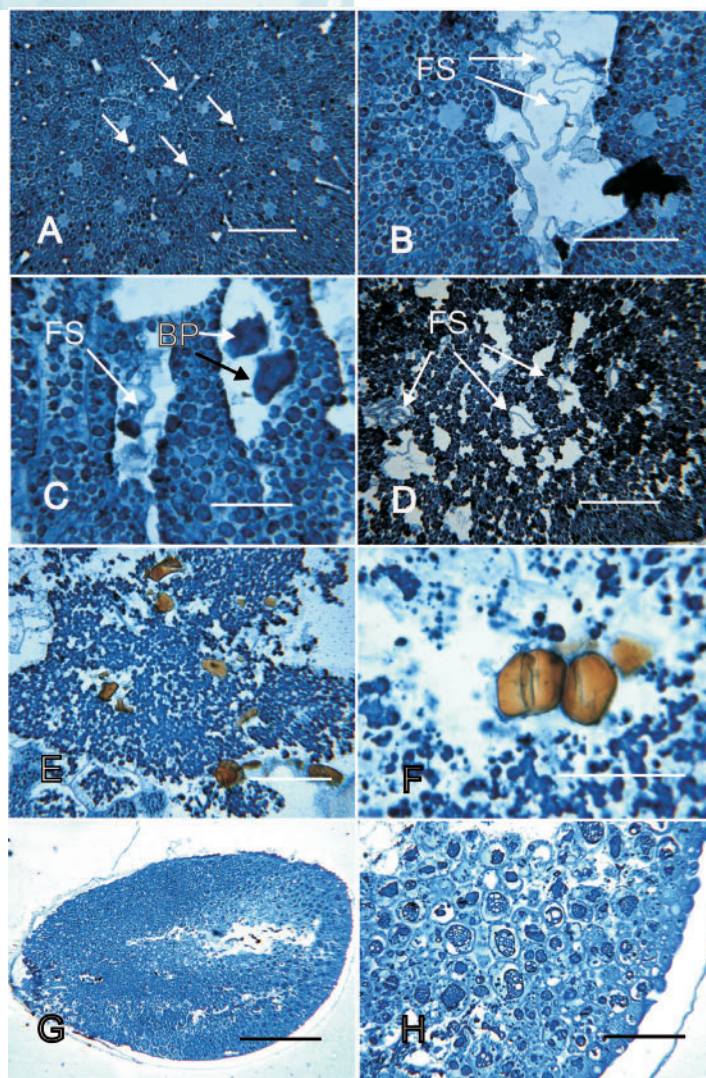


Plate 57. Degenerating seed of lodgepole pine. A to D) tissue degeneration type I: A. The first stage of degeneration was marked by many intercellular spaces (arrows); B. Filamentous structures (FS) in a larger intercellular space; C. Filamentous structures (FS) and blue particles (BP) in intercellular gaps; D. Type I tissue degeneration at late stage was marked by sandy appearance tissue with many spaces. Filamentous structures existed among the degenerating cells. E & F) type II degeneration: E. Appearance of cell liquidization and yellow particulate structures; F. A close look of yellow particles and amorphously coagulated cell contents in the background; G & H) Type III degeneration: G. Sign of mass cell death showed in a megagametophyte; H) A close look at degeneration type III at its early stage. Scale bar in G = 1mm, all other bars = 50µm.



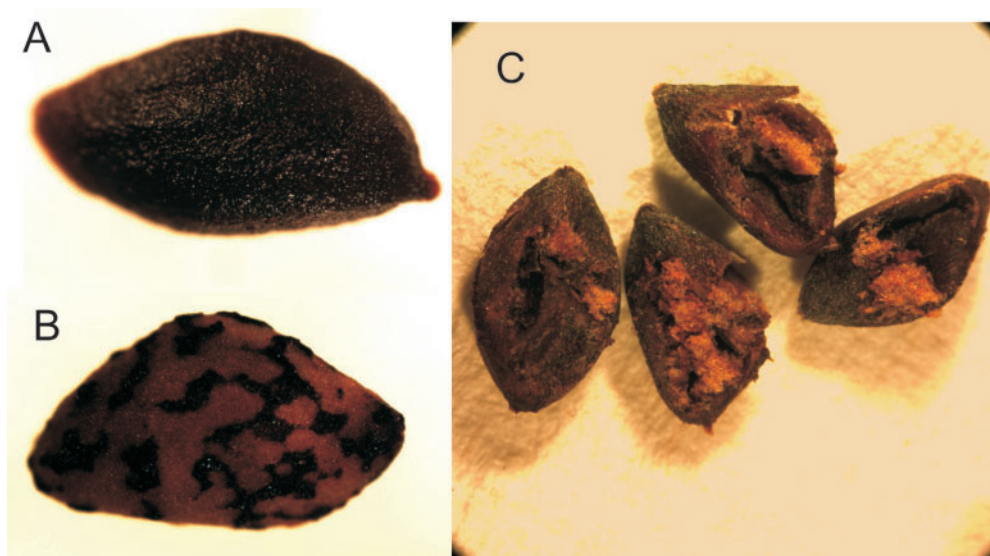


Plate 58. Comparison of normal and abnormal Pli seed coats. A) A seed showing its normal dark-brown seed coat. B) A seed with mottled seed coat. C) Empty seeds showing abnormal seed coats with unknown white structures on the surface.

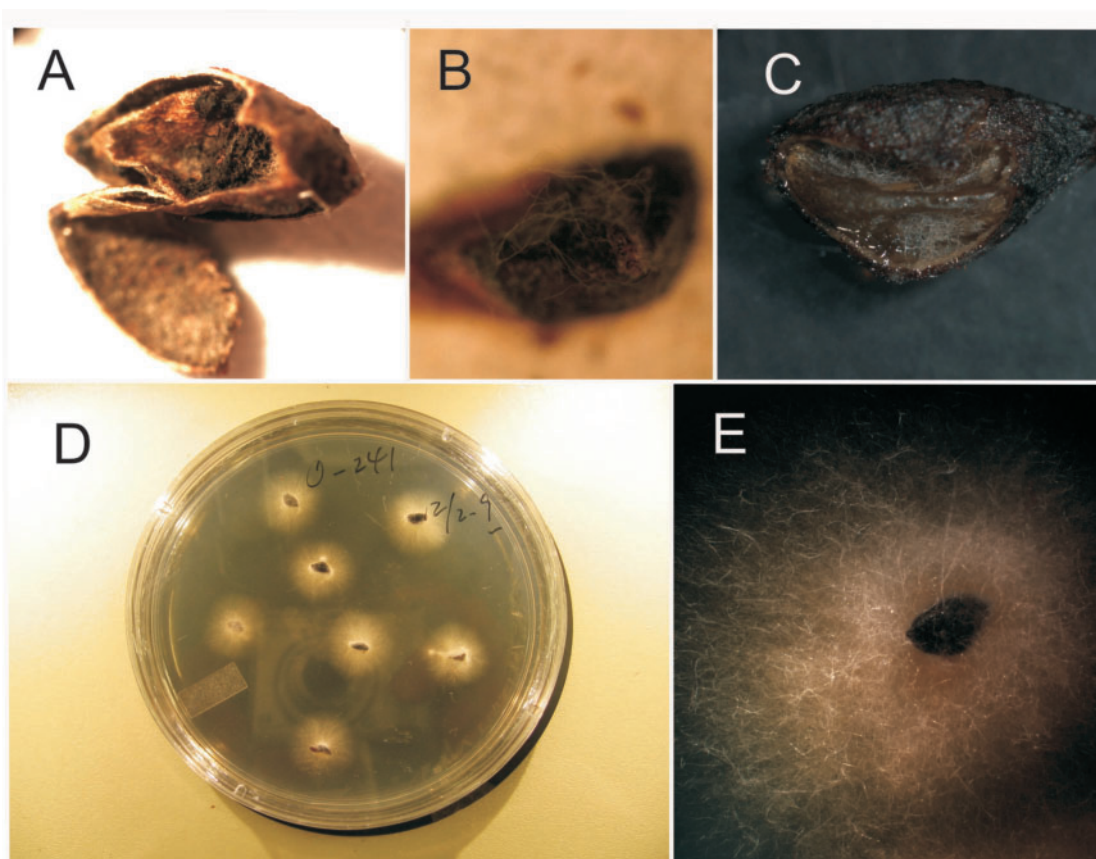


Plate 59. Seed borne fungi in lodgepole pine. A to C) show symptoms of fungal infection when seeds were dissected from the cone; D) infected seeds on fungus culture medium; E) Intensive fungus growth starting from a seed on the culture medium.

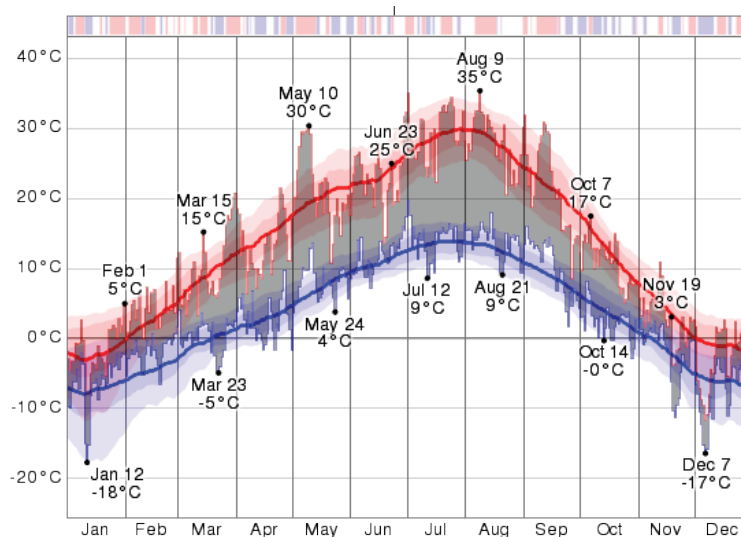


Figure 13. Annual temperature profile for Vernon BC in 2013 (<http://weatherspark.com/history/27809/2013/Vernon-British-Columbia-Canada>). The high temperature of May 10, 2013 was 30°C, which was 11°C higher than the average of 19°C. This high temperature in May of 2013 did not occur in the same month of 2012.

## Fungal identification

During the last quarter in 2013, we completed a preliminary molecular investigation on seed borne fungi isolated and cultured from surface-sterilized and non-sterilized seed (Plate 59 D, E). The identifications were based on sequence analysis of the internal transcribed spacer (ITS region) of the ribosomal DNA (rDNA) repeat region. In fungi, as in all higher eukaryotes, the highly conserved regions of the rDNA are separated by a highly variable ITS region. ITS RNA is spliced out of the precursor polycistronic RNA during the processing and folding of functional ribosomal RNA molecules. While the ribosomal RNA gene sequences are extremely well conserved, the intergenic spacer regions tend to be quite variable. Variation in this region is commonly used to identify fungi.

The most commonly identified fungi were *Sydowia polyspora* and/or *Rhizosphaera kalkhoffii*. *Sydowia* is a conifer pathogen associated with Current Season Needle Necrosis (CSNN), and *R. kalkhoffii* is the causative agent of a needle cast disease. *Sydowia polyspora* was recently discovered from seed in six out of seven conifer genera tested by Tølgø and coworkers (2012).

The most significant result to date was that fungi cultured from surface-sterilized seed consistently grouped with fungi of the *Rhizosphaera/Sydowia* species complexes. There were a small number of sequences that aligned with *Alternaria* spp. These same fungi were also found in

non-sterilized seed cultures along with some fungi that might be considered surface contaminants, e.g. *Gibberella/Fusarium*. The scores were very high for Query cover (99 – 100 %), low for E value (generally 0.0) and high for Identity (99%). However, confirmation of identity will rely on the amplification of additional markers. PCR primers could be designed for the specific amplification of each of these species groups from seed tissues as part of a routine screening process. Overall, this initial study with molecular tools proved that seed borne fungi were in Pli seeds.

## Project II. Cone induction in lodgepole pine and Douglas-fir

### Introduction

Flowering is a complex process that is controlled by multiple factors, among which plant hormones play an important role. Adjusting hormonal conditions *in vivo* does affect cone initiation and differentiation, thereby enhancing cone yield and seed production (Reviewed by Kong and von Aderkas 2007). Cone development differs in Douglas-fir and lodgepole pine. Consequently, the optimum treatment for cone induction will be species-dependent. In Douglas-fir, cone induction using combinations of a stress treatment, such as girdling, and gibberellin (GA) have proven more effective than using GA alone (Kong and von Aderkas 2007). Our experimental research suggested

the possibility of ethylene's involvement in cone bud initiation (Kong et al. 2012). Ethylene biosynthesis can be altered using compounds such as 2-chloroethyl phosphonic acid (ethephon) in combination of GA application to increase female cone yield in Douglas-fir. In the previous year (2012) cone induction treatments by stem injection of GA, ethephon, or a combination of the both were applied to Douglas-fir ramets of six genotypes, including five historically poor female cone producing genotypes. The treatment of GA in combination with ethephon was the best treatment based on data collected in spring 2013. Thus, more ethylene metabolism-related chemicals, methylglyoxal bis(guanylhydrazone) (MGBG) and putrescine (Put) (Botha and Whitehead, 1992), were applied in combination with GA for Douglas-fir cone induction in spring of 2013. In addition, 2,3,5-triiodobenzoic acid (TIBA), an auxin inhibitor, was also used to enhance effects of GA in cone induction.

In lodgepole pine, female and male cone development is normally restricted to different portions along an expanding long shoot. In addition, male cones are more abundant than female ones. Males are proximally distributed along the long shoot axis, whereas female cones are distally distributed: most are found near the branch apex. Occasionally, increased female cone formation is observed in proximal portions of long shoots normally restricted to male cone formation in a few genotypes. Our previous research demonstrated that certain phytohormone treatments further enhanced this phenomenon. For example, bud paste application of cytokinin and GA not only increased levels of a few important endogenous phytohormones, but also induced female cone formation in the proximal portion of long shoot buds (von Aderkas et al. 2010). Induction of cones in similar positions has been previously reported to incur during cone induction experiments in both Japanese red and black pines (Wakushima 2004).

Our goal is to develop cone induction treatments for both lodgepole pine and Douglas-fir for operational use in seed orchards. Three cone induction methods, i.e. bud paste, bud spray and stem-injection were used in 2012/13. Pli cone data collected in 2013 indicated that although higher female cone numbers were obtained with each of these methods, stem injection led to better results more consistently with a wider variety of genotypes. Greater genotype-dependent variation was observed with both the bud paste and bud spray treatment compared with stem injection. As a result, more attention was paid to stem-injection treatments for the spring/summer 2013.

## Plant materials and Methods

Ramets for 2013 cone induction were selected on the basis of previous cone yield records. Douglas-fir trees were selected and stem-injected at Pacific Regeneration Technologies Inc (PRT, Armstrong BC). Four ramets from each of six historically low cone-producing genotypes, i.e. reluctant genotypes, were treated. Stem-injections were applied to lodgepole pine trees at both Vernon Seed Orchard (VSOC, Vernon, BC) and Sorrento Seed Orchard (Sorrento, BC). At least six ramets per genotype were used for each treatment at each time point. For bud treatments, ten buds from each ramet were used per treatment, i.e. either bud paste or bud spray (Tables 15 to 18).

The type of GA used in our research was always a mixture of gibberellin A<sub>4</sub> and gibberellin A<sub>7</sub> (GA<sub>4/7</sub>). For stem-injections, GA was dissolved in methanol (MeOH, 50 mg/ml). MGBG and Put were dissolved in water (50 mg/ml). Usually 80 or 100 ml of each PGR was injected into one tree. Each hole held 1 ml of injection (Kong et al. 2012). Thus, multiple holes were needed to deliver PGRs in certain treatments. Bud paste was made by mixing the PGR solution with lanolin supplemented with either Vaseline or beeswax (Wakushima 2004). PGRs were first dissolved in a small amount of suitable solvent: GA in MeOH, TDZ in either MeOH or 1 N KOH, BA in 1N KOH, and TIBA in a solution of 50% pure MeOH and 50% 1N NaOH. The concentrated solution was then diluted with water for final concentrations for bud paste or spray. In the case of bud spray, the final solution also included 50 ml/l Tween 80 and was adjusted to pH 7 when possible. Approximately 1 ml of PGR paste was applied to each bud at each application time point. Bud spray was repeated one to two weeks following the initial treatment. Solutions for bud spray are as follows:

- 1) *GA-I* = GA 1g/l pH 7, Tween 80 50 ml/l
- 2) *GA-II* = GA 2g/l pH 7, Tween 80 0 ml/l
- 3) *GA+TDZ* = GA 1g/l + TDZ 30 mg/l, pH 8, Tween 80 50 ml/l
- 4) *GA+BA* = GA 1g/l + BA 0.5 g/l, pH 9-10, Tween 80 50 ml/l

Douglas-fir data was based on numbers of female cones per ramet. For stem-injection treatments, data was collected from ramets that had either received a PGR treatment or not, in the case of controls.

Lodgepole pine data on bud treatments was collected differently. The number of female cones per long shoot data for each type of bud treatment, e.g. bud paste or bud spray, were collected in the growing season following application. Controls were as follows: the same long shoot formed in the previous year before PGR treatment, as well as other long shoots from the same ramet that had not been given a PGR treatment in the previous two years. For stem-injection treatments, data was collected from ten randomly selected long-shoot/ramet. The ramets had either received a PGR treatment or not, in the case of controls.

## Results and discussion

### Douglas-fir

The highest response to hormone treatment was obtained with the combination of GA and ethephon (Table 15), which was significantly ( $P<0.05$ ) higher than the control. Four of six genotypes were higher than the controls. The average number of female cone per ramet was also highest in the GA and ethephon combination treatment (Table 16) and significantly ( $P<0.05$ ) higher than the control (no injection).

Treatment	Genotype 9137	Genotype 8208	Genotype 9115	Genotype 9148	Genotype 9550	Genotype 8237	Mean $\pm$ SE, n=6
GA100	0.0	100.0	100.0	33.3	66.6	0.0	50.0 $\pm$ 18.8
Eth100	0.0	66.6	100.0	0.0	33.3	0.0	33.3 $\pm$ 17.2
GA+Eth	33.3	100.0	100.0	66.6	100.0	0.0	66.7 $\pm$ 17.2
No injection	0.0	33.3	100.0	0.0	33.3	0.0	27.8 $\pm$ 15.9

Table 15. Percentages of female cone formation (n=3 for each genotype) in Douglas-fir. Treatments were applied to five reluctant genotypes in Douglas-fir by stem-injection. Treatment application was completed in spring 2012 in PRT seed orchard. Data was collected in spring 2013. GA- Gibberellin, Eth- Ethephon

Treatment	Genotype 9137	Genotype 8208	Genotype 9115	Genotype 9148	Genotype 8237	Mean $\pm$ SE, n=5
GA100	0.0	75.3	41.7	3.7	0.0	24.1 $\pm$ 15.0
Eth100	0.0	79.5	11.0	0.0	0.0	18.1 $\pm$ 15.5
GA+Eth	46.0	83.7	26.7	3.7	0.0	32.0 $\pm$ 15.4
No injection	0.0	0.7	9.3	0.0	0.0	2.0 $\pm$ 1.8

Table 16. Comparison of average number of female cones per ramet (n=3 for each genotype) in Douglas-fir. Treatments were applied to five genotypes of poor female cone yield in Douglas-fir by stem-injection. Treatment application was completed in spring 2012 in PRT seed orchard. Data was collected in spring 2013. GA- Gibberellin; Eth- Ethephon.



## Lodgepole pine

Three cone induction methods, i.e. bud paste, bud spray and stem injection, were used to induce cones in both the Sorrento and Vernon seed orchards. Female cone clusters were induced in atypical proximal positions in branches of ramets of numerous genotypes. These female buds were formed from buds that otherwise would have formed male cones (Plate 60 A-C), or long shoots (Plate 60 D). In previous years, female cone clusters were induced only with bud paste treatment. In 2012/13, the clusters were induced in seven genotypes (2082, 1799, 1822, 327, 4046, 4065, and 4142) by some or all of the three treatments (bud paste, bud spray, or stem-injection). Furthermore, a number of treatments were successful, including GA, or GA in combination with various cytokinins, (TDZ or BA). Cone induction treatments with cytokinins alone had little effect.

GA alone or in combination with a cytokinin stimulated female cone formation in many genotypes. Data in Table 17 shows induction results in six genotypes. Responses to bud treatments could be grouped into 4 types:

- 1) high response to both paste and spray, e.g. genotypes 2082, 1799, 1822
- 2) high response to spray but not paste, e.g. 4142, 4065
- 3) high response to spray only, e.g. 4036
- 4) low response in all treatments, e.g. 4066.

The following list gives a general genotype response to bud treatments from the best to the worse: 2082, 1799, 1822, 327, 4046, 4065, 4142, 4339, 4132, 4157, 4147, 4168, 4047, 1775, 4036, 4043, 4036, 4043, and 4066 (data not shown). Based on the results, some adjustments were made for bud treatments in order to optimize response for most genotypes in the trial that would carry over from 2013 to 2014.

			2082			1799			1822		
			CT	Tr	Tr/CT (%)	CT	Tr	Tr/CT (%)	CT	Tr	Tr/CT (%)
Paste	GA	Mean	1.06	2.16	203.92	1.33	2.42	182.26	0.78	1.83	233.81
		SE	0.25	0.28		0.33	0.37		0.29	0.67	
	GA+BA	Mean	0.94	1.46	154.42	1.26	1.80	143.24	0.60	0.70	116.67
		SE	0.18	0.48		0.37	0.44		0.07	0.25	
	GA+TDZ	Mean	0.50	1.67	334.00	1.13	2.69	238.89	0.29	1.34	457.53
		SE	0.38	0.07		0.31	0.60		0.11	0.20	
Spray	GA	Mean	0.67	1.97	296.00	0.83	1.13	135.60	1.08	2.64	245.51
		SE	0.42	0.47		0.18	0.32		0.17	0.14	
	GA+BA	Mean	0.48	1.22	255.94	1.27	1.59	124.80	0.43	1.00	235.29
		SE	0.24	0.76		0.02	0.59		0.25	0.36	
	GA+TDZ	Mean	1.00	2.20	220.00	1.31	1.13	85.88	0.50	1.25	250.00
		SE	0.4	0.6		0.19	0.63		0.3	0.6	
			4142			4065			4036		
			CT	Tr	Tr/CT (%)	CT	Tr	Tr/CT (%)	CT	Tr	Tr/CT (%)
Paste	GA	Mean	0.80	0.66	82.50	1.38	2.19	158.33	1.00	0.38	38.00
		SE	0.20	0.13		0.02	1.09		0.50	0.31	
	GA+BA	Mean	0.65	0.55	84.62	1.20	0.40	33.33	0.37	0.30	81.82
		SE	0.25	0.25		0.4	0.4		0.12	0.15	
	GA+TDZ	Mean	0.93	0.32	34.76	1.20	2.12	176.25	0.53	0.18	34.81
		SE	0.20	0.18		0.40	0.01		0.07	0.09	
Spray	GA	Mean	0.58	1.95	339.13	0.88	2.48	281.82	0.24	0.38	161.70
		SE	0.15	0.52		0.32	1.08		0.04	0.08	
	GA+BA	Mean	0.37	1.08	296.58	1.00	3.20	320.00	0.19	0.15	78.95
		SE	0.22	0.22		0.3	0.5		0.19	0.15	
	GA+TDZ	Mean	1.40	1.80	128.57	0.34	1.60	433.33	0.56	0.22	39.29
		SE	0.3	0.4		0.3	0.5		0.3	0.2	

Table 17. Comparison of average number of female cones per long shoot among various genotypes following application of plant growth regulators (PGRs). Treatments of bud paste or bud spray of PGRs were applied in early summer of 2012. Data collection was completed in summer 2013. Mean  $\pm$  SE, n= 40 long shoots, or occasionally n  $\geq$  10. CT – control, Tr – treatment, GA – GA<sub>4/7</sub>

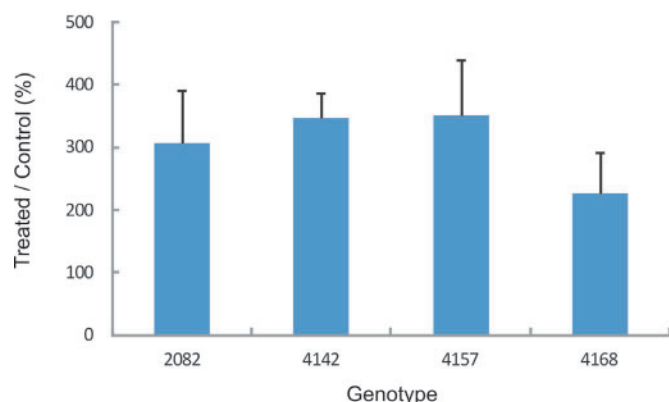


Figure 14. Cone induction percentage response of treated compared to untreated (control). Female cones were induced by stem-injection of GA in early summer of 2012. Data was collected in late spring 2013. Mean  $\pm$  SE,  $n = 4$  ramets.

Stem injection treatments had less variation in genotype response than was observed with bud treatments. This may be due to higher local concentrations of PGRs in bud treatments (not measured) or it may be due to genotype-PGR treatment interactions. Although some ramets of genotype 2082 are able to generate female cone clusters without any hormone supplementation, increases in female cone yield were widely obtained following injection of GA

(Figure 14). Injection with GA, especially in combination with BA, stimulated formation of female cone clusters (Plates 60 and 61). Female cone clusters were induced in the positions that male cones usually occupy. This implies that bud gender is under hormonal control. The number of female cones in each cone cluster occasionally exceeded twenty. Since these clusters are easily distinguishable from other female cones both by their high number of cones and their position on the long shoot, this would be a good system for studying gender determination in conifers.

In summary, enhanced cone induction in reluctant Douglas-fir genotypes was achieved by injection of GA along with ethephon. In 2013/14, a number of ethylene metabolism-related compounds were used in addition to GA<sub>4+7</sub>, for cone induction in Douglas-fir. In lodgepole pine, female cone clusters were induced from multiple genotypes with both pastes and sprays applied directly on developing buds. Stem-injection of GA or GA plus BA also stimulated female cone formation. This is the first time that we were able to induce female cone clusters by stem-injection. More treatments of stem-injection with GA plus cytokinin (BA or TDZ) were applied in a forward cone induction experiment that is to be evaluated in late spring 2014. There is always a certain amount of genotype variation in cone induction responses in both Douglas-fir and lodgepole pine.

Plate 60. Female cone induction in lodgepole pine by exogenously applied plant growth regulators. A & B) Female cone (FC) clusters induced by bud paste treatment (genotypes 1822, 4046); C) A female cone cluster induced by stem-injection (genotype 2082). The induced female cones (A, B and C) localized at the lower part of a long shoot where male cones (MC) usually develop. D) Female cones were induced from long-shoot buds (LSB) in genotype 4065.

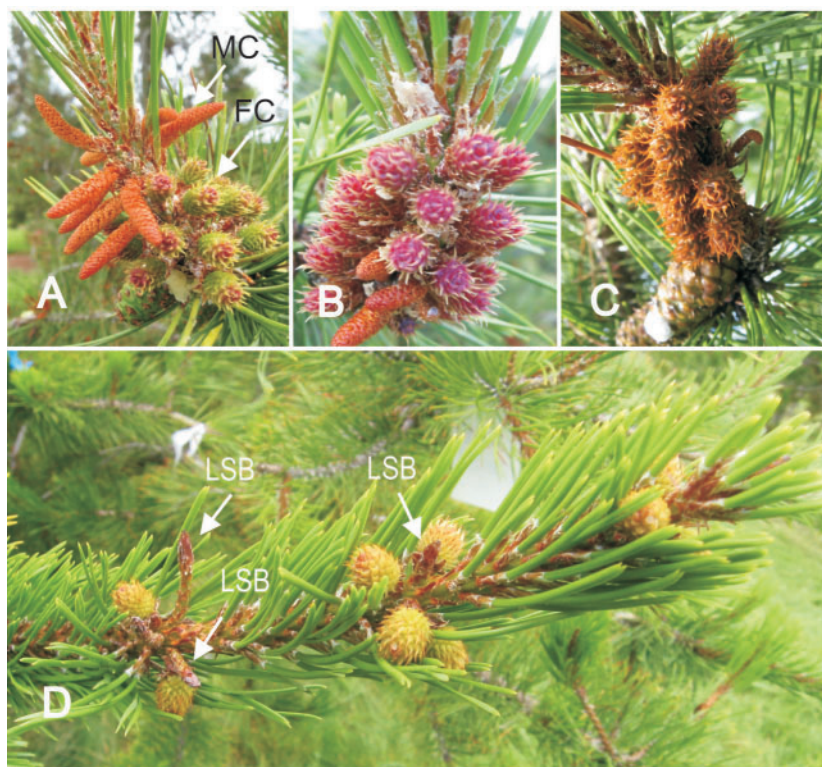




Plate 61. Female cone clusters (circled) induced by stem-injection treatment. GA in combination with BA was applied to ramets of genotype 2082 at Sorrento seed orchard in 2012. Photo was taken in early summer of 2013.

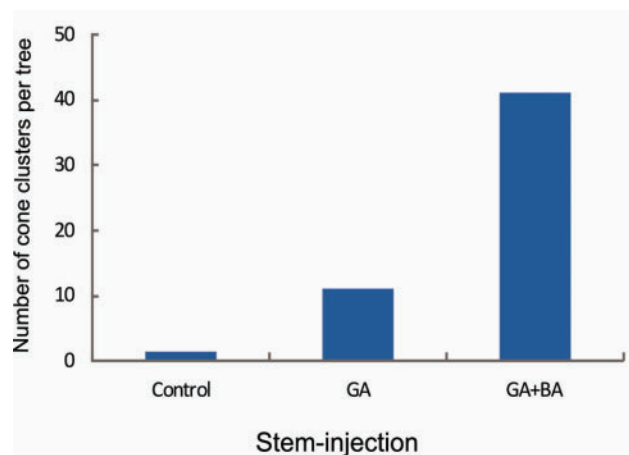


Figure 15. Number of female cone clusters per tree. Female cone clusters were induced by stem-injection of GA, or GA plus BA. PGRs were applied to ramets of genotype 2082 at Sorrento seed orchard in early summer of 2012. Data was collected in spring 2013. Mean, n= 4, or ≥2 ramets.

Date	Genotype Treatment	9196	9148	8238	8237	9220	8853
May16	GA	E1	A5	C25	E21	E28	P7
	GA	K28	C12	C32	H36	G34	C22
	GA	R2	F17	D38	K20	K24	D34
	GA+MGBG	S32	G7	I34	M26	L15	H5
	GA+MGBG	A34	G26	I20	S9	M3	H26
	GA+MGBG	X1	L3	M31	T17	M32	J11
	GA+TIBA	ZB6	L9	M14	V3	O37	J33
	GA+TIBA	ZC19	P31	P19	W39	P20	ZC33
	GA+TIBA	ZG2	Q6	R36	ZA35	Q27	ZE1
	GA+Put	ZH8	Q25	T4	ZF10	X3	ZG18
	GA+Put	ZJ10	S15	U16	ZG32	Y31	ZH12
	GA+Put	ZQ17	U33	U22	ZH20	ZA38	ZK6
June12	GA	ZQ22	V39	ZQ12	ZO9	ZD4	ZK6
	GA+TIBA	ZQ28	V7	Z3	ZK13	ZF34	ZN28
	GA+MGBG	ZQ39	ZB23	ZF8	ZI5	ZL25	ZP4
Control	No injection	ZZ1	ZB34	ZK37	ZO9	ZL37	ZP23
	No injection	ZZ23	ZK28	ZP37	ZO35	ZM3	ZR16
	No injection	ZZB30	ZO10	ZQ12	ZQ25	ZP34	ZZ26
	No injection	ZZC6	ZS29	ZY11	ZT30	ZU15	ZZ39
	No injection	ZZE19	ZS35	ZZA21	ZZ20	ZV33	ZZC15

Table 18. Douglas-fir cone induction treatments applied in 2013. Treatments were applied to ramets at the seed orchard of Pacific Regeneration Technologies Inc (PRT) by stem-injection. Treatments included gibberellins A<sub>6</sub> and A<sub>3</sub> (GA, 100 mg/tree) and combinations of GA and methylglyoxal bis(guanylhydrazone) (MGBG, 150 mg/tree), 2,3,5-triiodobenzoic acid (TIBA, 100 mg/tree) or putrescine (Put, 200 mg/tree) at two time points in May and June respectively.



## Application of cone induction treatments in 2013

Douglas-fir cone induction treatments were applied at PRT seed orchard starting May 16. Four treatments were included at two time points. The details of the treatments were provided in Table 18. For lodgepole pine, cone induction treatments were applied at VSOC and Sorrento seed orchard, starting middle of May. The majority of the work was completed during June and July. Hundreds of trees were used for induction treatments. Various plant growth regulators were applied including GA, TDZ, BA, MGBG, Putrescine as well as TIBA. These were introduced as either stem injection, bud paste, or bud spray.

## Acknowledgements

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## 7.0 Seed Orchard Pest Management

Jim Corrigan

The objective of the Seed Orchard Pest Management Subprogram is to provide research, extension support and orchard-level pest management to increase yields of high quality seed coming from the seed orchards of our province. Research and extension activities are carried out through the Tree Improvement Branch of the BC Ministry of Forests, Lands and Natural Resource Operations (FLNR). Dr. Ward Strong has a half-time responsibility to conduct pest management research, while Jim Corrigan delivers extension services to Interior and Coastal operations. Seed orchard personnel handle hands-on pest management duties at the orchard locations.

The Pest Management Technical Advisory Committee (PMTAC) manages annual LBIS funding allocations that come through the Forest Genetic Council to support relevant pest management research. Our Committee members come from the FLNR Tree Improvement Branch, the Canadian Forest Service, and industry. The PMTAC establishes research priorities and budgets through an annual process of proposal development and evaluation. In fiscal 2013, the PMTAC supported projects to test the efficacy of attract-and-kill techniques to control *Synanthedon sequoia*, the Sequoia pitch moth; assess novel pesticides for efficacy against *Contarinia oregonensis*, the Douglas-fir cone gall midge; determine if attacks by *Leptoglossus occidentalis*, the western conifer seed bug, can be detected on individual conifer seeds; conduct pesticide screening trials against the white-grub larvae of *Polyphylla crinita*, the long-haired June beetle; support research initiatives on cone and seed pests, and support pest management extension activities around the province.



Plate 62. Large numbers of white grub larvae of the long-haired June beetle, *Polyphylla crinita* can be found in the soil at the PRT seed orchards. (Jim Corrigan image).

Project	Species primarily impacted	Progress
Pesticide trials	Fd, Pw	Continued ongoing projects to find new, safe and effective formulations of pesticides for the control of <i>Contarinia oregonensis</i> in Douglas-fir. Results from these trials will be used to support applications for Minor Use registrations made to the Canadian Pest Management Regulatory Agency (PMRA).
Pesticide screening trial targeting the larvae of the long-haired June Beetle	Fdi, Pli	Treatments of Merit and Diazinon appear to have significantly reduced the amount of root damage to potted lodgepole pine rootstock. Reductions in larval numbers in the pots were less obvious. Diazinon was the only chemical to reduce larval numbers below those seen on untreated controls.
The <i>Synanthedon sequoiae</i> pitch moth attract-and-kill trial	Pli	This project replicated successful results obtained in similar trials done in 2012. Droplets that combined the sex pheromone for the Sequoia pitch moth, <i>S. sequoiae</i> , with a pesticide were applied in a grid inside lodgepole pine seed orchards. These treatments appeared to significantly reduce pitch moth attack rates at several Interior locations.
Detecting <i>Leptoglossus</i> seed bug feeding punctures on lodgepole pine seeds	Pli	This project replicated successful results obtained in similar trials done in 2012. Samples of lodgepole pine seeds that had been exposed to seed bug attack were put through a staining process. Staining heated seeds in a cooled ruthenium red solution could be used to identify individual lodgepole seeds that have been attacked by seed bugs.
Support for operations of the research lab	All species	Funding was provided to support on-going lab operations and provide for technical assistance in support of pest management research activities.
Support for extension activities	All species	Funding provided for on-going extension support to Interior and Coastal seed orchard locations and to the Tree Seed Centre.

Table 19. Summary of PMTAC Projects for fiscal 2013.

## Appendix 1 FGC Seed Planning Unit

Seed planning unit (SPU)					Program category
#	Species	Common Name	SPZ	Elev. band (m)	
1	Fdc	Douglas-fir	M	1-900	1
2	Cw	Western redcedar	M	1-700	1
3	Hw	Western hemlock	M	1-600	2
4	Sx	Interior spruce	NE	1000-1700	1
5	Sx	Interior spruce	NE	1700-2100	2
6	Ss	Sitka spruce	M	1-500	2
7	Pli	Lodgepole pine	NE	700-1600	1
8	Pw	Western white pine	M/SM	1-1000	1
9	Ba	Amabilis fir	M	1-1000	3
10	Pli	Lodgepole pine	TO	700-1400	1
11	Yc	Yellow cypress	M	1-1100	2
12	Pli	Lodgepole pine	PG	700-1400	1
13	Lw	Western larch	NE	700-1600	1
14	Sx	Interior spruce	PG	600-1400	1
15	Pw	Western white pine	KQ	500-1400	1
16	Pli	Lodgepole pine	TO	1400-1600	2
17	Pli	Lodgepole pine	BV	700-1400	1
18	Pli	Lodgepole pine	CP	700-1300	1
19	Fdc	Douglas-fir	SM	200-1000	2
20	Pli	Lodgepole pine	NE	1600-2000	2
21	Fdi	Douglas-fir	NE	400-1200	1
22	Fdi	Douglas-fir	NE	1000-1800	2
23	Sx/Ss	Spruce	SM/NST	all	2
24	Hw	Western hemlock	M	600-1100	2
25	Sx	Interior spruce	EK	750-1900	2
26	Pli	Lodgepole pine	PG	1400-2000	3
27	Cw	Western redcedar	SM	200-1000	2
28	Sx	Interior spruce	TO	1300-2100	2
29	Pli	Lodgepole pine	EK	1500-2000	2
30	Sx	Interior spruce	TO	700-1500	1
31	Fdc	Douglas-fir	M	900-1200	2
32	Pli	Lodgepole pine	EK	800-1500	2
33	Cw	Western redcedar	M	700-1500	2
34	Lw	Western larch	EK	800-1700	1
35	Sx	Interior spruce	BV	500-1400	2
36	Bg	Grand fir	M	1-700	3
37	Fdi	Douglas-fir	QL	700-1400	2
38	Hw	Western hemlock	M north	1-600 (part of SPU 3)	
39	Fdi	Douglas-fir	EK	700-1400	2
40	Sx	Interior spruce	PR	<650 & 650-1200	2
41	Fdi	Douglas-fir	PG	700-1200	2
42	Sx	Interior spruce	PG	1200-1550	2
43	Fdi	Douglas-fir	CT	600-1400	2
44	Sx	Interior spruce	NE	1-1000	1
45	Pli	Lodgepole pine	BB/CHL	All	3
46	Bl	Sub-alpine fir	all int.	all	3
47	Bn	Noble fir	M	all	3
48	Aspen/birch/poplar		Interior	-	3
49	Alder/poplar/maple		Coast	-	3
50	Lw	Western larch	NE	1200-1800	2
51	Py	Ponderosa pine	S. Interior	300-1200	2

## Appendix 2 Tree Species

CONIFERS	LATIN NAME	TREE SPECIES CODES
western redcedar	<i>Thuja plicata</i>	Cw
yellow cypress	<i>Callitropsis nootkatensis</i>	Yc
coastal Douglas-fir	<i>Pseudotsuga menziesii</i> var. <i>menziesii</i>	Fdc
interior Douglas-fir	<i>Pseudotsuga menziesii</i> var. <i>glauca</i>	Fdi
amabilis fir	<i>Abies amabilis</i>	Ba
grand fir	<i>Abies grandis</i>	Bg
noble fir	<i>Abies procera</i>	Bp
subalpine fir	<i>Abies lasiocarpa</i>	Bl
mountain hemlock	<i>Tsuga mertensiana</i>	Hm
western hemlock	<i>Tsuga heterophylla</i>	Hw
Rocky Mountain juniper	<i>Juniperus scopulorum</i>	Jr
alpine (subalpine) larch	<i>Larix lyallii</i>	La
western larch	<i>Larix occidentalis</i>	Lw
limber pine	<i>Pinus flexilis</i>	Pf
interior lodgepole pine	<i>Pinus contorta</i> var. <i>latifolia</i>	Pli
Ponderosa pine	<i>Pinus ponderosa</i>	Py
shore pine	<i>Pinus contorta</i> var. <i>contorta</i>	Plc
western white pine	<i>Pinus monticola</i>	Pw
whitebark pine	<i>Pinus albicaulis</i>	Pa
Engelmann spruce	<i>Picea engelmannii</i>	Se
Sitka spruce	<i>Picea sitchensis</i>	Ss
white spruce	<i>Picea glauca</i>	Sw
spruce hybrid (interior spruce)	<i>Picea</i> cross (Se and Sw mixtures)	Sx
Sitka x unknown hybrid	<i>Picea sitchensis</i> x	Sxs
western (Pacific) yew	<i>Taxus brevifolia</i>	Tw
<b>HARDWOODS</b>		
bigleaf maple	<i>Acer macrophyllum</i>	Mb
red alder	<i>Alnus rubra</i>	Dr
black cottonwood	<i>Populus balsamifera</i> ssp. <i>trichocarpa</i>	Act
hybrid poplars	<i>Populus</i> spp.	Ax
trembling aspen	<i>Populus tremuloides</i>	At
paper birch	<i>Betula papyrifera</i>	Ep
Garry oak	<i>Quercus garryana</i>	Qg



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