# WFGA – 50<sup>th</sup> Anniversary Looking back – Looking Ahead



Program

Joint meeting of:

Western Forest Genetics Association Northwest Seed Orchard Managers Association North American Quantitative Forest Genetics Group

> LaSells Stewart Center Oregon State University Corvallis, Oregon July 19-21, 2005





North West Seed Orchard Managers Association

NAQFGG

# WFGA Hosting Organizations:

# **Department of Forest Science, Oregon State University**

Forest Genetics Team, PNW Research Station, USDA Forest Service







# Organizing Committee:

Randy Johnson – Chair, USDA Forest Service Tom Adams, Oregon State University Mike Albrecht, Roseburg Forest Products Marilyn Cherry, Oregon State University Rich Cronn, USDA Forest Service Keith Jayawickrama, Oregon State University Glenn Howe, Oregon State University Nancy Mandel, USDA Forest Service Brad St.Clair, USDA Forest Service Steve Strauss, Oregon State University

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# WFGA – 50<sup>th</sup> Anniversary, Looking back – Looking ahead Program at a glance

# **Tuesday:**

7:15 am	Registration and Coffee
8:00 - 11:50	Plenary session I - Construction & Engineering Hall
11:50 - 1:00	Lunch (provided) – Giustina Gallery
1:00 - 2:20	Concurrent Session Ia - Student Molecular Studies – Construct. & Engineering Hall
1:00 - 2:20	Concurrent Session II – Student Presentations – Ag Production Room
2:20 - 2:40	Break
2:40 - 4:30	Concurrent Session Ib - Molecular Studies - Construction & Engineering Hall
2:40 - 4:30	Concurrent Session III - Seed Orchard Management - Ag Production Room
5:00 - 6:00	NWSOMA Business Meeting – Ag Production Room
5:00 - 6:30	North American Quantitative Forest Genetics Group – Construction & Eng. Hall
Dinner	On your own

# Wednesday:

Field Trip – Depart from LaSells Stewart Center 8:00 am
DLF International Seeds - Grass Breeding
Oregon Department of Forestry – Schroeder Seed Orchard (St.Paul, OR)
Lunch Provided
Landscape Plant Development Center (Aurora OR),
Oregon Garden (Silverton OR), Tour & Banquet, Speaker: Don Howse, Porterhouse Farms

# Thursday:

- 7:30 Coffee
- 8:00 9:10 Plenary session II Construction & Engineering Hall
- 9:40 10:10 Break
- 10:10 12:10 Concurrent Session IV Deployment & Breeding- Construct. & Engineering Hall
- 10:10 12:10 Concurrent Session V Population Genetics Ag Production Room
- 12:15 1:15 Lunch (provided)
- 1:20 2:40 Plenary Session III Disease Resistance Construction & Engineering Hall
- 2:40 3:00 Break
- 3:00 3:30 WFGA Business Meeting Ag Production Room

# WFGA – 50<sup>th</sup> Anniversary, Looking back – Looking ahead Session Schedules

# Tuesday, July 19

- 8:00 am Welcome and Introductions Construction & Engineering Hall Randy Johnson, Tom Adams
- 8:15 12:00 Plenary session I Construction & Engineering Hall Moderator: Tom Adams
  - 8:15 Overview of the past 50 yrs Bill Libby
  - 8:50 Genetics in horticulture Harold Pellett
  - 9:20 Genetics in agronomic crops Rich Cronn

#### 10:00 - Break

- 10:30 A forestry perspective to year 2020 Bob Kellision
- 11:10 A comparison of seedling vs. clonal forestry in Queensland Mark Dieters
- 11:50 1:00 Lunch (provided) Giustina Gallery
- 1:00 2:20 Concurrent Session Ia Student Molecular Studies Constuction & Eng. Hall Moderator: Brad St.Clair
  - 1:00 Candidate gene-quantitative phenotype associations for resistance against Fusiform rust and pitch canker in loblolly pine Elhan Erosoz
  - 1:20- Evidence of multiple glacial refugia for *Pinus contorta* inferred from a mtDNA minisatellite marker Julie Godbout
  - 1:40- Identifying candidate genes associated with cold hardiness in Coastal Douglas-fir using DNA microarrays Dana K. Howe
  - 2:00- Molecular evolutionary rates indicate a recent and rapid diversification of modern pine lineages Ann Willyard

# **1:00 – 2:20 - Concurrent Session IIa – Student Presentations – Ag Production Room** Moderator: Greg O'Neill

- 1:00- Genetic diversity in whitebark pine is affected by white pine blister rust infection -Andrew D. Bower
- 1:20- Improving breeding trial statistical analysis through post-hoc blocking S.A. Gezan
- 1:40- Phenotypic and genetic correlations of commercially important wood quality traits in Coastal Douglas-fir N. K. Ukrainetz
- 2:00- Genetic differentiation of ponderosa pine and a dominant understory grass in northern Arizona: implications for restoration Huarong Zhang

## **Tuesday, July 19**

# 2:40-4:30 - Concurrent Session Ib - Molecular Studies – Construction & Eng. Hall Moderator: Valarie Hipkins

- 2:40- Development of genomic tools for poplar Palitha Dharmawardhana
- 3:00- A conserved CC-NBS-LRR resistance gene in sugar pine K. Jermstad
- 3:20- Mapping adaptive traits in Douglas-fir using association genetics B. Pande
- 3:40- Polymix breeding with paternity analysis in Populus: A test for differential reproductive success (DRS) among pollen donors N. Wheeler

# 2:40-4:30 - Concurrent Session IIb - Seed Orchard Management – Ag production Room Moderator: Jim Smith

- 2:40- Cone stimulation of *Abies procera* Evaluating variable rates of GA 4/7, application timing and girdling Chal Landgren
- 3:00- Using Microsoft Access to manage your seed orchard information Jeff DeBell
- 3:20- Douglas-fir controlled mass pollination pilot project Rick Quam
- 3:40- Operational application and evaluation of Douglas-fir cone gall midge suppression using trunk-injected Imidacloprid Beth Willhite
- 4:10- Results of an operational stimulation in a young DF Seed Orchard Mike Albrecht

#### 5:00 – 6:30 - North American Quantitative Forest Genetics Group Moderator: Randy Johnson

- 5:00- Bridging breeding and genomics by quantitative genetics Bailian Li
- 5:30- Parameter estimation for stochastic simulation of multiple traits over more than one generation Jen Myszewski
- 6:00- Efficiency of spatial analysis of first-generation Coastal Douglas-fir progeny trials in the US Pacific Northwest – Terrance Ye

### Dinner – on your own

## Wednesday, July 20

Field Trip – Depart from LaSells Stewart Center 8:00 am
DLF International Seeds - Grass Breeding
Oregon Department of Forestry – Schroeder Seed Orchard
Landscape Plant Development Center (Aurora OR),
Oregon Garden
Tour of Garden
Banquet
Sharing of anecdotes over the last 50 yrs (Lauren Fins, MC)
Invited Speaker: Don Howse, Porterhouse Farms

# Thursday, July 21:

7:30 Coffee

8:00 – 9:10 Plenary session II - Construction & Engineering Hall Moderator: Glenn Howe

- 8:00- The genomics revolution- Implications on nutrition and medicine Emily Ho
- 8:30- Studying complex phenotypes in humans Patricia Kramer
- 9:00- Deployment of genetically improved material: Anything new lately? A Yanchuk
- 9:20- Growth vs. defense adaptations: Is there a trade-off? John King

#### 9:40- Break

- 10:10-12:10 Concurrent Session III Breeding and Deployment Construct. & Eng. Hall Moderator: Marc Rust
  - 10:10- The seed transfer impact calculator G. O'Neill
  - 10:30- Impacts of climate change and potential adaptation strategies through genetic resource management in BC A. Hamann
  - 10:50- Clonal forestry vs. seedling-based tree improvement for Coastal Douglas-fir in the US Pacific Northwest KJS Jayawickrama
  - 11:10- Douglas-fir tree improvement in the British Columbia Interior: results and opportunities B. Jaquish
  - 11:30- Recent Changes in the Genetic Improvement of Conifers in Queensland Mark Dieters
  - 11:50 Advanced propagation of Douglas-fir Christine Dean

## Thursday, July 21

- 10:10-12:10 Concurrent Session IVa Population Genetics Ag Production Room Moderator: Steve Strauss
  - 10:10- Genetic structure of core and peripheral populations: Implications for research and *ex situ* conservation S. Aitken
  - 10:30- Early allocation of resources to shoots versus roots of geographically diverse populations of western hemlock selected for improved juvenile height growth Charlie Cartwright
  - 10:50- Geographic variation for height and diameter among Black Spruce (*Picea mariana*) Provenances in Alberta – D. Rweyongeza
  - 11:10- Full-sib analysis of pollen flow and pollen structure in Guanacaste (*Enterolobium cyclocarpum*) in the Costa Rican dry tropical forest Peter Smouse
  - 11:30- Pollen dispersal redux: A look at recent analyses Bob Westfall
  - 11:50- Landscape genetics of western white pine: Genetic structure and evolutionarily significant units? G. McDonald

### 12:15-1:15 Lunch (provided)

- **1:20 3:00 Plenary Session III Disease Resistance Construction & Engineering Hall** Moderator: Keith Jayawickrama
  - 1:20- Genetic studies of Acacia Koa and Koa Wilt Disease J. Brewbaker
  - 1:40- Genetic resistance in Port-Orford-cedar to *Phytophthora lateralis* survival of seedlings from first orchard seed in short-term testing K. Mylecraine
  - 2:00- Time is On Our Side Blister Rust Resistance Traits "Holding Up" After 30+ Years - L. Fins
  - 2:20- White pine blister rust resistance in western white pine Family variation in infection percent and survival in field trials R. Sniezko

#### 3:00 – 3:30 - WFGA Business Meeting - Ag Production Room

### Posters – Authors are asked to be at their posters during the break on Tuesday afternoon

Flowering behavior of Port Orford Cedar from different provenances grown in a common garden J. Hill

Variation in wood quality of Coastal Douglas-fir - R. Johnson

30-year results of a Douglas-fir progeny test series. - R. Kelly

Inbreeding depression in Port-Orford-Cedar; early survival and growth of self-fertilized and openpollinated progeny – S. Kolpak

# **Fifty Years of WFGA**

# Bill Libby and Brad St.Clair

The Western Forest Genetics Association, officially founded in 1955, was established at a time when tree breeding and forest genetics were in an expansion and optimistic mode. It was preceded by a half-century of ideas and research in western North America, and by a much longer foundation elsewhere.

WFGA has been unusual, in that our meetings have not published *Proceedings*. Our philosophy has been to present and discuss things we are doing, or are planning to do, although reports of completed works have been presented and welcome. Field trips to work in progress have often been the highlights of our annual meetings.

Student involvement has been important, recognized since 1990 by the Critchfield Scholarship Awards that support outstanding student contributions to be given at other meetings. Support personnel, such as seed-orchard managers, nursery people, statistical analysts, etc., have been welcome members and participants. We have presented 10 short courses focusing on practical, theoretical, and high-tech topics in support of tree breeding and forest genetics.

We have often partnered with meetings of tree physiologists, ecologists, statisticians, and other tree-improvement organizations. Meeting venues have been in Merida, Yucatan; Prince George, British Columbia; Makaha, Hawaii; Fort Collins, Colorado; and lots of places in between. Unlike many scientific and professional meetings, at WFGA free exchange of ideas and information, as well as friendship and fellowship, have been the norm.

Perhaps our greatest disappointment has been the failure of academic forestry and natural resource management curricula to include forest genetics and tree improvement in the education of all their students of forestry and conservation. Perhaps in the next 50 years?

### Genetics of agronomic crops – a look at the past, present and future

**Richard Cronn** 

# USDA Forest Service, Pacific Northwest Research Station Corvallis, OR, 97330

Modern crops include only  $\sim 100$  species of the 200,000+ species described worldwide, yet they represent a stunning array of evolutionary, taxonomic and genetic diversity. Historical and contemporary trends in domestication and breeding of these diverse species are complex, but a number of common themes have emerged through comparative studies incorporating diverse sources of information, such as archaeological and molecular genetic evidence. In this talk, I review the past, present and future of crop genetics through the prism of a single crop, namely cotton. The general success of cotton as a fiber crop can be attributed to two general features common to many other important crops such as wheat, maize, soybean and potato. First, genetic redundancy introduced through recent polyploid formation opened the door to a greater level of genetic and developmental experimentation, and hastened the pace of crop improvement relative to diploid relatives. Second, improvement of fiber yield and quality was accelerated by a history of genomic exploration through hybridization, initially with closely related species, but more recently with distantly related species that differ in ploidy levels. Recent gains in cotton production are attributable to two key genetic factors, development of disease resistant cultivars, and transgenic insect protection. Future genetic improvement of cotton will depend on the continued introduction of novel variation, both through interspecific hybridization and transgenic technologies. Traits of interest in modern cotton breeding programs are mirrored by many other important crops, and include pathogen and herbivore resistance, improved drought and heat tolerance, increased tolerance of salinity, and possibly preserving productive hybrid crosses through novel processes such as apomixis.

#### A Forestry Perspective to Year 2020

R. C. Kellison

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The example has been set by the textile industry where failure to modernize their manufacturing plants has resulted in the production being shifted offshore, first to Latin America and then to Southeast Asia. Fast forward 20 years; the pulp and paper industry is on the same course. Since 1996, that industry has allowed capitalization to wane to about half that of depreciation and amortization. During that time, new pulp mills with attendant technology have been or are being The prognosis is that much of the pulp for built in South America and Southeast Asia. consumption in the United States will be imported. Numerous old pulp mills that have failed to meet air quality standards have been shuttered, and other ones will follow. Paper manufacturing will not be altered greatly because the U.S. market has greater demand for higher value-added products than other countries, with the EU being a distant second. A result of the decline in pulp production will be greatest in the South than in the Pacific Northwest because southern mills rely more on roundwood, and less on residual chips, than do their counterparts. That loss of market for pulpwood will dictate that the southern pine plantation forest will be geared more to sawtimber rotations than has been normal. Much of the roundwood that was destined for pulpwood will be increasingly used for engineered wood products (EWP), especially oriented strand board (OSB). New OSB mills with total capacity of more than 2.8 billion sq. ft. (3/8" basis) have been announced or are under construction in Georgia, Florida, Alabama and Louisiana. The new capacity is added to the 7.385 billion sq. ft. capacity that is already in production. The resultant sawtimber will replace a declining supply of lumber for U.S. consumption from Canada and it will limit imports from Latin America and New Zealand where the wood properties of constructiongrade lumber from plantation-grown trees do not meet U. S. building codes.

# **Comparison of Seedling versus Clonal Forestry in Queensland**

Mark J. Dieters

# The University of Queensland, School of Land and Food Sciences, Hartley Teakle Building, St. Lucia, QLD 4072, Australia

Clonal forestry with the interspecific hybrid between Pinus elliottii var. elliottii and P. caribaea var. hondurensis in Queensland (Australia) has been achieved operationally since 2002; involving the deployment of 4 to 5 million clonal plants per year. This hybrid has consistently demonstrated superior performance to both parental species across a range of sites on the coastal plain of subtropical south-east Queensland: delivering rapid growth, superior stem form, resistance to wind-damage, and excellent wood properties as well as broad adaptability to a range of site types. Queensland hybrid pine typically yields at least 50% additional wood production over P. elliottii, and P. caribaea var. hondurensis although fast growing has inferior wood properties for use as structural lumber for construction purposes (i.e. the predominant market for plantation-grown pine in south-east Queensland). Consequently, economic analyses have shown that significantly higher plant production costs can be justified in order to capture the benefits of hybrid pine. However, biological constraints (low numbers of viable seeds per cone, and maturation effects associated with reduced root initiation on stem cuttings as hedge age increases) make large-scale deployment of F<sub>1</sub> hybrid families as seedlings impossible and as cuttings impractical. Clonal forestry offers an opportunity to select for propagation traits, enabling utilization rates (i.e. ratio of shoots set as cuttings to the number of plants leaving the nursery gate) to be lifted to economically viable levels, and so making full-deployment of elite hybrid germplasm possible.

The focus of genetic improvement activities with hybrid pine in Queensland has recently been changed significantly. Previously, hybrid breeding activities focused on the identification of superior  $F_1$  hybrid families that could be either deployed as families, or screened for superior individuals that would subsequently be deployed as clones. However, genetic studies and simulations have clearly demonstrated that gain will be maximized by merging the two species into a synthetic hybrid, so that breeding activities now focus on the development of superior advanced generation ( $F_2$ ,  $F_3$ , ...) hybrid families. Advanced generation hybrids are significantly easier to produce than the initial  $F_1$  cross (50-80 seeds/cone vs. commonly <15 seeds/cone in the  $F_1$ ), making the operational deployment of full-sib hybrid families possible. I expect that over the next 3-5 years, as new (elite)  $F_2$  and  $F_3$  hybrid families are produced and evaluated, family forestry (possibly combined with limited vegetative propagation) will provide a viable alternative deployment strategy to clonal forestry, at a substantially reduced cost.

Clonal forestry is often viewed as the 'holy grail' of commercial forestry with plantation conifers, delivering rapidly growing, highly uniform stands. Any comparison of seedlings (taken here to be elite full-sib families) vs. tested clones as alternative deployment strategies, is both complex and subject to many assumptions. This paper will presents some advantages and disadvantages of clones and families as alternative deployment strategies for hybrid pine in Queensland.

# Candidate Gene-Quantitative Phenotype Associations for resistance against Fusiform Rust and Pitch Canker in Loblolly Pine

Elhan S. Ersoz (1,2), Santiago C. Gonzalez-Martinez (1,3), Gogce C. Kayihan (4), Geoff Gill (1), Garth Brown (1), Alison Morse (4), Dudley Huber (4), Tim White (4) John Davis (4), David Neale (1,2,5)

(1) Department of Plant Sciences, University of California, Davis, CA, 95616

(2) Graduate Group in Genetics, University of California, Davis, CA, 95616

(3) INIA- Forestry Research Institute, Madrid, SPAIN

(4) School of Forest Resources and Conservation, University of Florida, Gainesville, FL.

(5) Institute of Forest Genetics, Pacific South-West Research Station, US-Forestry Service, USDA.

Loblolly pine is the most commercially important forest tree species in the US. The fungal disease Pitch Canker, caused by the necrotrophic fungus *Fusarium circinatum*, is a necrotrophic pathogen that has been detected in loblolly pine plantations since 1974 where it causes extensive mortality associated with excessive pitch production of the host. Fusiform rust, caused by the biotrophic fungus Cronartium quercuum ssp. fusiforme, has been a major disease of southeastern conifer plantations since early 1920's. The challenge in developing disease resistant varieties, however, is that the genetic basis of stable resistance to both diseases is quantitative, rather than simple, gene for gene resistance common in other plant-pathogen systems. Our objective was to identify single nucleotide polymorphisms in candidate genes conferring resistance to these pathogens, which was tested for association with quantitative phenotypes. Candidate genes for disease resistance are categorized as: Positional Candidates, referring to candidate genes that are near QTLs for cell wall chemistry like pheylpropanoid pathway genes and cellulose synthase genes, Expression *Candidates*, referring to genes that are being identified by expression analyses (chitinases and myb and WD40 class transcription factors), Functional Candidates, referring to genes and regulatory regions whose roles in disease resistance have been identified in other plant systems through sequence homology to loblolly pine ESTs and bioinformatics. A total of 57 candidate loci have been screened first for identification of polymorphic locations that might lead to physiological changes in the disease resistance phenotype, and then selected SNPs have been inquired for their relevance to the variation observed in the phenotypes via association testing of these polymorphisms over an association population of 960 individuals. Progress to date on the successful associations made will be presented, two pathosystems will be compared and contrasted regarding allelic associations of the loci detected to be associated with resistance.

# Evidence of multiple glacial refugia for *Pinus contorta* inferred from a mtDNA minisatellite marker

Julie Godbout<sup>1</sup>, Aron Fazekas<sup>2</sup>, Craig Newton<sup>3</sup>, Francis C.H. Yeh<sup>4</sup>, and Jean Bousquet<sup>1</sup>

<sup>1</sup>Canada Research Chair in Forest and Environmental Genomics, Université Laval, Ste.Foy, Québec, Canada G1K 7P4

<sup>2</sup>Departement of Botany, University of Guelph, Guelph, Ontario, Canada N1G 2W1 <sup>3</sup> BC Research Inc., 3650 Wesbrook Mall, Vancouver, British Columbia, Canada V6S 2L2 <sup>4</sup>Department of Renewable Resources, University of Alberta, Edmonton, Alberta, Canada T6G

2P5

Lodgepole pine (*Pinus contorta*) is a hard pine with broad latitudinal distribution along the coast and inland across the Pacific Northwest. It is composed of up to four subspecies, including the widely distributed ssp. *latifolia* and the more restricted ssp. *contorta* along the coast. At the last glacial maximum, 18,000 years B.P., most of the range of lodgepole pine was covered by ice. It is likely that the current genetic structure of lodgepole pine has been shaped at least partly by historical events related to the last glaciation. In the Pinaceae, mtDNA is maternally inherited and dispersed through seeds. This mode of dispersion leads to reduced gene flow and higher population structure, as compared to paternally-inherited cpDNA or biparently-inherited nuclear DNA. Hence, maternal inheritance implies that the imprint of ancient genetic structures established at the time of glacial periods should remain for longer periods of time and that migration routes should be easier to track with mtDNA markers. We have used a mtDNA minisatellite-like marker discovered in the closely related jack pine (Pinus banksiana) to screen mtDNA haplotype diversity in 150 populations of lodgepole pine located across its natural range. Eighteen haplotypes were detected, which corresponded to different repeat numbers of a 32-nucleotides motif. A significant population subdivision was apparent, with at least three relatively homogeneous groups of populations presumably representative of three genetically distinct glacial populations: a first one along the coastal area in British Columbia, a second one in the interior of British Columbia, and a third one located at the junction between Alberta, Bristish Columbia, Idaho, and Montana. Zones of suture with increased mitotype diversity were also apparent between these three ancestral lineages.

# Identifying Candidate Genes Associated with Cold Hardiness in Coastal Douglas-fir Using DNA Microarrays

**Dana K. Howe<sup>1</sup>**, Amy M. Brunner<sup>1</sup>, Marilyn Cherry<sup>2</sup>, Konstantin V. Krutovsky<sup>3</sup>, David B. Neale<sup>3,4</sup>, and Glenn T. Howe<sup>1,2</sup>

<sup>1</sup>Department of Forest Science, Oregon State University, Corvallis, OR 97331 <sup>2</sup>Pacific Northwest Tree Improvement Research Cooperative, Corvallis, OR 97331 <sup>3</sup>Institute of Forest Genetics, Pacific Southwest Research Station, USDA Forest Service, Davis, CA 95616

<sup>4</sup>Department of Plant Sciences, University of California, Davis, CA 95616

Cold hardiness is an adaptive trait with considerable variation and importance in Douglas-fir (Pseudotsuga menziesii var. menziesii (Mirb.) Franco). We are working towards identifying genes associated with cold hardiness in coastal Douglas-fir using gene expression profiling. In the first year, we grew one-year-old seedlings outdoors in Corvallis, Oregon, and harvested them in the fall, winter, and spring (from bud set to bud burst) for RNA extraction. RNA samples were pooled and used to make three cDNA libraries (cold acclimating, maximum cold hardiness, and cold deacclimating). A fourth cDNA library was constructed from actively growing seedlings. In the second year, we studied cold hardiness in seedlings from two Douglas-fir populations. The Coos Bay (CB) population is from a coastal, low elevation site in Oregon, whereas the Yakima (YK) population is from an inland, high elevation site in Washington. RNA extractions and artificial freezing tests were conducted on the seedlings from these two populations at various times during cold acclimation, maximum cold hardiness, and cold deacclimation. Damage to needles, stems, and buds was assessed using visual inspection, and needle damage was also measured using electrolyte leakage. The cold hardiness of the two populations was significantly different for all tissues. For example, in late October, differences in predicted damage at -7°C were as large as 48% for needles (i.e. CB=50% and YK=2%), 47% for stems (i.e. CB=50% and YK=3%), and 35% for buds (i.e. CB=36% and YK=0%). Correlations between needle, stem, and bud damage were stronger in the fall than in the spring. RNA isolated from these two populations will be used to examine differences in gene expression associated with cold hardiness. Our four cDNA libraries have produced >20,000 ESTs that will be used to construct custom oligonucleotide microarrays for gene expression profiling. Genes that show differential expression (1) throughout the winter and (2) between populations will be classified as cold hardiness candidate genes and integrated into a larger association study of adaptive traits in Douglas-fir populations from Oregon and Washington.

## Molecular Evolutionary Rates Indicate A Recent And Rapid Diversification Of Modern Pine Lineages

Ann Willyard<sup>1</sup>, John Syring<sup>1</sup>, David S. Gernandt<sup>2</sup>, Richard Cronn<sup>3</sup>, and Aaron Liston<sup>1</sup> <sup>1</sup>Department of Botany and Plant Pathology, Oregon State University, Corvallis, Oregon 97331 USA <sup>2</sup>Centro de Investigaciones Biológicas, Universidad Autonoma del Estado de Hidalgo, Apartado Postal 1-69

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<sup>3</sup>Pacific Northwest Research Station, USDA Forest Service, 3200 SW Jefferson Way, Corvallis, Oregon 97331 USA

Previous estimates of pine evolutionary rates have varied widely, from comparable to woody angiosperm rates to among the slowest ever reported for any plant. This discrepancy in absolute rates is intimately entangled with age estimates, which in turn depend on pairing a fossil with a divergence between species. This pairing is not trivial, and requires a phylogenetic framework on which to place the fossil. Additionally, relative rates based on multiple, unlinked genes provide a more useful estimate than those based on a single gene. To create a benchmark for estimating rates and ages of the pine lineages, we used substitution rates for 12 nuclear (including 11 lowcopy) and four chloroplast genes (ca. 11,000 nucleotides) for a rooted quartet of Pinus species representing each of the major lineages. Two putatively non-orthologous nuclear loci and the nrITS were excluded from subsequent analyses. A robust sample of noncoding regions demonstrates that *Pinus* nuclear and chloroplast silent rates (third codons plus noncoding) are consistently slower than synonymous rates, a phenomenon that has been reported in other plants. Twelve loci showed internal rate constancy, even though there was substantial rate heterogeneity between genes. We used the silent sites from these loci to generate a nuclear and a chloroplast clock-enforced phylogeny. Although some have suggested that the higher level of divergence in subg. Strobus indicates an older lineage, all of the earliest currently accepted pine fossils (including the Cretaceous P. belgica) have been attributed to subg. Pinus. Hence, we calibrated the chronogram where the two subgenera branch, using the age of the oldest subg. Strobus fossils. Bracketing the subgenus split between 60 and 45 mya yields a mean absolute rate estimate of 1.10 x  $10^{-9}$  substitutions per site per year at nuclear and 0.36 x  $10^{-9}$  at chloroplast silent sites. These rates are comparable to those reported for long-lived angiosperm taxa. Projections using proportional branch lengths on the nuclear chronogram place the divergence of sect. Trifoliae from sect. Pinus at 21 to 16 mya, sect. Quinquefoliae from sect. Parrya at 34 to 25 mya, and the origin of the genus at 136 to 102 mya. Chloroplast rates were ca. three fold slower, and placed the origin of the genus at 117 to 89 mya. Our chronograms reveal unrealistically old estimates for the divergence of *Pinus* from *Picea* if *P. belgica* is used to calibrate any internal node, consistent with its position at the stem rather than the crown of the Pinus tree. To address the question of what happens when more species are added, the quartet chronograms were compared with a non-clocklike 12-species, four-gene phylogeny using a Penalized Likelihood method. The age range for hard pine section branching was comparable, but the soft pine section divergence occurs earlier (34 to 26 mya). We confirm previous reports of lower levels of variation within subg. *Pinus* and suggest that the paradoxical lack of Cretaceous subg. *Strobus* fossils may be explained by the earliest pines being "subg. Pinus-like", while most extant species of subg. Pinus arose from a relatively more recent, rapid radiation.

# Genetic diversity in whitebark pine is affected by white pine blister rust infection

### Andrew D. Bower and Sally N. Aitken

University of British Columbia

We have examined trends in heterozygosity to investigate the effects of inbreeding and the introduced disease white pine blister rust (caused by the fungus *Cronartium ribicola*) on genetic diversity in whitebark pine (*Pinus albicaulis* Engelm.). Isozymes were used to estimate the level of genetic diversity in three age cohorts (seedlings, young, and mature trees) from 14 sites with varying levels of rust infection in British Columbia, Idaho, and Montana. Values of observed and expected heterozygosity (H<sub>o</sub> and H<sub>e</sub>) and fixation index ( $F = 1 - H_o/H_e$ ) were compared among cohorts, and by cohort across sites. A significant heterozygote deficiency was detected at all ages, indicating a strong effect of inbreeding. When the level of rust infection is low, F decreases and H<sub>o</sub> increases with age, however, when rust infection is high, F increases and H<sub>o</sub> decreases in the mature cohort, relative to the young cohort. Trends in F and H<sub>o</sub> with increasing level of rust infected trees than for infected trees sampled. Our data suggests that more homozygous individuals may have a selective advantage in the face of blister rust possibly due to the increased expression of recessive resistance genes due to inbreeding, however, the number of sites included in the study was not sufficient to yield conclusive results.

# Improving Breeding Trial Statistical Analysis through Post-hoc Blocking

# **S.A. Gezan**<sup>1</sup>, T.L. White<sup>2</sup>, and D.A. Huber<sup>3</sup>

<sup>1</sup>Graduate Student, <sup>2</sup>Professor, <sup>3</sup>Research Associate SFRC, IFAS, University of Florida, Gainesville, FL, USA.

Experimental sites in forestry tend to have high environmental variability, which is usually in the form of patches, trends or both. The use of incomplete block designs in comparison to the randomized complete block design can provide considerable improvement in the precision of the estimation of genetic parameters and ranking of genetic entities. Due to the unknown pattern of variability on a field experiment, it is usually difficult to define an adequate block orientation and size. Hence, it is difficult to get the maximum improvement from using incomplete blocks. Blocking can also be implemented in the analysis stage, a technique that is known as post-hoc blocking consists of superimposing a one- or two-way blocking design and running the analysis as if these effects were present in the original design.

The goal of this study is to quantify the effects of *post-hoc* blocking. This is achieved through simulation of clonal trials for three different error surfaces (only patches, only gradients and both together). In the first stage several experimental designs were simulated and analyzed. Later for a randomized complete block design, incomplete block (IB) designs with blocks of different sizes were superimposed and reanalyzed. A row-column (R-C) design was also superimposed. In the later stage, the precisions of these different alternatives for *post-hoc* blocking together with several strategies to select models were compared. The statistic to discriminate the best model was based on average individual broad-sense heritability and the correlation between true and predicted clonal values (CORR). Also, more flexible mixed linear models were fitted for different IB and R-C variance components by replicate. Finally, some statistical justifications for implementing *post-hoc* techniques were discussed together in relation to spatial analysis of experiments, together with their relationship to blocking and randomization of treatments.

Considerable improvements are obtained with *post-hoc* blocking with average heritabilities as good as when the blocking was defined in the design stage. For IB designs, when average CORR values were compared, the best results corresponded to the design with the smallest incomplete block (*i.e.* 2 plots/block). For this study no reduction in genetic variance was found for smaller incomplete blocks. Once, all models were compared, the R-C design was the best due to the extra random effects in its linear model. And also, as expected, better statistics were found in mixed models that considered by replicate blocking variance components.

# Phenotypic and Genetic Correlations of Commercially Important Wood Quality Traits in Coastal Douglas-fir

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In tree breeding activities, the ability to accurately predict the genetic response of commercially important wood and fibre traits and growth rates through selection can significantly improve tree breeding efforts. In order to achieve such confidence, comprehensive knowledge of the relationships between easy to measure (volume) and commercially significant traits (density, fibre properties, etc.) is paramount. We explored the genetic control and relationship between growth and yield parameters (height, diameter and volume) and several wood quality attributes, including fibre length, cell wall thickness, microfibril angle, wood density (both earlywood and latewood) and wood chemistry (lignin and carbohydrate content) using broad-sense heritability (H<sup>2</sup>), and phenotypic and genetic correlations for coastal Douglas-fir (Pseudotsuga menziesii (Mirb.) Franco). This study employed 15 full-sib families replicated on four sites in Southwestern British Columbia from the BC Ministry of Forests Douglas-fir second generation progeny test program. The fifteen families and four sites were chosen to maximize variation in the sample population. Site and family variation were significant ( $\alpha = 0.05$ ) for all growth and yield and wood quality traits, and there was a slight family by site interaction for volume. Broad-sense heritability estimates varied from 0.23 to 0.30 for growth and yield traits, 0.10 to 0.18 for fibre traits, and 0.21 to 0.54 for wood density traits. Typically, growth and yield traits show significant positive phenotypic correlations with fibre traits, while both fibre and growth and yield traits show negative correlations with wood density. Genetic correlations follow the same general trends as phenotypic correlations with a few exceptions. These data combined with QTL mapping technology will be used to further explore the Douglas-fir genome and interactions of important wood quality traits.

# Genetic differentiation of ponderosa pine and a dominant understory grass in northern Arizona: implications for restoration

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Genetic diversity is the raw material for biodiversity, and its understanding provides important information for forest conservation and restoration. The association between environmental heterogeneity and genetic variation has been widely studied. However, little is known regarding possible relationships between genetic variation in forest canopy and understory species. Understanding these relationships could provide information for delineation of seed zones and identification of biodiversity hotspots for conservation of understory species. We hypothesized that there would be a correlation between genetic variation of ponderosa pine (Pinus ponderosa) canopy trees and bottlebrush squirreltail (Elvmus elvmoides) from ponderosa pine forests of northern Arizona, and a correlation between plants and environmental conditions. Allozyme data shows these two species exhibit different genetic structures which are likely due to the fact they have different life histories and mating systems. Ponderosa pine shows relatively high genetic variation within populations and low variation among populations, while bottlebrush squirreltail shows low genetic variation within populations and high variation among populations. Gene flow between populations of bottlebrush squirreltail is predicted to be much lower than in ponderosa pine, and genetic variations of ponderosa pine and bottlebrush squirreltail are affected by environmental conditions. Based on these findings, we will make recommendations for seed transfer and conservation genetics of ponderosa pine and bottlebrush squirreltail.

# **Development of Genomic Tools for Poplar**

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The recent sequencing of the poplar genome has paved the way to develop a range of genomic tools for understanding tree biology and jumpstarting tree improvement. Due to high level of putative functional redundancy in the gene families of Populus trichocarpa, sequencing of the complete P.trichocarpa genome was a pre-requisite for the comprehensive characterization of functional transcripts and their regulatory elements of this species. Through a collaboration with ORNL we have developed a microarray containing all predicted Populus trichocarpa gene models. The microarray represents 65,966 individual sequences including, nuclear, organelle and microRNA precursor genes. We will present the use of this array in global expression profiling of a range of poplar tissue types from different developmental stages. Comparison of differentiating xylem and phloem during late summer showed close to 4000 differentially expressed genes which is a significantly higher number than what has been reported thus far using cDNA microarrays. Similar gene expression data using the whole-genome array would be invaluable for elucidating poplar growth and development associated gene regulatory networks such as those controlling wood development.

# A conserved CC-NBS-LRR resistance gene in sugar pine

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We have isolated a 2926 base pair cDNA (Pl-RGC-CC 1) from sugar pine (P. lambertiana) that predicts a 777 amino acid peptide having high similarity to Arabidopsis CC-NBS-LRR type resistance genes such as ADR1, RSP2 and RPM1. The primary objective of this research was to isolate the nucleotide sequence corresponding to the white pine blister rust (WPBR) major gene of resistance (Cr1) that was identified by Kinloch (1992). Cr1 has been genetically mapped in several segregating sugar pine open-pollinated families using rescued megagametophytes (Harkins et al. 1998). PCR primers designed from Pl RGC-CC 1 were used to amplify nucleotide sequences from megagametophytes from our sugar pine mapping populations for obtaining single nucleotide polymorphisms (SNPs) segregation data. Surprisingly, not a single SNP was detected at this locus in either of our mapping populations; a result which prompted us to examine a small population of unrelated sugar pine trees and several western white pine (P. monticola) trees. Three SNPs (singletons) were found in the 3' non-coding region in the panel of unrelated sugar pine. In western white pine, several SNPs were detected in the 3' non-coding region, but as in sugar pine, none were found in the 2333 bp coding region. The low level of nucleotide diversity ( $\pi$ ) at the *Pl* RGC-CC 1 locus is anomalous when compared to other functional genes studied in conifers. indicating that it is either under direct negative selection or possibly under the influence of a genetic selective sweep. Its strong similarity to coil-coil sub-class R genes annotated in GenBank only hints at its possible function.

# Mapping Adaptive Traits in Douglas-fir using Association Genetics

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Through the accumulation of studies over a number of years, we have begun to understand and identify the genetic components of adaptation in conifer species. In Douglas-fir, our work has identified loci controlling several quantitatively inherited traits one being tolerance to frost and low temperatures, an important character for the timber industry. By mining cDNA libraries constructed in Douglas-fir, Loblolly pine and Norway spruce for genes known to contribute to the physiological processes involved in cold hardiness, we have identified the Douglas-Fir homologues of these genes. We are using association genetics to ascertain correlations between DNA polymorphisms that are being identified in these genes and phenotypic cold tolerance data collected in c.900 unrelated individuals from heterogenous environments across Washington and Oregon. I will report on the progress of SNP identification in candidate genes for cold tolerance and their genotyping in the association population.

# Polymix Breeding With Paternity Analysis In *Populus*: A Test For Differential Reproductive Success (DRS) Among Pollen Donors

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Polymix breeding with paternity analysis (PMX/WPA) of progeny has been proposed as an alternative to traditional full-sib breeding and testing schemes for out-crossing species. In short, PMX/WPA requires the application of pollen mixes of many male parents in controlled crosses, followed by paternity analysis of progeny using molecular markers. The approach offers good estimates of breeding values of parents and produces progeny populations with superior gain potential for forward selection. Other advantages of this approach include simpler and easier (thus less expensive) breeding and testing programs, greater flexibility for inbreeding control, and convenient tie-in with other molecular genetic applications such as clonal fingerprinting and MAS. To capture the full benefit of PMX/WPA, DRS of pollen parents must be inconsequentially low. To test the hypothesis that all pollen parents in a polymix are equally represented in a progeny pool, we created seven,  $F_1$  hybrid crosses, between eastern cottonwood ( $\mathcal{Q}$ ; *Populus deltoides*) and black poplar ( $\mathcal{O}$ ; *Populus nigra*) using a 16 parent polymix. Parents and progeny (~50 per maternal parent) were genotyped using an array of seven SSR markers selected for fidelity and information content. Of 359 progeny, 348 (96.9%) were unambiguously assigned to a single father, 10 could be identified as having one of two fathers, and one progeny could have been fathered by one of three parents. With a few exceptions, all 16 parents contributed to the progeny pool of all seven maternal parents, though contribution varied significantly among pollen parents. Pollen vigor (germination percent) was not correlated with paternal success. In many instances, SSR markers displayed differential amplification between parental species in hybrid progeny. Results will be discussed relative to the operational utility of PMX/WPA for poplar.

# Parameter Estimation for Stochastic Simulation of Multiple Traits over More Than One Generation

# Jen Myszewski<sup>1</sup>, Tom Byram<sup>2</sup>, and Floyd Bridgwater<sup>3</sup>

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In order for a specialty tree improvement program to be successful, selection indices that weight multiple traits appropriately and allow the breeder to select for a desirable aggregate genotype must be developed and tested. Given the long generation time of most pine species, empirical tests of breeding and selection alternatives are impractical so breeders rely on stochastic simulation to estimate the effects of different strategies. Stochastic simulation uses random normal deviates to generate variances about the mean. When more than one trait is involved, correlated standard normal deviates must be used to account for the correlations between traits. This poses a problem when simulations are conducted over multiple generations. If a population is limited in size, is under selection, and/or is suffering from inbreeding depression as occur in a tree breeding program, correlations between traits change from generation to generation. Thus, correlations must be predicted each generation before variances about the means of each trait can be estimated. For this to occur, it is necessary to examine how the additive genetic (co)variances and the dominance (co)variances are affected by departures from Hardy-Weinberg equilibrium.

Tuesday PM - NAQFGG

# Efficiency of Spatial Analysis of First-Generation Coastal Douglas-fir Progeny Trials in the US Pacific Northwest

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Nearly 1,000 first-generation progeny trials were established in the Pacific Northwest between 1967 and 1993, programs now associated with the Northwest Tree Improvement Cooperative. These trials typically had 200-300 parents tested as open-pollinated families collected from the parent tree growing in the wild, and planted on 6-12 locations. Most trials were in "Reps-In-Sets" designs where all replicates of a single set of families planted together; with the rest established as "Sets-In-Reps" where each replicate included all the sets of families. Although efforts were taken to minimize micro-environmental variation, spatial homogeneity seldom occurs within replicates in forestry experiments. With increasing computational power and new spatial analysis techniques available, it may be possible to get greater efficiency and genetic gain in forest tree improvement programs at minimal added cost.

This paper reports an evaluation of efficiency of spatial analysis using autoregressive models relative to our current "Reps-In-Sets" or "Sets-In-Reps" analyses (base models), for 139 first-generation coastal Douglas-fir progeny trials from 18 different breeding zones in western Oregon and western Washington. Measurements of tree height (*HT*) were made at ages 5, 10, and 15 years from seed for most of the trials. Breast-height diameter (*DBH*) at ages of 10, 15, and 20 years were available for 15~82 sites, and stem volume (*VOL*) was calculated approximately as  $HT \times DBH^2$  when both *HT* and *DBH* were available.

We found that 500 out of 506 trial × trait combinations had significant spatial variation based on likelihood ratio test. Positive spatial correlations among neighbors were found in all cases ( $\bar{\rho} \approx 0.8$ ). Spatial analyses using AR1 × AR1 models on average reduced the residual variance by 13~23% for *DBH* and 27~33% for *HT*. The resulting change in heritability after fitting spatial models varied among sites and traits with average increases from 0.20 to 0.24 for *DBH*, from 0.22 to 0.30 for *HT*, and from 0.32 to 0.37 for *VOL*. Spatial analysis also increased the accuracy of breeding value estimation (i.e. correlation between true and predicted genetic values) by 9~11% in most cases. Increases in relative genetic gain of changing from base models to spatial models were mostly less than 3% for backward selection (selecting the top 20% parents) and less than 5% for forward selection (selecting the best offspring from each of the 20% families). Sites with "Sets-In-Reps" design showed relatively higher spatial autocorrelation than sites with "Reps-In-Sets" design (0.88 vs. 0.79). Spatial autocorrelation was slightly higher in *DBH* (0.88) than in *HT* (0.80), and higher at age-10 than at age-5 or age-15. The pattern was rather random and patchy geographically and did not show significant longitudinal, latitudinal, or altitudinal trends (i.e. between breeding zones within the region).

Results indicated that adjustment for micro-environmental heterogeneity in progeny tests in the Pacific Northwest is desirable to improve estimates of genetic parameters and to maximize genetic gains from selection. It is therefore likely that spatial adjustment will be used in analyzing the second-generation data which is already coming available.

# **Deployment of Genetically Improved Material: Anything new lately?**

# Alvin Yanchuk and John Russell

## Research Branch, B.C. Forest Service

Although clonal and family 'block' forestry has been discussed over the last 50 years, implementation has been rather small and benefits still not very convincing in many localities. This may be due (independent of the economics of tissue culture production) to; i) large withinclone (C-effects) and family variation, ii) large environmental variations, iii) many clones/parents in use, and most importantly, iv) most block plantations that have been established are not old enough yet or planted on enough acreage to see the economic benefits.

Until more data are available, we will still largely be relying on theoretical analyses around deployment for factoring in the risk of loss of a stand to some future pest or disease. For this we still must rely on the original work of Libby (1981) and other supporting papers by Roberds, Bishir, and Namkoong. These models were largely for single-gene resistance models for clonal genotypes, and we are all aware of the important finding(s), that after 30 clones not much 'safety' is added. We have recently been building upon this body of research using the Tree and Stand Simulator (TASS) with a population dynamics model for a tree pest (spruce terminal weevil) to examine several other assumptions around deployment. The model does not induce mortality *per se*, other than that generated by competition, but includes three resistance mechanisms that behave in a quantitative fashion. The results first suggest that mixtures of clones (or genotypes) are preferred, and second, that around 18 is a safe 'optimum' number, although the range is quite large. Again, using more than 30 seems to provide little advantage and less than six seems risky.

For other pest or herbivore interaction, such as deer resistance in western redcedar, blister rust, or weevil resistance, investigations into the factors involved in reduced feeding and damage will force us to consider deployment with 'known threats' (in addition to future unknown biotic and abiotic threats). With additional factors such as climate change, deployment considerations are becoming more complex and will likely have to target 'stand resolve' rather than optimum economic returns.

### Growth vs. Defense Adaptations: Is there a trade-off?

John N King – BC Forest Service, Victoria BC Barry Jaquish – BC Forest Service, Vernon BC Randy Johnson - USDA Forest Service, Corvallis OR Sara Lipow - Oregon Department of Forestry, Salem OR

A commonly held view is that the allocation of resources by plants to chemical and structural defenses decreases growth by diverting resources from the production of leaf area and other vegetative structures. This is seen as a trade-off between investing in defensive mechanisms vs using plant resources for growth. We look at three cases from the Pacific Northwest that includes Douglas-fir and Swiss needle cast resistance, Douglas-fir and armillaria root-rot resistance and both interior and Sitka spruce resistance to tip weevil. In all of these cases and in an extensive look at the literature we could find no evidence that this trade-off is the rule. In fact it often appears that families with good defensive resources are just as likely to be inherently fast growing. We look at the nature of genetic and phenotypic correlations and how this is factored in this potential trade-off.

# The Seed Transfer Impact Calculator

Greg O'Neill B.C. Ministry of Forests

The Seed Transfer Impact Calculator (STIC) is an Excel spreadsheet program that estimates forest plantation productivity impacts associated with seed transfer in BC. Transfer curves for latitude, longitude and elevation at provenance test locations are used to develop site-specific transfer curves for plantations at any location in the province for three species. Inputs required include the coordinates of the seed source and plantation. Outputs include percent productivity and volume forfeited at rotation. Development and limitations of the STIC will be discussed.

Thursday AM - Concurrent Session III

# Impacts of climate change and potential adaptation strategies through genetic resource management in British Columbia

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Climate change observed during the past decades may have already affected forest ecosystems in British Columbia. Epidemics of forest pest and increased fire disturbance may in part be attributable to climate warming. If these trends continue as predicted by global circulation models, the forestry community will have to implement extensive changes to resource management policy, forestry operations, and conservation planning.

Adaptation strategies may include (1) changes to the protected area network for more effective conservation of genetic resources, (2) matching genotypes to new environments through modified seed zones, and (3) new tree breeding strategies aiming at a better adaptive match to environments or greater variance in deployment populations:

(1) We employ high-resolution climate modeling and bioclimate-envelope modeling to research potential impacts and adaptation strategies. Based on predicted changes in species habitats we identify tree species and populations at risk due to climate change and set priorities for gene conservation efforts. The effectiveness of selecting protected areas with a higher elevation range is tested, making various assumptions about species to migrate and adapt.

(2) By re-defining seed zones climatically, we match planting stock from current seed orchards to predicted future environments. Interior seed orchard target environments are predicted to shift northward at a rate of approximately 50-100 km per decade. Seed orchard target environments currently covering the southeast of British Columbia may provide suitable planting material for an extended portion of British Columbia in the future, whereas demand for other orchards seed may rapidly decline in B.C.

(3) In two case studies of experimental tree breeding with red alder and Douglas-fir we evaluate two adaptation strategies that may be implemented in tree breeding programs: breeding for increased genetic variance in deployment populations versus divergent selection of adaptive traits in multiple breeding populations to match predicted environments.

# Clonal Forestry vs. Seedling-Based Tree Improvement for Coastal Douglas-fir in the US Pacific Northwest

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Clonal forestry will yield the maximum genetic gain in a forest tree improvement program. Very successful clonal forestry programs are in place for some species, especially eucalypts, and there is strong interest in applying clonal forestry for some important conifer species.

The gains and costs of an aggressive seedling-based program were compared with a clonal forestry program. To be relevant to the PNW, we assumed the first generation of testing was complete and elite first-generation parents were available. Two scenarios were compared (A) initiate 200 clones, test them, select and deploy 20 top-performing clones; (B) make and test 220 crosses in the field, select top forward selections, establish and rogue an orchard and deploy control mass pollinated (CMP) seed. We then assumed this program served a landbase of 200,000 acres, with a rotation of 40 years, replanting 5,000 acres a year with 400 trees per acre.

Gains from the clonal testing option were greater than for the seedling-based option: 13.32 units vs. 11.86 units (1.13 times as much at the plateau) and were delivered faster (gain differential was 1.41: 1 at one point). Assuming \$0.03 seed cost for CMP seed, the seed cost over 20 years + the testing cost was estimated at \$1,465,000 (about \$0.037 per tree planted). Assuming \$0.50 propagating cost for clonal propagules, the propagule cost over 20 years + the testing cost was estimated at \$20,271,250 (about \$0.51 per tree planted). Assuming \$0.25 propagating cost for clonal propagule cost over 20 years + the testing cost was estimated at \$10,271,250 (about \$0.26 per tree planted). It therefore appears that the added gains from clonal forestry are obtained at a much higher cost; unless propagation costs are brought down significantly, seedling-based tree improvement remains a very profitable option.

We feel that clonal forestry will need to operate by the same guidelines as seed-based testing programs. For example, if we test a cross on five sites to age 12 for seedling-based testing prior to making final selection decisions, the same should apply to clonal testing as well. It would be reasonable, however, to test a clone with fewer ramets per site than a full-sib cross. Other issues to consider regarding implementing clonal forestry in the PNW include (A) whether clonal forestry will attract unwanted attention and opposition from those dubious of genetic improvement in forest plantations and (B) ownership and rights to the clonally tested germplasm.

A benefit of clonal forestry (uniformity of log quality and wood properties) will probably accrue only to integrated companies which mill the wood they grow. Most forest growers in the PNW sell logs and are unlikely to be paid more for improved uniformity. Some factors which make clonal forestry successful and profitable in some species (ease of propagation, low labor costs, rotations as short as 10 years, ability to select clones reliably as early as 2 years) do not apply to Douglas-fir in the PNW.

# Douglas-fir Tree Improvement in the British Columbia Interior: Results and opportunities

Barry Jaquish

# Research Branch, B.C. Ministry of Forests Kalamalka Forestry Centre, Vernon, BC.

Interior Douglas-fir (Pseudotsuga menziesii var. glauca [Beissn.] Franco) forms a major component of the rich coniferous forests in B.C.'s south-central Interior. The species is ecologically complex and its abundant intra-specific genetic variation is structured largely along moisture and elevational gradients. In 2004, Interior Douglas-fir represented about six percent of the total timber harvest in B.C. (66 million m<sup>3</sup>), and in spring 2005 over 15 million seedlings were planted on harvested lands. Interior Douglas-fir tree improvement began in 1982 and focuses mainly on the productive lands in the moist Interior Cedar Hemlock (ICH) biogeoclimatic zone. The first cycle of breeding was centred on multiple breeding populations from nine breeding zones, recurrent selection for general combining ability, wind-pollinated progeny testing, and delayed 1.5 generation seed orchards. The traits targeted for improvement relate to tree size and form. Since 1982, over 1,700 parent trees from the nine breeding zones have been selected, grafted, and established in breeding orchards and clone banks near Vernon. Between 1984 and 1992, wind-pollinated families from 1,661 of these parent trees were established in progeny tests on 39 sites. In the early 1990's, the original nine breeding zones were reduced to six and seed orchards were established for each zone in the north Okanagan Valley. These seed orchards are beginning to produce seed and the first collectable cone crops will be produced this year. Secondgeneration crossing is focused largely on the Nelson seed planning zone and is about 70 percent complete. Ten-year progeny test results will be presented and opportunities for increasing genetic gain through the use of parents from the BC submaritime zone and inter-varietal hybridization will be discussed.

# **Recent Changes in the Genetic Improvement of Conifers in Queensland**

## Mark J. Dieters

# The University of Queensland, School of Land and Food Sciences, Hartley Teakle Building, St. Lucia, QLD 4072, Australia

Plantation forestry with conifers in Queensland can be subdivided into three groups: *Pinus elliottii* var. *elliottii*  $\times$  *P. caribaea* var. *hondurensis* hybrid pine on near-coastal sites of southeast Queensland; tropical pines (primarily *P. caribaea* var. *hondurensis*) in north Queensland; and the indigenous conifer *Araucaria cunninghamii* on high-fertility and lower rainfall sites of southeast Queensland. Large geographical and climatic ranges, mean that we must utilize more than one species/taxa to maximise productivity in forest plantations throughout the State. Nevertheless, on a world-scale our plantation estate is small (approximately 130,000 ha of hybrid pine, 25,000 ha of tropical pine and 45,000ha of *Aracuaria*), and it is becoming increasingly important to maximize returns from investments in tree improvement. Recent changes to the genetic improvement of conifers will be discussed for each of the three major taxa used commercially in Queensland.

The genetic improvement of *P. elliottii*, *P. caribaea* var. *hondurensis* and their hybrid in Queensland has relied primarily on genetic improvement within the pure species, and intensive testing and selection of elite  $F_1$  hybrid families. A new breeding strategy has recently (in early 2005) been approved, which seeks to merge the two species into a stabilized, synthetic-hybrid rather than continued breeding and selection within the two pure species. Results of research activities that support this radical change will be outlined.

The changes adopted for the breeding of hybrid pine, now mean that *P. caribaea* will now only be required for deployment as a pure-species across a small plantation estate (approx. 25,000ha), chiefly in north Queensland. Breeding activities must therefore provide improved material for reforestation in these regions at a low cost. The wide variety of sites where tropical pines may be planted and some uncertainty regarding what will be the best taxon, requires a diverse range of germplasm be maintained for the future. Solutions adopted to balance resource limitations and plantation requirements for adapted, genetically improved planting stock will be outlined.

*Araucaria cunninghamii* is a relatively long-rotation crop (50-60 years) compared to hybrid or tropical pine (around 25 years) in Queensland. Further, the reproductive biology of this species means that generation intervals (15-20 years) and the time-lag from establishment of clonal seed orchards to seed production (usually 10 years or more) are considerably longer than the pines. This means that benefits from tree breeding activities are unlikely to be realized for around 80-90 years. Consequently, tree improvement activities with *A. curninghamii* now focus on identification of superior (growth, form and wood-quality) parents that can incorporated rapidly into an operation control-crossing program to produce full-sib families for deployment in plantations.

Thursday AM - Concurrent Session IV

# Genetic structure of core and peripheral populations: Implications for research and *ex situ* conservation

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The majority of studies of population genetic structure are conducted using samples collected from continuous populations in the core of species ranges. These types of populations are usually typical of regions where species have high economic importance and therefore receive more attention. From these studies, we have learned that populations of most conifers have low inbreeding levels, little within-population spatial genetic structure, high levels of gene flow, and many pollen parents contributing to pollen clouds. In our studies of core and peripheral populations of Sitka spruce (Picea sitchensis) we have found that while these general characteristics describe the population genetics of core populations, peripheral and disjunct populations are markedly different. Peripheral populations are more inbred, have strong spatial genetic structure and have few pollen parents contributing to open-pollinated progeny of individual parents. Despite higher inbreeding levels, disjunct peripheral populations may have a higher average degree of fitness than continuous peripheral continuous due to a lack of gene flow allowing more effective selection. Standard sampling strategies for population genetic studies may underestimate diversity in peripheral populations due to strong spatial genetic structure. For ex situ gene conservation collections, seed should be sampled from more individuals spread over a wider area in peripheral than in core populations to capture the same proportion of standing genetic variation.

Thursday AM - Concurrent Session IV

# Early Allocation of Resources to Shoots Versus Roots of Geographically Diverse Populations of Western Hemlock Selected for Improved Juvenile Height Growth

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Selecting trees for better stem growth traits raises concerns for consequences this might have on root development and resulting wind firmness. As well, nurserymen recognize different growth rhythms for trees from different latitudes, which might also have an impact on the readiness of stock for the field. In general, as trees come to an age where roots are several centimetres in diameter their growth can respond to wind stress, but before this action can ameliorate the problem, juvenile plants may be susceptible to damage. Apprehension on this account was raised when some of the largest and most exposed trees in progeny trials of F-1 hemlock were observed to be leaning. Root/shoot ratios for poly-crossed best height growth families from BC and Washington are contrasted to wood's run standards for container grown seedlings, as well as potted one and two year olds.

# Geographic Variation for Height and Diameter Among Black Spruce (*Picea mariana*) Provenances in Alberta

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Black spruce (*Picea mariana* (Mill) B.S.P) is a widely distributed tree species in the boreal region of North America, which is mainly used for pulp and timber production. Between 1989 and 1990 the Alberta Tree Improvement and Seed Centre established seven field provenance trials of black spruce to study the amount and pattern of its geographic variability, provenance  $\times$  site interaction and develop seed transfer guidelines for Alberta. The 22 provenances involved originated in Alberta between 52015' and 59030' N, 111028' and 119042' W and 244-1276 metre above sea level. Test sites are located between 52037' and 59008' N, 112012' and 117034' W and 370-990 metre above sea level. Variation among provenances for 15-year height (H15) and diameter at breast height (D15) were generally low. Across sites, the range of provenance means was 2.2 -3.1m (H15) and 3.4 - 3.9cm (D15). On individual sites, the variance among provenances was 1.0 - 1009.1% (H15) and 0.5 - 8.1% (D15) of the provenance phenotypic variance. When data for all sites were pooled, the provenance variance was significant (P = 0.05) accounting for 2.7% (H15) and 3.1% (D15) of the provenance phenotypic variance. The provenance  $\times$  site interaction was also significant (P = 0.05) accounting for 1.9% (H15) and 2.5% (D15) of the provenance phenotypic variance. The interaction variance was 72.4% (H15) and 81.9% (D15) of the between-provenance variance. This suggests that population differentiation for early growth in the sampled Alberta region is low and that genotype  $\times$  environment interaction is substantial and should be considered in planning for seed source transfer for reforestation. At four of the seven sites, height (r = 0.45 to 0.60) and diameter (r = 0.51 to 0.69) growth were significantly (P < 0.05) linearly related to mean annual temperature at the place of seed origin. Only at one site was height (r = 0.68) and diameter (r = 0.77) linearly related to mean annual precipitation at the place of seed origin. The results show that provenances from the cooler and drier northern Alberta had lower growth potential than those from the warmer and moister central and southern Alberta. In particular, results from these provenance trials show that temperature rather than precipitation is the main determinant of variability among black spruce populations in Alberta.

# Full-Sib Analysis of Pollen Flow and Pollen Structure in Guanacaste (*Enterolobium cyclocarpum*) in the Costa Rican Dry Tropical Forest

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Patterns and rates of gene movement across the landscape are key factors in the development of genetic structure under both natural and human-influenced conditions. Conservation biologists and forest geneticists have become interested in how landscape features (population densities and propagule dispersion, degree of population fragmentation and isolation, different habitat types) influence rates and patterns of gene flow across the landscape. Increasingly, we need to understand how heterogeneous spatial arrays and other landscape features influence patterns of gene flow.

We here elucidate factors that affect breeding patterns of the tropical dry forest dominant Guanacaste (*Enterolobium cyclocarpum*), using full-sib paternity analyses, coupled with pollen structure analyses. Guanacaste is common in the Tropical dry forest, occupying a wide variety of habitats, and exhibits a wide range of ecological and demographic situations. Our studies utilize 1,101 seed pods, collected from 27 seed-parents from the Palo Verde National Park in Guanacaste Province, Costa Rica, over a five year period. Some of these trees flowered in more than one year, so there were 44 seed-tree collections in total, with an average of n = 25 pods / single-tree (within year) collection. This is a mimosoid legume, with pollen dispersed in polyads of 32 pollen grains, so that individual pods contain full-sib families from a single donor.

We have developed procedures that combine full-sib paternity analysis with a genetic structure approach, allowing us to extract inference on the structure and flow of pollen across the landscape, without physically locating the pollen donors. The central premise of the structure analysis is that maternal plants, scattered across a landscape, sample pollen from different sets of pollen donors, most pollination being somewhat localized. Genetic divergence of pollen donor pools among seedparents increases with maternal physical isolation, but it decreases with increasing pollen dispersal distance. Comparing degrees of pollen donor divergence between pairs of sampled seed-parents, we gain some inference on the pattern of pollen flow, without pinpointing pollen donor locations.

In any one year, the average probability of paternal identity for seed pods, drawn randomly from the same mother, is  $r_0 \approx 0.0382$ , which implies an effective number of pollen donors of  $N_{ep} \approx 26.2$ per seed parent. There is some shared paternity among pods, but generally with only a few pods from any one donor. The PPI estimate for the total collection is  $R_0 = 0.005$ , without regard to female parent, as compared with a pannictic expectation of (PPI<sub>e</sub> = 1/490 = 0.002, with 490 different male contributors). That implies an  $N_{ep}$ : N ratio of ~ 0.4 for the whole population. It seems clear that pollination is not pannictic, but it is nevertheless quite widespread in Guanacaste.

We do not expect much year-to-year pollinator consistency for a single seed-parent, and the average cross-year PPI value is very small, with most of what there is being found in seed parents with higher numbers of multiple-pod pollen donors. We can also estimate the average distance of pollen flow, and while there is a concentration of local pollination, at least some pollen is flowing considerable distances across the landscape. We discuss these findings and indicate the direction of future studies aimed at elucidating the impact of changing ecological conditions.

Thursday AM - Concurrent Session IV

# Pollen Dispersal Redux: A Look At Recent Analyses

### Bob Westfall

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Conventional wisdom holds that pollen dispersal functions are uniform or nearly so and interpretations of direct estimates of paternity support that assumption. However results from recently developed indirect estimators, MLTR, a maximum likelihood method (Ritland 1989), and TwoGener, a non-parametric least squares analogue (Smouse et al, 2001), indicate that the effective number of pollen parents (Nep) is much lower than those previously assumed: Most published estimates of Nep are less then 100. In contrast, studies using paternity analysis indicate large numbers of contributing males. However, these estimate census numbers of males only where data are available in these studies, effective numbers are much lower. Moreover, results from extensions of the Adams' (Adams and Birkes 1990) maximum likelihood model which estimate components of male dispersal, such as the dispersal function, anisotropy, and plant height, from paternity data (Austerlitz et al. 2004, Burczyk et al. 2002) are consistent with the indirect estimators. Most dispersal functions estimated in these studies are highly leptokurtic, i.e., the estimated shape parameter, b, in  $\exp[-bx]$ , where x is distance from the male parent, is less than 1 (leptokurtic distributions are those with b < 2). With these, most of the matings are near the mother parent. However, distributions with b < 1 are long-tailed, meaning that some contributing males, though infrequent, are quite distant from a female parent. Dispersal behavior such as this results in local population structure or protected polymorphisms (Namkoong and Gregorius 1985), while facilitating rapid adaptations to climate change or in dispersing to suitable habitats. Data we have for Sierran sugar pine are consistent with this hypothesis:  $Nep \approx 5$  (Hipkins and Westfall, in prep), single (Fst = 0.038; Westfall, unpublished), multilocus (Westfall and Conkle 1992), and quantitative trait differentiation (Harry et al., 1983) among populations along elevational transects is high, considering distances among populations, and fossils of sugar pine dating to 800-1350 CE have been found at 3090 m elevation in the eastern Sierra, 600 m higher than the current upper elevation for the species (Millar, et al., submitted).

# Landscape Genetics of Western White Pine: Genetic Structure and Evolutionarily Significant Units?

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The emerging discipline, landscape genetics, offers a valuable approach for managing wild genetic resources. Suture zones (SZ), narrow geographic bands displaying multi-species genetic discontinuities, signify restrictions to genetic homogenization and define co-migrating species assemblages otherwise known as evolutionarily significant units (ESU). Maternal molecular markers have suggested a SZ in the northern Rocky Mountains between two geographic variants of ponderosa pine. A congruent genetic discontinuity was observed in western pine beetle populations. Similar markers in whitebark pine (WBP) suggest a SZ in the north central Cascades. An ESU that correlates with pest behavior may be indicated by this SZ, since north of this SZ, rates of blister rust infection were 72x greater on WBP than on western white pine (WWP), whereas south of this zone, they were only 4x greater. Distributions of endemic pests might also indicate ESUs. If so, the limitation of WWP dwarf mistletoe to the Siskiyou Mountains of southwestern Oregon indicates an ESU. Similarly, WWP ESUs in the northern Rockies and southwestern Oregon may correspond to distributions of western larch and tanoak respectively.

For our study of WWP landscape genetics, we used a rangewide collection of 60 WWP seedlots (OP seed from 5-10 trees), assembled during the 70s. Problems with seed quality and quantity eliminated five seedlots. Plantings were established from 1980 to 1983 at several nurserv and field locations. A 55-entry x 32-character matrix was analyzed using multidimensional scaling to classify seedlots into provisional ESUs and indicate geographic location of possible genetic discontinuities. The matrix includes means and standard deviations of 12 traits assessed at multiple locations (variance differences can also indicate population differences), five traits assessed at one location, and co-occurrences with WWP dwarf mistletoe, western larch, and tanoak. Bud break, bud set, rate of shoot elongation, and total height of three-year-old seedlings were recorded at three locations. Primary-needle color, first-year bud set, first-year height, second-year shoot color, and secondary-needle freezing tolerance were recorded at one location. Provisional ESUs from this analysis were used for analysis of additional data. Seedlings from the original seed lots were planted in long-term plantations (tests) at two locations within the range of WWP (Priest River Experimental Forest in northern Idaho, and Whidbey Island in western Washington), and in central Idaho outside the range of WWP. Total height, blister rust incidence, blister rust mortality, and five-year survivability were recorded after 20 years at 1 location and after 21 years at two locations. Means and standard errors were computed for each test x ESU combination. These data were used to construct generalized reaction norms that were interpreted across paired test locations. Finally, we will report preliminary results of genetic diversity and structure based on AFLP and microsatellite markers obtained from 15 populations collected from across the entire range of WWP. All results from studies presented will be incorporated with results from published reports for associated populations of plants, animals, and fungi to hypothesize numbers and locations of glacial refugia for WWP. Importance of putative ESUs to management, restoration, and conservation of WWP dominated ecosystems will be discussed.

#### Genetic Studies of Acacia Koa and Koa Wilt Disease

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Koa (*Acacia koa* Gray) is Hawaii's premier hardwood, a tetraploid species of this large leguminous genus. As one of only five legume trees indigenous to Hawaii, it became a dominant at mid-elevations where annual temperature averages range between 16° and 22°C. The species is shorter-lived or rare under higher temperatures, at elevations generally below 600 m. It has become evident that koa wilt (causal organism *Fusarium oxysporum* f. sp. *koae*) is a major factor in this failure. The disease is found throughout the islands and has been particularly severe in our progeny trials at 650 m elevation on the Hamakua coast of the island of Hawaii. Several trials were replicated at a low elevation on the island of Oahu, with similar severity.

Our evaluations of genetic diversity trace to collections begun in the 1960's, which showed extensive variation in morphology and isozymes. Progeny trial plantings between 1991 and 2003 involved 550 half-sib families (koa is generally self-sterile) gathered from all major islands in the state. About 50 families were grown annually, and planted in two reps of 10-tree plots. Koa is a fast-growing species under these protected conditions, where weed management is aggressive and ruminant animals carefully excluded (rarely true in nature). Annual increments in height and dbh averaged 1.5 m and 2 cm, resp., and 12-year old trees attained heights >15 m and dbh's >40 cm. Between 40% and 70% of phenotypic variation in dbh at four years could be ascribed to genetic factors. Great diversity occurred in forking, fluting, limbiness, growth-rate and the shape of pods and phyllodes. Few trees were outstanding in form. Inbreeding among provenances was evident in the high uniformity of some half-sib progenies.

Koa wilt caused great mortality among trees in these trials, with average survival percentages of 62% in two years, 43% in 4 years, 28% in 6 years and 7% in 12 years. These values reflected overtopping to some extent, as planting densities were high (1 x 1.5 m), but wilt clearly was a major factor. Family variations in survival were high enough to encourage selection for tolerance. At 4 years, 15% of the families had survived with mortality values <20%. Seeds from these half-sib trees, obtained in 4 to 5 years, were planted in two sets of seed orchards that are now under study for growth and koa wilt tolerance. Seeds from the first of these orchards is available from the Hawaii Foundation Seed Facility as "UH Koa Comp 1".

Koa was highly refractory in our studies of vegetative propagation. We've been able to clone it from juvenile seedlings in greenhouse. Some cloning success also occurred with etiolated branches on nursery-grown trees. It would clearly be valuable to clone (or breed) trees with outstanding wood quality (figure, color) for the craft and furniture industries.

# Genetic Resistance in Port-Orford-cedar to *Phytophthora lateralis* – survival of seedlings from first orchard seed in short-term testing

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Port-Orford-cedar (Chamaecyparis lawsoniana) is highly susceptible to the non-native, invasive root pathogen Phytophthora lateralis, the cause of Port-Orford-cedar root disease. Mortality from this disease has impacted forest ecosystems and has greatly restricted the utilization of Port-Orford-cedar (POC) for restoration and reforestation. Fortunately, a low incidence of genetically resistance trees is found within some POC populations. In the mid-1990's, the USDA Forest Service and USDI Bureau of Land Management began an operational program to develop genetically resistant populations of POC. Working in conjunction with researchers at Oregon State University, over 10,000 field selections have undergone the first stage of resistance screening, a stem dip test. The second screening stage, a root dip test, is well underway for the top 10% of parents from the stem dip test. Selections with high survival in the root dip test are placed into containerized seed orchards at Dorena Genetic Resource Center. To date, orchards have been established for only a few breeding zones, but more will be added as screening is completed. The first orchard cone crops, harvested in 2002, were used in seedling tests of bulk orchard seedlots in 2004. We report here on two short-term trials: a greenhouse test using a root dip inoculation, and a raised bed test using soil previously inoculated with P. lateralis. These trials include bulked seed orchard lots from several breeding zones, bulked woodsrun seedlots (no selection for resistance), and individual control families spanning a wide range of resistance. There were significant differences among seedlots for mortality in both trials. Seedlot mortality in the greenhouse test ranged from 0 to 100% over the 12 months of the test, while mortality in the raised bed test ranged from 25 to 88%. Ten seedlots and seven control families are common to both trials, and there was a significant correlation (r=0.78) between the two tests. In the greenhouse, the orchard lots showed 44 to 50 percent less mortality than the woodsrun seedlots for the three orchards with >22 parents; for the three orchards with <7 parents, the orchard seedlots showed 12 to 33 percent less mortality than the woodsrun lots. Potential gains may be underestimated here due to [1] an unknown level of pollen contamination from more than 1000 parent trees in the breeding arboretum, [2] the nature of some of the woodsrun seedlots used, and [3] the potential presence of a few susceptible parents in the unrogued orchards. Additional selections will be added to orchards as the screening results are updated and orchards will be rogued as needed, following further verification of resistance or susceptibility. Seed from several orchards are now being used by various landowners. Current work in the resistance program involves testing new field selections for resistance, breeding to increase levels of resistance, and implementing field trials to examine utility and durability of resistance.

# "Time is On Our Side – Blister Rust Resistance Traits "Holding Up" After 30+ Years"

# Lauren Fins and Paul Lupo

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The primary objective of this study was to evaluate long-term field performance of Phase I  $F_2$  planting stock, with particular attention to individual rust resistance mechanisms (Lupo 2004). Field assessments in the fall 1999 of  $F_2$  progeny at 31-32 years from seed, showed 19 and 21 percent survival in plantings established from tests begun in 1967 and 1966, respectively, but only 1% survival of the controls under the same conditions. While the low survival percentages of the  $F_2$  may suggest cause for concern, they *should not* be used to estimate long-term survival based on rust resistance, as many factors are known to have contributed to the high mortality, including cattle damage and sunscald. Furthermore, the Phase I  $F_2$  families used in the study were descended from a very limited gene pool (only 14 grandparents), at least ten of which originated within 15 miles of each other, and some of the mortality likely reflects errors in identifying resistant parents, approximately one fifth (21%) of which were subsequently found to be susceptible to blister rust.

A comparison of the proportions of individuals with different rust resistance traits at the time of planting and after 25-27 years of field exposure revealed similar survival among resistance types. With the possible exception of bark reactions, resistance traits appeared to provide consistent protection against blister rust, even with repeated inoculations under field conditions. Height and diameter measurements taken in 1999 confirmed previous reports of little or no significant phenotypic relationships between resistance traits and growth.

We conclude that the Phase I breeding program successfully increased rust resistance in the  $F_2$ , that different resistance traits confer similar long-term survival potential under field conditions, that selection for resistance does not sacrifice growth, and that additional gains in resistance are available through selection and breeding. Nonetheless, since we could not separate losses due to blister rust from losses due to other causes, results of this study cannot be used to generate good estimates of long-term survival due blister rust resistance.

#### Acknowledgements

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# White Pine Blister Rust Resistance in Western White Pine – Family Variation in Infection Percent and Survival in Field Trials

## Richard A. Sniezko, Scott Kolpak, Angelia Kegley, and Robert Danchok

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All nine white pine species in North America are susceptible to white pine blister rust, caused by the non-native invasive pathogen Cronartium ribicola. Western white pine (WWP, Pinus *monticola*) is one of the most susceptible species, suffering high mortality throughout much of its range. Fortunately, there is a low frequency of naturally-occurring genetic resistance to this pathogen. Resistance breeding programs in British Columbia, Idaho, and Oregon now cover much of the species range. These programs started with field selections of phenotypicallyresistant trees and have used artificial inoculation to evaluate seedling progeny from thousands of these selections in short-term testing. A great range in the degree and type of resistance has been found the artificial screening tests, although most seedling families have shown little or no resistance. Until recently, there have been few tests to confirm the field resistance and durability of the resistance individual families. A series of plantings established in Oregon and northern California beginning in 1996 provides some of the first evaluation of family variation in the field. We focus here only on the sites heavily infected with blister rust. Through the 2003 assessment of 13 common families established at two sites, family means for stem infection ranged from 19% to 90% (mean = 52%) at Happy Camp (HC) and 31% to 96% (mean = 59%) at Kerbluey (K). There was a significant correlation (r=0.74) between stem infection incidence at the HC and K. Rust related mortality (family means) ranged from 0 to 56% (mean = 20%) at HC and 0 to 28% (mean=7%) at K. Mortality often lags several years behind infection and is expected to increase. Infection will likely continue to increase as new infection events take place. A separate trial ('Optical') was established in 1998; percent stem infection ranged from 25% to 100% for the 49 families (mean = 89%) at the 2003 assessment. The top family at Optical is an advance-generation cross and has also been outstanding in artificial inoculation testing. High levels of stem infection are not unexpected in most of these families on high hazard rust sites. The most interesting results for these families will come in the next few years since the best families have shown bark reaction or slow canker growth responses in short-term screening - many individuals in these families may survive and grow well with cankers. In the oldest known field test of WWP families in Oregon, planted between 1968 and 1972 with predominantly canker-free survivors from artificial inoculation trials, 92.5% of the trees across the 97 surviving families at the Grass Creek site (GC) had cankers. Individual trees had 0 to 200 cankers. Despite the heavy infection, many trees are alive and growing well after >30 years. At several sites in western Oregon, one type of major gene resistance has been overcome by a virulent strain of blister rust, and it is other types of resistance that show the lowest infection levels and offer the best potential for restoring the species. Future efforts will concentrate on breeding to increase levels of resistance, further field testing, and establishing advance-generation orchards.

Posters

# Flowering Behavior of Port Orford Cedar from Different Provenances Grown in a Common **Garden** <sup>1</sup> Jerry Hill<sup>1</sup>, Don Zobel<sup>1</sup>, and Richard Sniezko 3

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Maintaining genetic diversity in breeding and production seed orchards is difficult. The genetic contribution of a given tree in an open-pollinated orchard can vary greatly depending on flower abundance and timing of flowering. To estimate these parameters for Port-Orford-cedar (*Chamaecyparis lawsoniana*), clones of 25 trees representing three geographical or physiographic extremes of its distribution were studied in a common garden setting in a greenhouse containerized seed orchard (CSO). These sources were northern coastal (low elevation), northern interior (high elevation), and southern interior (high elevation). Cone development stages are illustrated in greater detail than previously available. Preliminary analysis shows that, in general, trees from low elevations began and completed flowering earlier than higher elevation trees, though there was considerable variation within each group. With cooler weather, pollen cone development slowed greatly and pollination drops tended to be more abundant or persist longer. Some trees showed a bimodal pattern of development of female cones (two peaks separated by several weeks), a feature which was more common in trees from southern sources. There may also be differences between genotypes in the stages of female cone maturity where pollination drops occur. All of these features have definite implications relative to the management of, and preserving genetic diversity in, the Port-Orford-cedar CSO.

# Variation in Wood Quality of Coastal Douglas-fir

# **Randy Johnson**<sup>1</sup> and Barbara Gartner<sup>2</sup>

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Like most species, the wood quality we know most about in coastal Douglas-fir is wood density. Strength (modulus of rupture, MOR) and stiffness (modulus of elasticity, MOE) are of greater importance to the lumber industry, but have received much less attention than wood density because it is inexpensive and straightforward to measure. Variation patterns of wood density have been examined at the within-tree, among-tree, among-stand and among-provenance levels. Variation at the within-tree level is much greater than the variation among-tree level, but is fairly well understood. The variation among trees within a stand is 3 to 4 times greater than the among-stand variation, and therefore of importance to forest managers. The reported heritabilities of density range from 0.54 to 0.90, suggesting that most of this within-stand variability is controlled by genetics. Based on our data, we estimate that over 35% of the variation in MOE in the region is controlled by genetics (for a fixed rotation age).

We used the Director HM-200® to estimate dynamic MOE of logs from 39 open-pollinated families at four progeny test locations in the Oregon Coast Range. The HM-200 measures the speed of sound through a log, which is highly correlated with MOE. Based on preliminary analyses, the estimated heritabilities for the speed of sound (MOE) at the individual sites ranged from 0.37 to 0.81. The heritability calculated over all four sites was 0.46; across-site heritabilities for height and DBH were 0.23 and 0.32 respectively. The family-by-site variance for "MOE" was only 16% of the family variance component; suggesting strong genetic correlations among sites. An adverse family-mean correlation was found for diameter growth and MOE (r = -0.2 to -0.3), but not for height growth and MOE (r < -0.1).

Posters

# Genetics and Silviculture Results - NWTIC Umpqua/Noti Breeding Unit – 10-30 Year Data

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A cooperative tree improvement testing program was established in the Umpqua cooperative, Noti Breeding Unit (Oregon Coast Range) to develop improved seed. 326 families (11 sets of 30 families) were tested in eight progeny test sites (established in 1972-1975). The sites were measured at 5, 10, 15 and 30 years of age. Five sites were thinned at approximately age 25 years (removing about one third of the trees). Thirty year measurements were taken in 2001 and 2002 to determine long term results, quantify growth and yield results, and verify genetic gain predictions. The NWTIC completed a BLUP analysis for these data in 2005. Nine sets are summarized in this report.

The estimates of across site heritabilities of individual-tree for age-15 dbh, age-30 dbh, age-10 height and age-15 height were  $0.09\pm0.01$ ,  $0.12\pm0.02$ ,  $0.09\pm0.01$  and  $0.12\pm0.01$  respectively. The family-mean heritabilities for these traits were  $0.59\pm0.04$ ,  $0.58\pm0.04$ ,  $0.61\pm0.03$  and  $0.67\pm0.03$  respectively. The family-mean correlation between age-15 predicted gain for dbh, and age-15 height with age-30 predicted gain for dbh were 0.77 and 0.70 respectively.

The top 20 families (based on predicted dbh-15 within-set gain) had an average of 15.7% predicted gain. The "realized gain" [((top 20 – overall mean)/overall mean)\*2\*100] of these same 20 families for age-30 dbh was 16.3%. Thus family selection at age-15 appears to provide good gains at age-30. The average predicted genetic gains for the top 3 families in each set (diameter at age 30) was 12.4%, while the actual growth of these same families (comparison to plantation mean on each of the eight sites) averaged 7.7%.

				Relative	
<u>Site</u>	<u>DBH</u>	BA/Acre	Trees/AC.	<u>Density</u>	<u>Thinning</u>
Ferguson	284	155	219	46	Yes
Pataha	229	106	223	35	Yes
Oxbow	250	129	232	41	Yes
Smith Creek	223	110	250	37	Yes
Clay Creek	258	200	381	62	No
Coyote	271	163	255	50	Yes
Steampot	210	123	307	42	No
Triangle Lake	248	191	330	60	No

Diameter, basal area, trees per acre, and relative density for the eight sites were:

# Inbreeding depression in Port-Orford-cedar: early survival and growth of self-fertilized and open-pollinated progeny

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*Phytophthora lateralis* is a non-native, invasive root pathogen that has caused extensive mortality of *Chamaecyparis lawsoniana* (Port-Orford-cedar) in some areas of its natural range in southwest Oregon and northwest California. An operational breeding program to develop populations of Port-Orford-cedar that are resistant to *P. lateralis* is underway. Several containerized orchard populations have been established and more are being developed as disease resistance testing is completed.

We are investigating inbreeding as a tool to elucidate the patterns of inheritance for root disease resistance. Several well characterized families suggest a single dominant resistant gene is present in high survival families. Disease resistance testing on progeny from self-fertilized parents is a potential strategy to determine the prevalence of single gene resistance in the orchard populations. However this strategy requires a minimal impact of inbreeding depression on seed yield and seedling survival. In addition, selfing or related matings may be common in natural populations that have been decimated by *P. lateralis*. In these populations, if significant inbreeding depression exists, then Port-Orford-cedar may lose competitiveness with other tree and shrub species.

Inbreeding depression has been examined in several conifer species. Generally, inbreeding depression causes a decrease in early height and survival, and a reduction of filled seed due to the expression of embryonic lethals. We examined the effects of inbreeding in Port-Orford-cedar by examining growth and survival in self-fertilized and open-pollinated progeny. Eighteen parent trees having both first generation self-fertilized ( $S_1$ ) and open-pollinated ( $F_1$ ) seed were sown in March 2005. Seedlings were grown in 'supercell' containers (164 cm<sup>3</sup>) in a greenhouse for several months then moved outdoors. Early survivorship and growth were examined in 5 month old seedlings.