## **PROCEEDINGS OF THE TWENTY-FOURTH MEETING**

### **OF THE**

## CANADIAN TREE IMPROVEMENT ASSOCIATION

PART 1 Minutes and members' reports PART 2 Symposium



## COMPTES RENDUS DE LA VINGT-QUATRIÈME CONFÉRENCE

DE

## L'ASSOCIATION CANADIENNE POUR

## L'AMÉLIORATION DES ARBRES

1<sup>re</sup> PARTIE Procès-verbaux et rapports des membres 2<sup>e</sup> PARTIE Colloque



### PROCEEDINGS

### **OF THE**

### **TWENTY-FOURTH MEETING**

### OF THE

## CANADIAN TREE IMPROVEMENT ASSOCIATION

## Part 1

## THE FUITURE FORESTS: OPTIONS & ECONOMICS

Held in Fredericton, New Brunswick August 15-19, 1993

Editor:

J. Lavereau

Additional copies of this publication may be available from:

Joy Lavereau Editor, C.T.I.A./A.C.A.A. Natural Resources Canada Petawawa National Forestry Institute P.O. Box 2000 Chalk River, Ontario K0J 1J0

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### **COMPTES RENDUS**

## DE LA

## VINGT-QUATRIÈME CONFÉRENCE

DE

## L'ASSOCIATION CANADIENNE POUR L'AMÉLIORATION DES ARBRES

## PARTIE 1re

## LES FORÊTS DE L'AVENIR: LES OPTIONS ET L'ÉCONOMIE

Fredericton (Nouveau-Brunswick) du 15 au 19 août 1993

**Rédactrice:** 

J. Lavereau

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### Préparé par

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# C.T.I.A./A.C.A.A. 24th BUISINESS

## MEETING MINUITES



### C.T.I.A./A.C.A.A. 24th BUSINESS MEETING MINUTES

Kathy Tosh chaired the 24th Business Meeting of the CTIA/ACAA held in the K.C. Irving Theatre, Fredericton, New Brunswick on Wednesday August 18, 1993.

### 271. Minutes of the 23rd Meeting

(as printed in the proceedings from the 23rd meeting (Part I))

Motion: The minutes of the 23rd Business Meeting be approved as published.

Moved by: Gordon Murray Seconded by: Yill Sung Park Carried.

### 272. Membership

### **New Active Members**

The names of nominated new active members were presented as follows:

Robin Browne	Agriculture Canada Manitoba
Stewart E. Cameron	Natural Resources Canada New Brunswick
Campbell G. Davidson	Agriculture Canada Manitoba
Peter de Groot	Natural Resources Canada Sault Ste. Marie, Ontario
Marie Deslauiers	Ressources naturelles Canada Region du Quebec
Ronald D. Hallett	Natural Resources Canada New Brunswick
Richard Hamelin	Ressources naturelles Canada Region du Quebec
Peng Li	Université Laval Quebec
John Major	Natural Resources Canada Chalk River, Ontario
Spencer McDougald	Weyerhaeuser Canada Saskatchewan

Gene Namkoong University of B. C. British Columbia

Ben Sutton

Forest Biotechnology Centre British Columbia

Brenda Vanstone

University of Toronto Ontario

Motion: That the nominated new active members be duly elected.

Moved by: Yill Sung Park Seconded by: Guy Caron Carried.

### **New Corresponding Members**

The following were recorded:

Rhys Andrews	Selkirk College British Columbia
Murray Ferguson	Can. Pac. Forest Products Ltd. Dryden, Ontario
Nathalie Isabel	Université Laval Quebec
W. A. Kenney	University of Toronto Ontario
Sakti Jana	University of Saskatchewan Saskatchewan
Gwen McGimpsey	Dept. of Natural Resources Manitoba
Michel Rioux	Min. des Forests du Quebec St. Modeste, Quebec
Kimberly Stinson	Ont. Fed. of Anglers & Hunters Peterborough, Ontario
Kristjisn Vitols	University of Toronto Ontario

Joy Lavereau noted that 9 members requested withdrawal from active member status, due to no longer being involved with tree improvement and 7 members asked to be changed to corresponding members. Active Members total 127, Corresponding members 177, Canadian institutions and libraries 72, U.S.A. addresses total 101 members and other foreign addresses 137. Members are urged to assist the Executive Secretary by notifying promptly of changes of address, names of new prospective Active and Corresponding members, changes of responsibility that directly affect members status in the Association, retirements and deaths.

### 273. Chair's Report

Hosting a conference such as this requires many hours of hard work and I would like to take this opportunity to thank the other members of the executive committee, Judy Loo-Dinkins, Yill Sung Park, Don Fowler, Greg Adams, and especially Dale Simpson. I would also like to thank the New Brunswick Department of Natural Resources and Energy for agreeing to host the 24th meeting as well as, the many organizations that contributed to the success of this meeting.

The meeting was well represented from all parts of Canada as well as various points in the U.S. and overseas. All of the events scheduled in the conference went smoothly and from the comments we have received I think very successful.

To conclude, I would like to thank my staff at the Tree Improvement Unit for all their hard work preparing for the conference and helping with all the details that make a conference such as this a memorable event.

### 274. Treasurer's Report

The revised amended financial statement for the period of May 10, 1989 to June 30, 1991\* was prepared by appointed Treasurer Joy Lavereau and was tabled for membership information and acceptance (See attachment #1). The statement shows an account balance of \$8,736.20 and Guaranteed Investment Certificates (G.I.C.) totalling \$16,500.00 as of June 30, 1991.

The financial statement prepared by appointed Treasurer Joy Lavereau (for Treasurer Steen Magnussen) for the period of July 1, 1991 to December 31, 1991 and was tabled for membership information and acceptance (See attachment #2). The statement, as of December 31, 1991 shows a balance of \$2,676.33 in the association's account and \$22,500.00 in G.I.C.'s.

The financial statement for the period of January 1, 1992 to July 30, 1993 was prepared by appointed Treasurer Joy Lavereau was tabled for membership information and acceptance (See attachment #3). The statement shows a balance of \$2,726.78 in the association's account as of July 30, 1993 and G.I.C.'s totalling \$24,000.00.

Motion: That the financial statements as presented be accepted.

Moved by: Ron Smith Seconded by: Jack Woods Carried.

### 275. Financial Contributions

Can/NB Cooperation Agreement on Forest Development Fraser Incorporated J.D. Irving Limited Juniper Lumber Company New Brunswick Department of Resources and Energy New Brunswick International Paper Forest Products Limited Miramichi Pulp and Paper Incorporated Natural Resources Canada - Maritimes Region Stone-Consolidated Incorporated Université de Moncton

Motion: That the CTIA/ACAA express our sincere appreciation to these contributors.

Moved by: Yill Sung Park Seconded by: Dale Simpson Carried.

### 276. Editor's Report

The proceedings were printed and distributed to all Active members, Canadian universities and libraries, all participants of the 23rd meeting, sponsoring and honorary members during the second month of 1992. Three hundred and four proceedings were mailed. Invited speakers papers from the 1991 CTIA conference were published in full, and appeared in the October 1992 issue of "The Forestry Chronicle".

The Science and Sustainable Development Directorate and the Petawawa National Forestry Institute of Natural Resources Canada paid all costs of printing and distribution of Part I and publishing in the Forestry Chronicle.

A form letter advising the theme of the 23rd biennal meeting, the proceeding context and a request for ten dollar donation to obtain one copy was sent to all Corresponding members, U.S.A. addresses and Other Foreign Countries. There were 38 donations from Canadian addresses, 34 from U.S.A. addresses and Other Foreign countries obtained 31 copies.

The next fiscal year printing and distrubution will also be supported by the Sustainable Development Directorate.

Prompted by a request from C.W. Yeatman a complete set of Canadian Tree Improvement Association proceedings and NRC Subcommittee meeting dating back to 1937 have been archived at Petawawa Publication Distribution Centre in a fireproof vault.

### 277. Educational Committee

To promote students' awareness in tree improvement activities and forest genetics research, the forestry faculties of Canadian universities were encouraged to nominate a student for the travel award to attend the 24th CTIA/ACAA meeting. The following students received the awards, which provide all the costs of meeting, including registration, accommodation, pre-conference tour and travel:

Marilyn Cherry Christine Hansen Nathalie Isabel Stephanie Nicholas Emanuel Sildor Kristjan Vitols

University of British Columbia University of Alberta Université Laval University of New Brunswick Université de Moncton University of Toronto

### 278. Working Groups Reports

This year marks the 10th anniversary of the Tree Seed Working Group. The TSWG has held regular Workshops at all but one CTIA/ACAA meeting since its creation. From

the original 93, membership has risen to 226, of which 14% are from outside Canada. Hugh Schooley presented at this CTIA/ACAA meeting a poster on the short history of TSWG.

The Tree Seed Working Group held its regular Biennial Business Meeting on August 16 with 25 present.

Four issues of the TSWG NewsBulletin were published in the past two years and distributed to all members and 32 institutions. A membership survey was conducted in 1992 and response was great. Hugh Schooley will continue his great work as Editor of the NewsBulletin.

This year's TSWG workshop themed "Seed Testing" tied in with the newly created Tree Seed Processing and Testing Working Party. Since 1991 Dave Bewick resigned as Coordinator of the TSPTWP owing to job reappointment. Dave Kolotelo acted as interim coordinator until his official appointment at this TSWG business meeting. The TSPTWP will try to serve the need of the many seed users of the TSWG. Objectives and activities planned for the TSPTWP will be published in the upcoming November issue of the NewsBulletin.

Efforts will be made in the next two years to increase participation to the Cone and Seed Insect Working Party. Peter de Groot will encourage members of the IPMISO (Integrated Pest Management in Seed Orchards) Network to submit information reports of their activities and research findings to the NewBulletin. IPMISO is comprised of entomologists, pathologists, and seed orchard pest managers with a focus of developing integrated pest management systems for seed orchards. Through this information exchange, both the CSIWP and IPMISO should benefit. It was also acknowledged that the CSIWP is more likely to persist than the IPMISO.

The TSWG intends to be actively involved in the 25th CTIA/ACAA meeting to be held in Victoria, B.C. in 1995 by hosting another Workshop. In addition, a tour to the Surrey Seed Center has been requested by membership. Dave Kolotelo will act as liaison with the organizing committee of the 25th CTIA/ACAA meeting.

This concludes my report,

Guy E. Caron Chairperson 1991-95

279. New Business

#### 279.1 Retirement

Motion: That in light of the pending retirement of Dr. E.K. Morgenstern that the current CTIA/ACAA executive compose and send a letter on behalf of the CTIA/ACAA to the Faculty of Forestry, University of New Brunswick urging the Faculty to maintain a staff position in forest genetics.

Moved by: Don Fowler Seconded by: Hugh Schooley Carried.

#### 279.2 Distribution of proceedings

Joy Lavereau presented a couple of questions, as to who should receive gratuitous copies of the CTIA/ACAA proceedings and/or should the association request a donation to cover the printing and mailing costs. Joy Lavereau advised costs of the printing and

distribution of Parts I and II. Tim Mullin suggested keeping the costs to a minimum and to request a donation. Jean Bousquet suggested using a box system for which level of membership/proceedings status preferred. Gordon Murray stated all Active members, Participants of the meetings, Canadian Universities and Libraries, Honorary and Sponsoring members should receive gratuitous copies of the proceedings.

Motion: Distribution of the 25th CTIA/ACAA proceedings will be determined by the 1995 executive committee.

Moved by: Jack Woods Seconded by: Gordon Murray Carried.

### 279.3 Name change of CTIA/ACAA

There was some dicussion on a name change for the Canadian Tree Improvement Association as a result of the words "Tree Improvement". John Russell suggested a change to Canadian Forest Genetic Association.

Motion: That an official name change be held off, until members were advised of the proposal and that the 1995 executive committee would be responsible for notification of the motion to all Active members.

Moved by: Jerry Klein Seconded by: Alex Mosseler Carried.

### 279.4 CTIA/ACAA Finanical Holdings

There was much deliberation on the excess investments of the CTIA/ACAA. Suggestions on how to effectively use some of the holdings were as follows:

- 1. Why not sponsor two Forestry students per university per Canadian Tree Improvement Association Meeting.
- 2. Perhaps sending forestry students on or to specific forestry related meetings, poster sessions, etc.
- 3. That a scholarship fund to be set up for graduating Forest Genetic students.
- To award Forestry Graduate students on papers presented at the CTIA/ACAA biennal meetings.

Motion: To grant the 1995 executive committee the decision on student awards and draw no more than \$1000.00 per meeting.

Moved by: Bruce Dancik Seconded by: John Russell Carried.

### 280. Future Meeting

### 280.1 Location of the 1995 meeting

John Russell confirmed that B.C. Ministry of Forests and W.F.G.A. will host the 25th biennial meeting in Victoria, British Columbia.

### 280.2 Location of the 1997 meeting

Motion: That the 1997 CTIA/ACAA meeting be jointly hosted by Yves Lamontagne (Ministere des Forêts), Jean Beaulieu (Natural Resources Canada), and Jean Bousquet (Université Laval) and will be held in Quebec City.

#### 280.3 Location of the 1999 meeting

Tentative support from the Praire Provinces or Thunder Bay, Ontario to hold the 1999 meeting.

### 281. Election of New Executive

The following slate of officers for election to the 1993/95 CITA/ACAA executive:

Chairperson: Alvin Yanchuk B. C. Forest Service Research Branch

Vice-Chairperson: John Russell B. C. Ministry of Forests

Vice-Chairperson Arrangements: Michael Stoehr B. C. Ministry of Forests

Treasurer: Joy Lavereau Natural Resources Canada, PNFI

Motion: That the slate of officers proposed be elected.

Moved by: Gordon Murray Seconded by: Hugh Schooley Carried.

#### 282. Adjournment

Motion: That the members of the CTIA/ACAA thank the executive for their efforts over the past two years and for an exciting and successful meeting.

Motion: That the 24th business meeting of the CTIA/ACAA be adjourned. Moved by: Gordon Murray

### Attachment # 1.

### CTIA/ACAA Financial Statement Amendment 5.0 Treasurer's Report May 10, 1989 to June 30, 1991\*

	Balance May 10, 1989	•	\$4,317.78
Credit:			
Interest earnings (GIC, Account)	\$ 4.008.32		
Surplus from 22nd meeting	1,231.06		
Back pay of advance of 22nd meeting	1,000.00		
<u>Contributions</u>			
Canadian Pacific Products Ltd.	\$ 500.00		
Ont. Forest Industries Association	200.00		
Weyerhaueser Canada Ltd.	250.00		
Fotal Credit	\$ 7,189.38		\$ 7,189.38
Guaranteed Investment Certificates: (as of M	lay 10, 1989)	,	
GIC Principal	\$ 9,180.00		
GIC Principal	1,100.00		
GIC Principal	4,500.00		
Fotal GIC's		\$14,280.00	
Guaranteed Investment Certificates: (as of Ju	ıne 10, 1991)		
GIC Principal	\$ 6,000.00		
GIC Principal	6,000.00		
GIC Principal	4,500.00		
Fotal GIC's		\$16,500.00	
Invested (Debit)			\$ 2,220.00
Debit:		<u>, , , , , , , , , , , , , , , , , , , </u>	
Printing of stationary	\$ 368.88		
Printing cost of announcement and pro	ogramme 119.94		
Service charges for safety deposit box	62.14		
Fotal Debit	\$ 550.96		\$ 550.96
Cash balance	lune 30, 1991*		<u>\$ 8.736.20</u>
Invested GIC	Dalance		210,200,00
Total Holding	<u>s</u>		<u>\$25,236.20</u>
Total Holding	<u>s</u>		<u>\$25,236.20</u>

\* excluding expenditures and registration fees for the 23rd meeting.

### CTIA/ACAA Financial Statement July 1, 1991 to December 31, 1991

	Cash Balance July 1	, 1991	\$ 8,736.20
Credit:			
Interest earning (GIC, Account)	\$ 1.497.13		
Accumulated monies from 23rd meeting	; 14,800.83		
Total Credit	\$16,297.96	<u></u>	\$16,297.96
Guaranteed Investment Certificates: (as of July	y 1, 1991)		
GIC Principal	\$ 6,000.00		
GIC Principal	6,000.00		
GIC Principal	4,500.00		
Total GIC's		\$16,500.00	
Guaranteed Investment Certificates: (as of De	cember 31, 1991)		
GIC Principal	\$10,000.00		
GIC Principal	8,000.00		
GIC Principal	4,500.00		
Total GIC's		\$22,500.00	
Invested (Debit)	<u></u>		\$ 6,000.00
Debit:			
Announcements Aboriculture (U.S.A.)	\$ 179.25	,	
Printing costs for 23rd meeting program	mes 91.05		
Cost of 23rd meeting	16,087.53		
Total Debit	\$16,357.83		\$16,357.83
Cash balance D	ecember 31, 1991	, ,	\$ 2,676.33
Invested GIC ba	alance		\$22,500.00
Total Holdings			<b>\$25,176.33</b>
		•	

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### Attachment # 3.

### CTIA/ACAA Financial Statement January 1, 1992 to July 30, 1993

	Balance January 1,	1992	\$ 2,676.33
Credit:			
Interest earnings (GIC, Account)	\$ 2,198,29		
Donations for 23rd Meeting Proceeding	s 1,052.28		
Total Credit	\$ 3,250.57		\$ 3,250.57
Guaranteed Investment Certificates: (as of Jan	nuary 1, 1992)		
GIC Principal	\$10,000.00		
GIC Principal	8,000.00		
GIC Principal	4,500.00		
Total GIC's		\$22,500.00	
Guaranteed Investment Certificates: (as of Jul	y 1, 1993)		
GIC Principal	\$10,000.00		
GIC Principal	8,000.00		
GIC Principal	6,000.00		
Total GIC's		\$24,000.00	
Invested (Debit)			\$ 1,500.00
Debit:	<u></u>		
24th CTIA/ACAA advance	\$ 1,500.00		
Distribution fees: of the 23rd Proceedin	es 193.62		
Money order fees	6.50		
Total Debit	\$ 1,700.12		\$ 1,700.12
Cash balance I	uly 1, 1993		<u>\$ 2,726.78</u>
Invested GIC t	balance		\$24.000.00
Total Holdings	5		<u>\$26,726.78</u>
# IN MEMORY OF



#### IN MEMORY OF S. ROSS

Although Steve has not been with us for well over a year now, frequent encounters with his past work serve to keep my memories of him as a colleague and friend fresh in my mind. His absence as a dependable reviewer of manuscripts for the CJFR as well as an always helpful and enthusiastic colleague still provides an unfillable void, underscored because there aren't that many who make their living trying to make trees flower. But in a way I now ...rejoice that I am so often reminded of him, because the usefulness of his work will endure for some time.

My friendship and professional acquaintance with Steve began in 1974, when I met him as my first Weyerhaeuser research review in Washington, DC, at the beginning of the Golden Era of the Companies tree improvement program. I was fascinated with the skillful way he rolled his own cigarettes, while he discoursed emphatically with a slight stutter on girdling, water stress, root pruning and other torments that make trees flower. But he was also very helpful as we began our North-South rivalry to see who could make flower stimulation operational first on either Douglas fir or loblolly pine. My job was to follow Steve's lead, because he and Dick Pharis had published an abstract in 1973 reporting that GA<sub>4/7</sub> promoted flowering in Douglas fir grafts. However, the potential usefulness of these results was greeted with some skepticism by both company geneticists and tree breeders, so Steve and I welcomed each other as allies in our struggle to convince them to support our work, so that Tissue Culture research did not gobble up all available funds. Synergism was one of Steve's favorite words, and although in his enthusiasm he sometimes over used it, I like to think our relationship had a certain synergy of its own.

The fact that I have been asked to present this eulogy is a testimony to the many contributions Steve has made to manipulating conifer development, and I perform this small task with a great deal of gratitude to him, that I am confident many of us share. I am very glad I saw Steve a couple of months before he died. He was still rolling his own cigarettes, and discoursing on his work with as much enthusiasm as ever, as we drove through much of BC viewing his projects, and planning further collaboration.

> M.S. Greenwood University of Maine

#### IN MEMORY OF W.E. RAITANEN

On November 14, 1991 William Edward Raitanen (Bill) died suddenly in Ottawa at age 41. He is survived by his wife Diane, their two sons and sister Liisa.

Bill joined the Ontario Ministry of Natural Resources in the mid-1970s in Brockville. Under Bill's leadership, a very successful program of hybrid poplar technology development and transfer was initiated in the eastern part of the province. As the program grew and broadened its scope, strong ties were established with the International Energy Agency and the International Poplar Commission. In memory of Bill, a preferred very fast-growing and resilient hybrid poplar clone has been named "Williamsburg".

Following his poplar days, Bill spent 5 years in Timmins initiating and coordinating the Northern Forest Development Group. The Group's activities focused on forest science and technology development, including the tree improvement program.

In the later part of 1989, Bill moved his career to Forintek in Ottawa. He very much enjoyed his work encouraging private investors to support Forintek's products.

Those who knew Bill, knew him as a strategic thinker and a creative person who liked to start new things. He was also known for the tremendous enthusiasm and dedication he brought to his work. Although Bill's life and career were cut short, he had already made a meaningful contribution to forestry in Ontario. This contribution will live on, but his presence shall be missed.

> R. Ford O.M.N.R.

#### IN MEMORY OF J. PITEL

Jacob ("Jack") Andrew Pitel, a graduate of Carleton University, joined the staff of the Petawawa Forest Experiment Station in 1967 as a technician in forest biochemistry. Two years later Jack moved to Ottawa as a member of the Forest Ecology Research Unit where he worked under Dr. Durzan developing new methods for analysis of nucleic acids and chromosomal proteins. In 1975, he returned to Petawawa, serving briefly as Unit Head before moving into other Projects to participate in research that progressed from seed biochemistry to biochemical and molecular genetics.

As his career progressed from the responsibilities of a research technician to those of a research scientist, Jack earned a well deserved reputation for the high quality of his work, his dedication, and his productivity. Most recently he is remembered for his work on gene cloning and characterization, and for his key role in generating the conifer cDNA and genomic DNA banks which constitute a unique and valuable genetic resource supporting ongoing research.

When he died on the 17th August, 1991, Jack was a respected member of the research team in the Forest Genetics and Biotechnology Program at the Petawawa National Forestry Institute. He is much missed by his many colleagues and friends.

G. Murray P.N.F.I.

# ACTIVITY REPORTS

FROM ACTIVE CTIA

MEMBERS



## IMPLEMENTATION OF THE NEW DIRECTIONS PLAN IN NEWFOUNDLAND AND LABRADOR

#### C.M. Harrison

## Department of Forestry and Agriculture P.O. Box 2006, Herald Building Corner Brook, Newfoundland A2H 6J8

Keywords: Seed orchards, grafting, inter-agency co-operation.

In my report two years ago (Harrison, 1991), I described a new five-year plan which was put in place as a result of a comprehensive review of the province's tree improvement programme. The objective of this report is to bring the membership up to date on the implementation of that plan.

The first priority of the plan was to complete all first-generation seed orchards. That objective will largely be realized in 1993. Site preparation is complete or nearly complete for white spruce clonal orchards and black spruce seedling orchards at Wooddale Provincial Tree Nursery, near Grand Falls, and at Pynn's Brook Management District Office near Corner Brook. A site is also being prepared for a larch clonal orchard at Wooddale Nursery. The orchards at Wooddale will be for the Island, those at Pynn's Brook will be for Labrador and the Northern Peninsula. A former white spruce orchard at Lady Slipper Road, near Corner Brook, was abandoned due to a conflict with watershed management, amongst other reasons. The soundest and healthiest grafts were moved from that orchard to the present site at Wooddale. By the end of 1993, a total of 1,700 grafts will have been planted in the Wooddale orchard.

Seedlings have been raised from seed collected from 196 black spruce plus trees, and the 12 Quebec families that performed best in the progeny tests of Quebec plus-tree families that were established in 1985. These seedlings will be planted in the seedling seed orchards, as well as in progeny tests in western and central Newfoundland and Labrador, in 1993.

Seed from the Quebec plus trees, both in 1985 and in 1992, was supplied by MFO (Ministère des Forêts). Other co-operative efforts with that agency have been undertaken, recently and in the past. This year, trials of European and Dunkeld larch will be established simultaneously at various locations in the two provinces, using seedlots obtained by MFO. Trials of introgressants between sitka and interior spruces were established at two Newfoundland locations in 1992, using seedlots supplied by the British Columbia Research Corporation.

In other inter-agency co-operation, 1.8 ha of native red pine representing 100 provenances were planted at a gene pool reserve/orchard at Wooddale Nursery. A similar plantation was established on the Thomas Howe Demonstration Forest at Gander. Seed for this project was gathered from stands all over the island and a few mainland sources by Alex Mosseler, then of NFRC, now of PNFI. A similar project of native white pine is planned for 1994, also in co-operation with Dr. Mosseler.

The vegetative propagation project envisioned in 1991 had to be put on hold due to budgetary constraints. For similar reasons, no new biotechnology projects have been undertaken. However, we do still try to keep ourselves abreast of developments in that field.

### REFERENCES

Harrison, C.M. 1991. New Directions in Tree Improvement in Newfoundland and Labrador. Proc. 23rd CTIA Meeting, Ottawa, On. pp 37-38.

#### J.D. IRVING LTD. - TREE IMPROVEMENT SUMMARY

#### Greg Adams

## J.D. Irving Ltd. - Sussex Tree Nursery R.R. #4 Sussex, New Brunswick E0E 1P0

## Keywords: Picea mariana, P. glauca, P. abies, P. rubens, Pinus banksiana, Larix laricina, seed orchards, tree breeding, vegetative propagation.

First generation seed orchard management continues and includes establishment of a new red spruce orchard. The establishment of second generation black spruce and jack pine clonal orchards using selections from family tests is nearing completion. Tree breeding and field testing to allow roguing of first generation orchards is also nearly completed for black spruce, white spruce and jack pine. All seed used for reforestation with the exception of red and Norway spruces originates from seed orchards.

Work is also continuing on vegetative propagation focussing mainly on amplifying full-sib crosses among second generation black spruce selections.

#### SEED ORCHARDS AND SEED PRODUCTION

Maintenance continues on the 61 ha of first generation clonal seed orchard including black spruce (*Picea mariana* (Mill.) B.S.P.), white spruce (*Picea glauca* (Moench) Voss), Norway spruce (*Picea abies* (L.) Karst.), jack pine (*Pinus banksiana* Lamb.) and eastern larch (*Larix laricina* (Du Roi) K. Koch). A 2.4 ha red spruce clonal seed orchard was established at the Sussex Tree Nursery using grafts provided by the Nova Scotia Tree Improvement Working Group (NSTIWG). This orchard will supply stock requirements for Nova Scotia and south-eastern New Brunswick. Roguing is scheduled to begin in the jack pine clonal orchard in 1993 based on New Brunswick Tree Improvement Council (NBTIC) family test information. Because of the current seed supply, cone harvesting is only being conducted in the oldest, most heavily rogued jack pine and black spruce seedling seed orchards.

All jack pine, black spruce, white spruce and eastern larch seed grown originates from orchard seed. These species account for 80 percent of nursery production. In 1991 and 1992, 143 kg and 77 kg respectively of orchard seed was processed.

Establishment of second generation black spruce and jack pine orchards is essentially complete for a total of 17 hectares. The first cones were harvested from the second generation jack pine orchard in 1992.

#### TREE BREEDING AND PROGENY TESTING

Tree breeding and progeny testing for roguing the clonal seed orchard is nearing completion for black spruce, white spruce and jack pine. Accelerated greenhouse progeny tests are continuing for black spruce and jack pine to provide information for assortative pair-mating

among clones. The first pair-matings are being field planted in 1993 for future selection. Tree breeding for second generation selections continues *in situ* as well as in the breeding hall at Sussex Tree Nursery in cooperation with NBTIC to produce polycross families for field testing. Breeding and testing Norway spruce is just starting because of recalcitrant flowering in this species. This is aggravated by the young age of the selections. Work with gibberellic acid ( $GA_{4/7}$ ) to induce flowering is continuing to try and overcome this obstacle. Breeding and testing of eastern larch is not a priority at this time because of the limited amount of stock planted.

#### **VEGETATIVE PROPAGATION**

A pilot-scale clonal production program has been initiated using full-sib families of second generation black spruce parents which were produced in the breeding hall in 1992. It is anticipated that rooted cutting production will be scaled up over the next few years.

#### PUBLICATIONS

- Adams, G.W. 1992. Pollen monitoring studies in J.D. Irving Ltd. seed orchards and orchard management implications. In Proc.: Challenges in Pollen Dispersal and Pollen Contamination Workshop, Ed. by F. Di-Giovanni and D. Joyce, Report CCAD- 92-008, Environment Canada, Centre for Atmospheric Research Experiments, Egbert, Ont., Feb. 5, 1992, pp.21-26.
- Adams, G.W. and Greenwood, M.S. 1992. Optimization of environmental regimes for flowering in an indoor breeding hall for black spruce, white spruce and jack pine. In Proc.: IUFRO meeting P2.02.02, S2.01-05, S2.01-13, S2.02, S2.04, P2.04. Mass Production Technology for Genetically Improved Fast Growing Forest Tree Species, Bordeaux, France, Sept. 14-18, 1992, 9pp.
- Carter, K.K., Adams, G.W. and Greenwood, M.S. 1991. Early family selection in black spruce. In Proc.: 1st North. For. Gen. Assn. meeting, Burlington, Vermont, July 1991, 7pp.
- Sulzer, A.M., Greenwood, M.S., Livingston, W.H. and Adams, G.W. 1993. Early selection of black spruce using physiological and morphological criteria. Can. J. For. Res.: In press.

#### FRASER INC. – TREE IMPROVEMENT PROGRAMME

### Ray LeBlanc

## Fraser Inc. 27 Rice Street Edmundston, New Brunswick E3V 1S9

Keywords: Picea mariana, Picea glauca, seed collections, seed orchards, testing.

Since 1991, the establishment of the first generation white spruce and second generation black spruce orchards have been completed and the white spruce orchard is now producing seed. The testing effort continues to progress and the second roguing of the black spruce seedling orchards has started. Cooperative trials are ongoing.

#### SEEDLING SEED ORCHARDS

The second genetic roguing of the 1978 black spruce (*Picea mariana* (Mill.) B.S.P.) orchard (2.8 ha) was done in 1992 based on 14-year measurements collected in the fall of 1991 from family tests. Due to adequate seed in storage the only seed collected from black spruce over the past two years was during a partial collection in 1991.

#### CLONAL SEED ORCHARDS

The establishment of 3.6 ha of first generation white spruce (*Picea glauca* (Moench) Voss) clonal orchard is completed. The first cone crop was collected in 1990 and yielded 0.5 kg of seed. The harvest in 1992 yielded 1.3 kg. Topping of the ramets was done in 1993 to control height growth and gibberellin  $A_{4/7}$  was applied as a stem injection to stimulate cone production on the larger ramets.

The establishment of 1.5 ha of second generation black spruce clonal orchard comprised of 1300 ramets is also complete.

#### TESTING

In cooperation with the New Brunswick Tree Improvement Council (NBTIC), polycross matings of second generation black spruce selections are continuing in family tests established by Fraser. All selections made during 1989–91 have been mated to produce progeny test stock.

During 1992 and 1993 four tests were established, one black spruce and two white spruce progeny tests and one black spruce realized gain test. Realized gain tests are established to determine if the gain predicted from the use of orchard seed relative to unimproved sources will be achieved. This brings our total established test area for tree improvement to 48 ha.

#### COOPERATIVE PROJECTS

Cooperative projects in relation to orchard management are continuing. Areas addressed include cone and seed pests, variation patterns in wood characteristics of black spruce families, seed production potential and optimal time for pollination in black spruce, pollen monitoring in a black spruce orchard, site and family effects on seed quality in black spruce, and panmictic assessment of a white spruce clonal orchard.

Also, assistance and a location was provided to Forestry Canada-Maritimes to establish a white spruce clonal test.

#### PUBLICATIONS

Caron, G.E. and R. LeBlanc. 1992. Pollen contamination in a small black spruce seedling seed orchard for 3 consecutive years. For. Ecol. & Manage. 53: 245–261.

## TREE IMPROVEMENT AND RELATED STUDIES AT THE UNIVERSITÉ DE MONCTON

#### Guy E. Caron

## École de sciences forestières Université de Moncton, C.U.S.L.M. 165 boulevard Hébert Edmundston, New Brunswick E3V 2S8

## Keywords: Picea mariana, Picea glauca, Pinus strobus, full-seed yield, pollen contamination, proliferation, bisexuality, family and site effect.

This report summarizes on-going and completed research activities related to tree improvement and seed quality at the Université de Moncton over the past two years. These activities include: i) site and family effect on black spruce (*Picea mariana* (Mill.) B.S.P.) seed quality, ii) timing of pollen application to maximize black spruce full-seed yield, iii) survey of proliferated seed cones and pollen cones, and hermaphroditic strobili in black spruce orchards, iv) pollen monitoring in a number of orchards, and v) wood specific gravity in black spruce half-sib families.

#### SEED QUALITY AND QUANTITY STUDIES

Site and family effect on seed germination and rate of germination was investigated for black spruce (*Picea mariana* (Mill.) B.S.P.) half-sib families in three New Brunswick orchards (see Caron 1991). In addition, seed germination and seedling growth for the same material was determined in the nursery (Bouchard 1992, Bouchard and Caron 1992). An expanded study to relate seed quality to site and family effect was initiated in 1992. Mr. Emanuel Sildor, a M.Sc. student, selected families common to five black spruce seedling seed orchards in the Maritimes.

Pollen applied to receptive seed cones appears to maximize full-seed yield when applied 4-6 days after the start of receptivity. However, important variation exists among trees within half-sib families (Lévesque 1992). Further studies to better define timing for controlled pollination in black spruce are underway.

#### PROLIFERATION AND BISEXUALITY IN BLACK SPRUCE

Presence of proliferations (a combination of needles and bracts on the same structure; see Caron and Powell 1991) and bisexual strobili has been on-going since 1987 in two black spruce orchards. Results indicate that the number of proliferations and bisexual strobili fluctuates annually. Full-seed yield from seed-cone proliferations was investigated (Thériault 1993). This latter study indicated that, unlike previous reports, full-seed yield can be obtained from proliferated seed cones. Trees bearing bisexual strobili tend to bear them repeatedly, and this occurs most often in years of heavy seed-cone and pollen-cone production.

### POLLEN MONITORING STUDIES

Pollen monitoring in a white spruce (*Picea glauca* (Moench) Voss) clonal orchard and a black spruce seedling seed orchard was conducted in 1991 (Caron et al. 1992) and 1992 at St. Elzéar, Quebec, with the Quebec Ministry of Forests. A white spruce clonal orchard, located amidst a large burnover area near La Malbaie, Quebec, was monitored for the first time in 1992 and a second time in 1993. An eastern white pine (*Pinus strobus* L.) clonal orchard located at Fort Coulonge, Quebec, was monitored in 1992 and 1993 to verify influx of potential pollen contaminants. Orchard pollen contamination in a small black spruce seedling seed orchard at Second Falls, N.B. (see Caron and LeBlanc 1992), was evaluated for a fourth time in 1992 (Ritchie 1993; Ritchie and Caron 1993).

#### WOOD SPECIFIC GRAVITY

A study to investigate within family wood specific gravity in black spruce was initiated in 1992 in collaboration with Dr. Kris Morgenstern of the University of New Brunswick and Mr. Ray LeBlanc of Fraser Inc. Early results indicate that certain families had both rapid growth and high specific gravity (Plourde 1993).

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## TREE IMPROVEMENT AND RELATED STUDIES AT THE UNIVERSITY OF NEW BRUNSWICK

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#### GENETIC VARIATION AND PARAMETERS

Several studies or projects in the genus *Larix* were completed. Eight New Brunswick populations of *Larix laricina* (Du Roi) K. Koch sampled by isoenzyme methods revealed large within-population variability. Only 3.8% of total genetic variation resided among populations (Ying and Morgenstern 1991). A comparison of species with several provenances each in the two blocks of our Larch Arboretum gave a general idea of their phenology and growth. *Larix sibirica* Ledeb., *Larix occidentalis* Nutt., *Larix gmelini* (Rupr.) Kuzeneva, *Larix gmelini* var. *olgensis* (Henry) Ostenf. and Larsen generally initiate growth too early in spring and suffer from spring frost, but also stop growth too early in summer. *Larix decidua* Mill., *Larix eurolepis* Henry and *Larix kaempferi* (Lamb.) Carr. and *L. laricina* are much better adapted to the growing season and grow well (Carswell 1992, Carswell and Morgenstern 1992). Full-sib families produced in 1989 by controlled crosses (mainly *L. eurolepis* but also *L. sibirica* x *decidua* and *L. sibirica* x *kaempferi*) in cooperation with Dr. D. Fowler, were established in three locations in 1991 with help from Scott Canadian Timberlands, J.D. Irving Ltd., and Forestry Canada.

Breeding options for *Picea mariana* (Mill.) B.S.P. were explored. The advantages of a method to derive additive, dominance, and epistatic genetic variances from a clonally replicated test of full-sib families were demonstrated (Mullin et al. 1992). A breeding strategy based on clonal propagation via somatic embryogenesis was discussed by Morgenstern and Park (1991) at a IUFRO meeting in Finland.

#### CONE AND TREE-CROWN DEVELOPMENT

Pollen-cone and seed-cone development in *L. laricina* was described, and the pollination mechanism detailed (Powell and Tosh 1991). Sizes of pollen grains varied widely by clone in *Picea glauca* (Moench) Voss and *P. mariana* (Luke 1992). This will affect proportional representation of pollen of different clones in polymixes formed volumetrically. Seed weight varied predictably with seed position in the cone of *Abies balsamea* (L.) Mill. (Greebe 1993). Proliferated pollen and seed cones with various proportions and positioning of leaves occur widely in young *P. mariana* and suggest that the developmental pathway has some degree of flexibility (Caron and Powell 1991).

The distribution of cones on branches and along and around shoots follows patterns associated with those of branch development in young crowns of *P. mariana* (Caron and Powell 1992; 1993). Documentation of such patterns is a key to elucidating physiological factors of cone differentiation, and to devising rational crown-management techniques for sustaining cone production on modified crowns (Powell 1991a). Patterns of shoot-extension, branching on

#### TREE PHYSIOLOGY

Regulation of wood formation at the biochemical level continues to be the focus of the physiology research program. Coniferin was qualitatively and quantitatively correlated to cambial growth phenology in *L. laricina*. Neither auxin (indol-3-ylacetic acid, IAA) nor photosynthate (sucrose) contents of cambium varied in parallel with coniferin, indicating that coniferin biosynthesis must be regulated by factors other than auxin or photosynthate availability (Savidge 1991). Protoplasts from cambial ray and fusiform cells of *Pinus banksiana* Lamb. and *Pinus strobus* L. were obtained and their coniferin contents were compared with those of the tissues from which the protoplasts were derived. The data suggest that coniferin conferin concentrations are minor (Leinhos and Savidge 1993). An ultrastructural study of induced tracheid differentiation in *Pinus contorta* Dougl. elucidated protoplasmic changes associated with secondary-wall deposition. Two notable observations were that microtubules appear to be unnecessary for microfibril deposition and that electron-dense exocytotic blebs are specific to and evidently essential for wall growth (Savidge and Barnett 1993).

In vivo cambial growth of L. laricina was simulated in vitro on a fully defined growth medium (Savidge 1993b). 'Chips' from 8-year-old stem regions comprising cambium sandwiched between phloem and xylem were cultured xylem-surface down on agar media. Wood formation in this system required the presence of a critical concentration of auxin (l-naphthalene acetic acid), and  $[NO_3^{-3}]$  was identified as a factor affecting cambial growth. The radial increments achieved were modest and entirely earlywood like; sub-culturing did not support additional growth.

A new enzyme of lignification, coniferyl alcohol oxidase, was discovered in *A*. *balsamea*, *L*. *laricina*, *Picea rubens* Sarg., *P. banksiana* and *P. strobus* (Savidge 1992, Savidge and Udagama-Randeniya 1992).

Diameter growth in trees has again been reviewed (Savidge 1993a).

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## FOREST GENETICS RESEARCH AT FORESTRY CANADA - MARITIMES REGION

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Forest genetics work at Forestry Canada - Maritimes Region (FC-MR) continued in four major research areas: (1) genetic studies of important tree species; (2) operational research relating seed production and vegetative propagation systems; (3) biotechnology involving tissue culture and genetic engineering; and (4) biodiversity.

In November 1992, the project was peer reviewed and reorganized. Messrs. R.D. Hallett and S.I. Cameron, formerly with the Seedling Culture project, joined the project. Also, within the project, a "Somatic Embryogenesis (SE) Working Group" was formed to bridge the gaps between the advances in SE techniques and operational application.

Forestry Canada - Maritimes Region provides technical coordination and direction in the operation of the New Brunswick Tree Improvement Council (NBTIC), which completed its 16th year of operation. NBTIC is directly responsible for all operational aspects of the program. Progress of NBTIC is reported separately.

Dr. D.P. Fowler completed his assignment with FC-MR as a one-third-time research scientist. His outstanding contribution to Forest Genetics is well recognized, and he has been named an Emeritus Research Scientist. It is expected that Dr. Fowler will continue to contribute to the project.

#### GENETIC STUDIES OF IMPORTANT TREE SPECIES AND TREE BREEDING STRATEGIES

Most of the "species and provenance trials" and "population genetic studies" have been completed after receiving final assessments and publication of results. These test plantations will be maintained as a diverse source of material for future breeding work. Currently, rangewide provenance tests of black spruce and tamarack are considered to be active experiments. Data on 16-year performance of black spruce and 5-year height of tamarack are being evaluated.

Over the past several years, tree improvement strategies using clonal approaches as an alternative to conventional seed orchard breeding have been emphasized. The implementation of an effective clonal breeding strategy requires accurate genetic information at different levels, i.e., population, family, individual tree, and clones. To provide a means of obtaining such information, the genetic testing procedures have been modified by the use of clonal replicates, which enables one to obtain estimates of additive, dominance, and epistatic variances. During 1991-92, a series of clonally replicated tests of white spruce has been established at three locations. The material for the tests was derived by using two cycles of rooted cuttings representing 80 full-sib families resulting from disconnected diallel crosses of NBTIC's second-generation breeding population.

Three possible "breeding-cloning" options incorporating vegetative multiplication and breeding hall technology for advanced generation are proposed: "Backward GCA selection and polycrossing", "Backward SCA selection and repeat-crossing" and "Forward clonal selection" (Mullin and Park 1992, Mullin et al. 1992, Morgenstern and Park 1991).

#### CONE AND SEED RESEARCH/SEED ORCHARD MANAGEMENT

FC-MR staff from both Forest Genetics and the Forest Insect and Disease Survey continue to provide assistance to orchard managers in the Region through technology transfer, and by conducting and coordinating operational, problem-oriented research trials. Many of the cooperators are now actively conducting their own operational research trials. In 1992, a cooperative project began to test the feasibility and economic viability of miniaturized seed orchards in Norway spruce, jack pine, and hybrid larch.

The cooperative cone induction and topping trials established 5 years ago have essentially been completed. The results from these trials are being used in many orchards, e.g., removing 2-years' growth every second year is becoming an operational practice in many spruce orchards.

The strong cooperative infrastructure among orchards in the Region has allowed for a shift in program emphasis towards more basic research, specifically in the field of flowering physiology. Several studies have recently been initiated including examining the effects of gibberellin, cytokinins, and adjunct cone induction treatments on photosynthesis, stomatal conductance and cell division within developing buds. This work is part of Ron Smith's PhD program.

#### SOMATIC EMBRYOGENESIS OF WHITE SPRUCE

Since 1991, an extensive experiment was conducted to determine the degree of genetic control and the effects of cultural treatments on SE with 30 full-sib families derived from six-parent diallel crosses, including reciprocals. Thirty zygotic embryos, from both immature and mature cones, were cultured in media with either 2,4-D or Picloram immediately after collection of cones and after 2 months of cold storage. A total 5,572 explants were used in the experiment.

There were significant differences in SE initiation between immature and mature explants, and fresh and cold-stored seeds, but there were no significant culture media effects. Significant variances due to families and family x treatment interactions were found. The mean percentages of explants that initiated SE in each family ranged from 3.3 to 54.6%, with an overall average of 30.5%.

The partitioning of family variances revealed that 21.7% was due to general combining ability, 3.5% was due to maternal effects and 5.5% was due to reciprocal effects; however, the specific combining ability was negligible. Variance due to interactions of family x treatments collectively accounted for 32.6%, while the remaining 37.8% of variation was accounted for by random error. When comparing the responses obtained using immature explants with mature ones, there were drastic changes in composition of genetic variances, which indicate that we are dealing with rapid shifts in the activity of several genes. However, while the

dominance variance may be transitional, the additive gene effect maintains an important role in initiating SE.

It appears that SE is a promising tool for vegetative multiplication of genetically improved white spruce because SE was obtained from all the full-sib families in relatively high percentages. Furthermore, since the variation in SE was under strong genetic control, it can be manipulated into breeding schemes. In conjunction with cryopreservation, SE will have a great impact on implementing clonal breeding strategies (Park et al. 1993)

Results from the maturation, germination, and cryopreservation phase of the experiments are currently being compiled. With extensive genetic information on all phases of SE and relatively high initiation rate, the potential for industrial application is greatly enhanced. Therefore, we have formed a SE Working Group, consisting of Dr. J.M. Bonga (tissue culture), Dr. Y.S. Park (genetics), Mr. S.I. Cameron (physiology) and Mr. R.D. Hallett (silviculture), to increase our capacity for mass production for potential industrial application. Research is being carried out to improve protocols and for development of an efficient and partially automated "embling" production system. Field testing of emblings in comparison with seedlings and stecklings is planned. To date, about 500 emblings have been field planted.

Micropropagation of mature *Larix* continued. To date, SE in conifers has been obtained only in tissues from highly juvenile donors (zygotic embryos or seedlings up to a few weeks old). In our efforts to micropropagate 30-year-old *Larix decidua* trees, we have, over the last 5 years, routinely obtained a few cultures that are displaying the early stages of SE. The most critical factor in this process is the collection date of the shoot buds placed in culture. Only buds collected in early April are responsive. We have also determined that Picloram stimulates the process. A histological study of this material has been completed. The results of this study are being prepared for publication.

#### HORMONAL CONTROL OF WOOD FORMATION AND GENETIC ENGINEERING

Research on the hormonal control of wood formation and the enhancement of wood quantity and quality using genetic engineering was continued in collaboration with personnel at the Swedish University of Agricultural Sciences, Umeå, Sweden, and the University of New Brunswick, Fredericton.

It was demonstrated in *Pinus sylvestris* that the cambium's ability to respond to exogenous indole-3-acetic acid (IAA) was qualitatively the same in 1-year-old and older stem internodes, however with increasing internode age there was a decrease both in the number of tracheid produced by a particular IAA concentration and in the optimal IAA concentration for inducing tracheid production (Little and Sundberg 1991; Cui et al. 1992). Treating an intact stem with IAA increased tracheid production, indicating that the level of endogenous IAA in the cambial region is less than optimal for producing wood. The endogenous free IAA concentration in the cambial region was observed to vary radially and seasonally, and endogenous IAA conjugates (both amides and esters) were detected (Sundberg et al. 1991). Applying gibberellin  $A_{4/7}$  to seedlings increased tracheid production, longitudinal growth and the cambial region concentrations of IAA and gibberellins  $A_4$ ,  $A_7$  and  $A_9$  in the current-year leader (Wang et al. 1992).

In Abies balsamea, it was observed that cambial frost hardiness, IAA-induced cambial cell cycling, and the IAA-induced xylem to phloem ratio varied independently with season, temperature and photoperiod (Mellerowicz et al. 1992a). The seasonal variation in the cambial growth response to IAA and in cambial frost hardiness was shown not to be specifically

related to changes in the nuclear genome size of fusiform cambial cells, determined cytophotometrically (Mellerowicz et al. 1992b).

Measurement of the activity of five enzymes involved in lignification indicated that post-harvest lignin synthesis in etiolated asparagus spears is associated primarily with an increase in the activity of syringaldazine oxidase (SyrOx), a peroxidase. Other evidence suggested that ethylene is involved in the control of SyrOx activity (Hennion et al. 1992).

Genetic engineering experiments showed that the IAA concentration was elevated in *Nicotiana tabacum* plants transformed with the *Agrobacterium tumefaciens* T-DNA i*aaM* and *iaaH* genes and that varying the promoter used to express each of these genes altered the IAA level measured in a particular organ (Sitbon et al. 1992a,b). Finally, a non-destructive technique was developed for measuring light emission from the leaves of hybrid *Populus* plants transformed with a 35S promoter-luciferase gene fusion. It was used to demonstrate that promoter activity in a particular leaf can be monitored repeatedly over time (Nilsson et al. 1992).

#### BIODIVERSITY

Biodiversity is a new research area for the FC-MR's Forest Genetics project. Dr. Judy Loo-Dinkins has been establishing a research program around the genetic aspects of biodiversity with studies comparing genetic diversity of forest tree species after different kinds of disturbance and a comparison of genetic diversity levels in several tree species from different parts of the Maritimes Region. In addition, work is being undertaken to support and enhance the selection of potential forested protected natural areas for conservation purposes.

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#### NEW BRUNSWICK TREE IMPROVEMENT COUNCIL

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Technical coordination and direction of the operations of the New Brunswick Tree Improvement Council (NBTIC) is provided by Forestry Canada. Member agencies are continuing to provide funding for a full-time data analyst, Dave Steeves. NBTIC completed its 16<sup>th</sup> year of operation in 1992. Efforts are concentrating on selecting, breeding, and testing second generation black spruce (*Picea mariana* [Mill.] B.S.P.) and jack pine (*Pinus banksiana* Lamb.) and breeding and testing first generation white spruce (*Picea glauca* [Moench] Voss.) and tamarack (*Larix laricina* [Mill.] Karst). Establishment of second generation seed orchards and management of first generation orchards are also important program components.

### SEED ORCHARDS AND SEED PRODUCTION

Establishment of first generation seedling and clonal seed orchards was completed in 1987 and 1991, respectively. Planting of second generation clonal orchards started in 1989 and there are currently about 9 ha each of black spruce and jack pine. For these species the phenotypically best tree from each of the 40 best performing families from the three oldest series of family tests is clonally propagated by grafting. Clone placement in the orchards is determined by a computer algorithm utilizing the permutated neighborhood design concept. Planting spacing ranges from 2 x 5 m to 3 x 6 m.

Seed production from seed orchards has been beyond our expectations. Seedling seed orchards of black spruce and jack pine have produced abundant, annual crops without, for the most part, having to utilize any cone stimulation techniques. At least 100 million seed has been collected annually the last several years. The first seed was collected in 1992 from a 2<sup>nd</sup> generation jack pine orchard! All seedlings for reforestation of the four species are grown from genetically improved seed. Practically all black spruce and jack pine seed is collected from seedling seed orchards which have been rogued once.

#### TESTING

Establishment of tests has been an integral part of the Council's program. Over the past 16 years, 177 tests have been planted on 230 ha. These tests are yielding a wealth of information used to rogue seedling seed orchards, to estimate genetic parameters, and identify families and trees for second generation selection.

Analysis of the 7-year and 10-year measurements of first generation half-sib jack pine and black spruce family tests, respectively, for second generation selection and first generation seedling seed orchard roguing continues. The final series of jack pine tests should be complete this year. Eight black spruce test series remain to be measured and analysis of these is scheduled for completion by 1997. Height heritabilities at a given age have been quite consistent among series for both species, particularly in light of differences in the numbers and locations of environments sampled. "Genotype x environment" interactions were inconsequential at best and easily managed by removing two or three "unstable" families at worst. A verage individual tree and family narrow-sense heritabilities for 7-year and 10-year heights of jack pine and black spruce, respectively, are presented in Table 1. In some of the older series, data have been collected at ages ranging from five years to 14 years. Family age-age height correlation coefficients ranged from 0.80 to 0.95 for both species.

Breast height diameters were measured in several of the family test series of both species and individual total tree volumes were estimated using standard volume equations developed for these species in central and eastern Canada. Correlation coefficients between family height and volume means range from 0.83 to 0.92 for jack pine and between 0.90 and 0.95 for black spruce. Tree volume has been used for the second roguing of older jack pine and black spruce seed orchards. It tends, however, to be less heritable and fails to take stem form into account.

The seven year old jack pine test trees were graded for straightness by assigning a score from "1" (crooked) to "6" (straight). This trait has proven to be quite heritable at the family level ( $\overline{h}^2_f = 0.64$ ) and in all cases is negatively correlated, both phenotypically and genetically, to height. Both independent culling and index selection have been used to select simultaneously for these two traits.

Polycross progeny tests are being established for all four species. The experimental design of polycross tests is two-tree plots randomly planted in each of 15 replications at 4 to 5 sites throughout the province. Seedlings are planted at a 2 x 2 m spacing. Four to five month old greenhouse container seedlings are used. These tests are scheduled for first measurement at age 5. Pair-mate tests or selection plantations are planted such that each family is replicated once on a site in a 48-tree plot.

rable I.	Average individual free and family narrow-sense heritabilities with standard deviations for
	7-year heights of six jack pine family test series and 10-year height of five black spruce test
	series.

Species	$\overline{h}_{i}^{2}$	s.d.	$\overline{h}^2_{f}$	s.d.
Jack pine	0.26	0.07	0.82	0.05
Black spruce	0.26	0.03	0.79	0.04

#### SELECTION

Selection of trees for second generation breeding populations is based on 10-year family test data for black spruce and 7-year data for jack pine. Top performing black spruce families are identified based on height growth while for jack pine stem straightness and height are used. Candidate trees for both species are initially selected based on height growth, phenotypically graded in the family tests, and the final selection made from each family based on a combination of superior phenotypic and metric traits. The goal is to form breeding populations containing 400 trees. Family and within-family selection intensity depends on total number of families tested and number of trees tested per family. Family selection intensities are 1 in 3 and 1 in 2 for black spruce and jack pine, respectively and within-family selection intensity ranges from 1 in 60 to 1 in 160. A total of 174 black spruce and 299 jack pine trees has been selected. The remaining number of jack pine will be selected over the next two years.

#### BREEDING

The Council conducts a complimentary breeding program. The polycross, consisting of a mix of 20 unrelated pollens, is used to determine breeding value. Pair-mating, involving specific crosses among pairs of selected trees, is done to produce material from which two selections will be made for the next generation. Excellent progress has been made in polycrossing first generation white spruce and tamarack with 62% and 69%, respectively, of the clones crossed. Forty three percent of white spruce pair-matings is also complete. Due to declining interest in planting and managing tamarack, polycrossing will be the only breeding conducted. Polycrossing of second generation black spruce and jack pine selections has, for the most part, taken place *in situ* in the family tests. The purpose of this is to enable the establishment of progeny tests 3 to 4 years sooner than would be possible with intensive growth and culture of grafts. Ramets from trees not producing sufficient seed will be bred in a clone bank or breeding hall. Polycrossing has been completed on 85% of the black spruce and 14% of the jack pine selected to date. Pair-mating will be conducted in an assortative manner based on results from early tests.

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#### TREE IMPROVEMENT PROGRESS IN NEW BRUNSWICK

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Keywords: Accelerated breeding, cone collection, cross-pollinations, flower induction, progeny tests.

The tree improvement program in N.B. is progressing smoothly. Our efforts continue to be focused on our 4 major reforestation species, black spruce (*Picea mariana* (Mill.) B.S.P.), jack pine (*Pinus banksiana* Lamb.), white spruce (*Picea glauca* (Moench) Voss) and tamarack (*Larix laricina* (Du Roi) K. Koch.). Secondary species for Tree Improvement include balsam fir (*Abies balsamea* (L.) Mill.), and Norway spruce (*Picea abies* (L.) Karst).

All first generation selection and seed orchard establishment has been completed for the four major tree species. Second generation selections, cross-pollinations and orchard establishment are well underway. The following report highlights our Tree Improvement activities for the past two years.

#### TREE BREEDING/TESTING

The 1991 and 1992 black spruce polycrosses done on second generation selections provided sufficient seedlots to grow and outplant the second series of black spruce progeny tests. The polycross seedlots included those from NBTIC members, totalling 35 families. In the past, all crosses on jack pine were carried out in the NBTIC family tests, however, the trees have now reached sufficient size to enable crosses to be done in the breeding garden. Black spruce crosses are still being carried out in family tests. A third series of white spruce progeny tests were also established in 1992. Breeding on white spruce is now 62% complete, while tamarack is 69% complete.

#### SEED ORCHARDS

The best individuals selected from the best families in NBTIC (New Brunswick Tree Improvement Council) family tests are being identified for second generation material. To date, we are 44% complete for black spruce selections and 75% complete for jack pine. A total of 2 ha each of second generation jack pine orchard and second generation black spruce orchard has been established at the Kingsclear Nursery.

#### CONE COLLECTION IN SEED ORCHARDS

Both the clonal and seedling orchards have started to produce substantial amounts of seed (Table 1). The black spruce, white spruce and jack pine orchards are producing enough seed to meet our annual reforestation requirements with improved seed.

Seed yield from the white spruce clonal orchard was phenomenal in 1992 (Table 1). The tamarack clonal orchard produced some seed in 1992, although production is low, primarily as a result of larch cone maggot infestations.

		Cones (1)		Seed (kg)	
Species	Seed Orchard	1991	1992	1991	1992
Black Spruce SSO	Bettsburg Pokiok	4 454 803	5 186 5 971	21.8 3.1	14.3 34.2
Jack Pine SSO	Otter Brook	6 928	3 560	62.6	20.9
White Spruce CSO	Queensbury	219	3 621	2.5	57.3
Tamarack CSO	Queensbury	-	354	-	0.8

Table 1. Cone collection and seed yield from orchards in 1991 and 1992.

#### ACCELERATED BREEDING PROGRAM

Gibberellin A  $_{4/7}$  was applied by stem injections to white spruce grafts for flower induction in June of 1992. The treated grafts had not yet produced the females necessary to complete the first generation breeding program. Only 27% of these grafts produced enough flowers to breed this spring. In general, flower production in all conifer species in our program was very low this year, regardless of hormone injections.

Another trial was undertaken in June of 1993. Potted black spruce second generation selections, as well as unco-operative white spruce and tamarack clones in the first generation selections were injected. This will enable us to finish up a good portion of the first generation breeding in 1994.

## **COOPERATIVE TREE IMPROVEMENT IN NOVA SCOTIA**

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## Keywords: Picea mariana, P. glauca, P. rubens, P. abies, Pinus strobus, seed orchards, flower induction, breeding.

The Tree Improvement Working Group (TIWG) was established in 1977, and is the coordinating body for tree improvement in Nova Scotia. Active members include both Federal and Provincial Governments, Bowater Mersey Paper Company Limited, Scott Worldwide Inc., Stora Forest Industries, and J.D. Irving Limited. Meetings are held in the spring and fall of each year to review progress and plans, while day-to-day activities are handled by the Department of Natural Resources. Species of interest include *Picea mariana*, *P. glauca*, *P. rubens*, *P. abies*, and *Pinus strobus*.

#### ORCHARD ESTABLISHMENT

First generation orchard establishment was completed with the planting of 2000 Norway spruce trees in May 1993. Total orchard area for the five species we work with are as follows:

Species	Location (Managing Agency)	Туре*	Approximate area (ha)	1992 Seed Prod. (kg)
Black Spruce	Fast Mines (Scott)	S	64	11 5
Diach oprace	Aldershot (Stora)	Š	71	26
	Debert (DNR)	s	3.1	-
White Spruce	Debert (DNR)	С	8.6	22.0
•	East Mines (Scott)	С	5.6	26.6
	Waterville (Stora)	С	5.4	4.8
Red Spruce	Melvern Square (Bowater)	С	4.3	2.2
•	Waterville (Stora)	С	5.3	5.4
	Lawrencetown (DNR)	С	1.6	-
	Debert (DNR)	С	7.9	0.5
	Sussex, N.B. (Irving)	С	2.4	-
White Pine	Debert (DNR)	С	1.8	0.9
Norway Spruce	East Mines (Scott)	С	4.6	-
- •	Debert (DNR)	С	11.7	6.0
TOTALS 75.8 82.5				82.5

\* S = Seedlings, C = Clonal (Grafted)

The above table also displays 1992 seed production; excluding Norway spruce. Eighty percent of the 1993 conifer crops sown in provincial nurseries utilized orchard produced seed.

#### TESTING

In the spring of 1992 and 1993 both white and red spruce polycross tests were again established following good flower crops in preceding years. Norway spruce testing is still hampered by inconsistent flowering and a high susceptibility to late spring frosts, application of  $GA_{4/7}$  for flower induction should help overcome the inconsistent flowering problem. An open-pollinated white pine family test was established in 1992 and 1993 to evaluate genetic variation within this species.

#### BREEDING

The general strategy for white and red spruce consists of polycrossing to determine GCA and a series of paired matings from which second generation selections will be made. A summary of breeding to date follows:

Species	Cross	Target	To Date	% Complete
wS	poly	466	393	84
wS	specific	466	146	31
	-			
rS	poly	<b>499</b>	207	42
rS	specific	250	100	40

During the winter of 1993, second-generation black spruce material was selected from within our 10 year old family tests. These selections have been grafted and will form a portion of our second generation black spruce orchard and clonebank.

#### FLOWER INDUCTION

Stem injections of  $GA_{4/7}$  are being used in soil-based clone banks to stimulate those clones which have shown poor flower production to date, application rates are dependent upon tree size. A trial in our Norway spruce orchard should help refine a rate suitable for general application which will increase the orchards overall seed production. Another trial dealing with the topping of our larger Norway spruce trees will assess the feasibility of controlling the size of these trees and 'topping' effect on cone production.

#### MINIATURIZED SEED ORCHARD

In cooperation with Forestry Canada, a non-conventional Norway spruce seed orchard was initiated. This orchard will test the effectiveness of intensively managing a smaller number of ramets as well as clones in contrast to our conventional orchard which utilizes a high number of clones and a high number of ramets per clone. The miniaturized orchard will incorporate collecting and extracting all pollen from within the orchard and using a mass pollination technique to breed the female flowers with unrelated pollen.

## TREE IMPROVEMENT CONSULTING SERVICES OFFERED IN THE MARITIMES

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## Keywords: Breeding strategy, China, clonal forestry, computer simulation, economic analysis, seed procurement.

The writer has offered independent consulting services since the fall of 1990, specializing in biological and management aspects of forest renewal. This report summarizes the major contracts, completed or in-progress, which are related to tree improvement and forest genetics.

#### **REVISION OF SEED ZONES IN ALBERTA**

Data analysis services were provided under subcontract to the University of New Brunswick to evaluate provenance variation in Alberta as a basis for revision of seed zones (Morgenstern et al. 1991, 1992a, b). Data were available from test installations of white spruce (*Picea glauca* [Moench] Voss), lodgepole pine (*Pinus contorta* var. *latifolia* Engelm.), and jack pine (*Pinus banksiana* Lamb.). In addition to standard analyses of variance and correlation with environmental variables, the standardized data were subjected to a multiple regression procedure to derive an adaptation index value based on geographic location and elevation. Seed zone revision will probably incorporate response gradients from the regressions together with ecoclimatic data.

#### GENETIC PARAMETERS FROM CLONAL TESTS

Procedures for estimation of genetic parameters from clonal tests, described originally as part of the writer's doctoral thesis, were prepared for journal publication (Mullin and Park 1992). These procedures were applied to the analysis of a five-year-old black spruce (*Picea mariana* [Mill.] B.S.P.) progeny test with clonal replicates, established at four locations (Mullin et al. 1992). The 10-year data from this test series will be available in the fall of 1993 and will be analyzed under a contract funded by the Canada-NB Cooperation Agreement for Forestry Development (CAFD).

#### SEEDLING AND CLONAL OPTIONS FOR REFORESTATION

A project funded by the Canada-NB CAFD investigated factors that should be considered in the evaluation of clonal propagation as a breeding tool and deployment method for reforestation. Comparisons of gains were made by means of a stochastic computer simulation model, and a financial analysis was made. It was concluded that clonal forestry is worthwhile pursuing if allowable-cut effects are available. Even in the absence of allowable-cut effects, clonal forestry should be viable at low to moderate rates of interest (Mullin 1992a).

#### MANAGEMENT OF GENETIC DIVERSITY IN TREE BREEDING PROGRAMS

A three-year project under the Canada-NS CAFD was initiated to study the joint management of tree breeding populations for genetic gain and maintenance of genetic diversity. The approach adopted was to investigate strategy options by means of computer simulation. A stochastic simulation program has been written for use on personal computers to predict the effect of population management, breeding and selection on genetic variance structure and effective population size. An extensive series of scenarios is being simulated in an attempt to describe population management strategies that will resolve the conflict between maximizing genetic gain and maintaining adequate levels of genetic diversity.

#### GANSU FOREST TREE NURSERY PROJECT, CHINA

Starting in 1992, consulting services were retained over a five-year period by ROCHE Ltée, Canadian Executing Agency for the Gansu Forest Tree Nursery Project, funded by the Canadian International Development Agency. These services include advice on procurement and handling of seed, training on seed biology, seedling physiology and vegetative propagation. On-site missions to the Longnan Prefecture of Gansu Province are conducted to evaluate current practices and provide recommendations for seed procurement, storage and testing, and tree improvement, and to deal with on-going problems with seed quality and seedling physiology encountered during nursery production. In addition, a PhD training program for the Chinese Laboratory Director has been arranged with the Chinese Academy of Forestry, with cosupervision and in-Canada training provided in collaboration with the Faculty of Forestry at the University of New Brunswick.

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# TREE IMPROVEMENT PROGRESS IN P.E.I.

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Keywords: Miniaturized seed orchard, polycross, flower induction, provenance trials, density trial, gene pool conservation.

This progress report notes the P.E.I. Forestry Branch's tree improvement activities during 1992 and 1993. The Forestry Branch has concentrated primarily on four major reforestation species: black spruce (*Picea mariana* (mill.) B.S.P.), white spruce (*Picea glauca* (Moench) Voss), eastern larch (*Larix laricina* (Du Roi) K. Koch.) and white pine (*Pinus strobus* L.). Secondary species include red spruce (*Picea rubens* Sarg.), balsam fir (*Abies balsamea* (L.) Mill.), yellow birch (*Betula alleghaniensis* Britton.), red oak (*Quercus rubra* L.), white ash (*Fraxinus americana* L.), black ash (*Fraxinus nigra*, Marsh) and hop-hornbeam (*Ostrya virginiana* (Mill.) K. Koch).

First generation orchards have been completed for our major reforestation species. Orchard establishment for red spruce and balsam fir are 60 percent and 50 percent complete respectively.

#### PLUS TREE SELECTION

In 1993 twenty-five new white pine selections were made and grafted for the establishment in a miniaturized seed orchard. Second generation selections from the best families in NBTIC (New Brunswick Tree Improvement Council) black spruce family tests in P.E.I. may be carried out in 1994.

#### TREE BREEDING

In 1992, white spruce polycrosses were carried out on 35 of 80 families. There were no female flowers produced in 1993.

#### CONE COLLECTION IN SEED ORCHARDS

All of the orchards except balsam fir have produced seed. The black spruce seedling seed orchard has been in full production since 1988 producing much more seed than our annual requirements. The next nursery crop of eastern larch will be of orchard origin. The remaining species except balsam fir should provide adequate seed requirements within a few years.

## FLOWER INDUCTION

In June of 1992, one third of an eastern larch clonal orchard had Gibberellin  $(GA_{4/7})$  applied by stem injection together with monthly fertilizer application. Results thus far are very confusing with only one portion of the treated area reacting to the injection. Male production was much lower than female flowers in the treated area.

#### PROVENANCE TRIALS/RESEARCH PLANTING

In 1993 two provenance trials were established. The species used were Scots pine (*Pinus sylvestris* L.) and Norway spruce (*Picea abies* (L.) Karst) each with nine provenances. A larch species trial was planted as well as observational plantings of red spruce. In cooperation with Forestry Canada -Maritimes Region, a hybrid larch test was also established.

#### HARDWOOD RESEARCH - GENE POOL CONSERVATION

Tree improvement hardwood research concentrates primarily on yellow birch, red oak and white ash. Gene pool conservation is underway with hop-hornbeam and black ash.

In 1992 tree improvement established several yellow birch density trials. These trials contain four densities which were replicated three times on each site. Plot size was 10 ha for a total of 1.20 ha for each of the sites. The 1993 trials consist of red oak and yellow birch. White ash density trials will be established in 1994.

# AMÉLIORATION DES ARBRES FORESTIERS À LA DIRECTION DE LA RECHERCHE DU MINISTÈRE DES FORÊTS

# Roger Beaudoin, Yves Lamontagne, Marie-Josée Mottet, André Rainville, Ante Stipanicic, Gilles Vallée, Michel Villeneuve

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## AMÉLIORATION DU PIN GRIS (PINUS BANKSIANA LAMB.) ET DU PIN DE MURRAY (P. CONTORTA GOUDL. VAR. LATIFOLIA ENGELM.) PAR ROGER BEAUDOIN

Mots-clés : Tests de descendances, tests de provenances, sélection d'arbres, éclaircie génétique, croisements dirigés.

Un test de descendances sur *P. banksiana* a été établi en 1991 et 4 autres en 1992 pour un total de 86 000 plants. À ce jour, le nombre de tests de descendances de pin gris établis en rapport avec les 12 vergers à graines de semis est de 21.

Une éclaircie génétique a été réalisée dans un verger à graines de pin gris (Duverny) en 1991 et une autre en 1992 (Chasseur). Une éclaircie génétique partielle (25 p. 100 des familles) a été réalisée en 1992 dans le verger à graines de pin gris de Harrington. L'éclaircie finale de ce verger sera pratiquée en 1994 à partir de données sur la hauteur prise dans un test de descendances âgé de 11 ans après la plantation.

En 1991, des greffons ont été prélevés sur 24 arbres dans un test de descendances de pin gris (Briand) et en 1992 sur 52 arbres dans un autre test (Chasseur) en vue de l'établissement de parcs de croisements.

Vingt-quatre croisements dirigés ont été réalisés en 1991 et 27 en 1992 sur *P. banksiana* entre les clones d'arbres-plus d'une provenance recommandée (Briand) et évalués par un processus de sélection récurrente.

En 1992, une éclaircie génétique a été pratiquée dans un test de provenances de *P. contorta* (Iles-de-la-Madeleine) et une autre dans une plantation conservatoire (New-Richmond «A») en vue de constituer une aire de production de semences améliorées pour le reboisement des régions concernées.

## SÉLECTION D'ARBRES ET ÉTABLISSEMENT DE VERGERS À GRAINES PAR YVES LAMONTAGNE

Mots-clés : Vergers à graines, sélection d'arbres, résineux, greffes, récolte de cônes.

Cette activité a pour objectif l'établissement et l'aménagement d'un réseau de vergers à graines pour les résineux qui fournira éventuellement toutes les semences améliorées génétiquement nécessaires pour le programme de reboisement du ministère des Forêts (MFO).

L'établissement du réseau provincial est complété depuis l'implantation d'un dernier verger en 1992. L'aménagement se poursuit dans tous les vergers. Depuis le début du programme, environ 20 000 arbres ont été sélectionnés et 83 vergers d'une superficie totale de 1 083,9 ha ont été établis avec 12 espèces. La production prévue est de 236 millions de plants par année. Pour accompagner ces vergers, 75 tests de descendances avec 6 espèces ont été établis. Six vergers ont déjà subi une éclaircie génétique. La récolte de semences améliorées a débuté en 1988. Jusqu'à 1992, 602,5 hl de cônes ont été récoltés.

Depuis le 1<sup>er</sup> avril 1993, l'aménagement des vergers à graines est sous la responsabilité du Service de la production de plants du MFO. Le Service de l'amélioration des arbres demeure responsable des analyses des tests de descendances et de la préparation des prescriptions relatives au martelage des familles et des descendances à éliminer des vergers.

## SÉLECTION D'ARBRES FEUILLUS POUR LEUR RÉSISTANCE AUX MALADIES PAR MARIE-JOSÉE MOTTET

Mots-clés : Populus, Betula, sélection, résistance, Septoria musiva, Hypoxylon mammatum, Nectria galligena.

Les travaux ont porté sur le peuplier et le bouleau. Depuis 1986, la méthode employée pour évaluer la sensibilité des clones et semis de peuplier au chancre septorien (*Septoria musiva*) consiste à inoculer artificiellement le pathogène en pépinière. La stabilité de la résistance des plants sélectionnés est suivie en plantation. À chaque année, l'évolution de la virulence des isolats de *Septoria musiva* est étudiée dans un dispositif spécial de 20 clones en pépinière. D'autre part, environ 500 nouveaux clones issus de semis ayant déjà subi une sélection primaire ont été inoculés en 1991. Des mesures ont été prises dans ce dispositif pendant deux années consécutives pour suivre la progression des dommages.

Les résultats d'inoculation en serre et en pépinière nous ont permis de noter des différences de sensibilité à *Hypoxylon mammatum* entre certains clones de peuplier. Par ailleurs, quelque 500 semis de peuplier de la section Leuce ont été sélectionnés à la suite d'inoculations avec *H. mammatum* en pépinière.

Concernant le bouleau, un volet sur la sélection pour la résistance au chancre nectrien a été entrepris. Une méthode d'inoculation artificielle en serre et en pépinière est à l'essai avec différents clones et provenances de bouleau jaune afin d'étudier la variabilité génétique de la sensibilité de cette essence à *Nectria galligena*, champignon responsable du chancre.

# AMÉLIORATION GÉNÉTIQUE DES FEUILLUS À BOIS NOBLE PAR ANDRÉ RAINVILLE

Mots-clés : Amélioration génétique, croisements dirigés, feuillus nobles, sélection d'arbres, greffage.

Le programme d'amélioration génétique du chêne rouge (*Quercus rubra* L.) se divise en deux volets, soit celui orienté vers le long terme et ayant pour base les tests de provenances-descendances, et l'autre à court terme basé sur les arbres-plus. Dans le premier cas, cinq tests de provenances-descendances ont été établis en 1993 à partir de diverses sources de glands de l'Ontario, du Vermont et principalement du Québec; ils sont situés dans la partie sud de la province. Chacun de ces tests sera complété en 1994 avec des provenances du nord-est des États-Unis et du Québec en grande majorité.

Le second volet du programme d'amélioration du chêne rouge se déroule simultanément au premier. Une centaine d'arbres-plus ont été sélectionnés en forêt sur des critères de qualité et greffés pour l'établissement de parcs à croisements en vue de produire des glands le plus rapidement possible pour le programme de reboisement du MFO. Ces semences seront aussi évaluées dans des tests de descendances.

Dans le programme d'amélioration du bouleau jaune (*Betula alleghaniensis* Brit.), plus de 120 arbres-plus ont été sélectionnés. Plusieurs de ces arbres ont fait l'objet de croisements dirigés intraspécifiques et interspécifiques avec *Betula papyrifiera*, *B. verrucosa* et *B. pubescens*. Très peu de semences de croisements interspécifiques ont été obtenues jusqu'à présent; les plants produits à partir de toutes les semences de croisements seront installés en tests en 1993; des observations permettront d'évaluer la valeur des croisements. Entre-temps, de nouveaux croisements ont été réalisés au printemps 1993. Ils sont principalement interprovenances et plutôt intraspécifiques qu'interspécifiques; toutes les étapes du processus de pollinisation, allant de la méthode d'extraction du pollen à l'enlèvement des sacs sur les fleurs femelles, ont été étudiées. Chez cette essence aussi, la stratégie d'amélioration axée sur les croisements interspécifiques sera réévaluée à la lumière des résultats de 1993 et d'une revue de documentation complète.

Chez le frêne d'Amérique (Fraxinus americana L.), seuls quelques arbres ont été sélectionnés jusqu'à présent et greffés en vue de constituer des parcs à croisements, mais la quantité de matériel est encore très faible. À l'automne 1993, des demandes seront faites à des collègues d'autres provinces du Canada et des États-Unis afin de récolter des lots de semences pour constituer des tests de provenances-descendances au même titre que le chêne rouge.

Finalement, des efforts d'amélioration plus modestes sont consacrés pour reproduire les noyers noirs ayant démontré une certaine résistance au froid, ainsi que pour identifier des provenances et descendances d'érable à sucre ayant un taux de sucre supérieur et un phénotype intéressant.

# AMÉLIORATION DES MÉLÈZES (LARIX SP.) ET DE L'ÉPINETTE DE NORVÈGE (PICEA ABIES KARST.) PAR ANTE STIPANICIC

# Mots-clés : Tests de descendances, tests de provenances, croisements dirigés, vergers à graines.

Au cours des deux dernières années, les travaux de croisements dirigés se sont poursuivis dans le but de produire et d'évaluer les hybrides interspécifiques entre le mélèze d'Europe et le mélèze du Japon. En 1991, 45 croisements ont été effectués, mais seulement 25 ont réussi. Aucun croisement n'a été effectué en 1992, faute de floraison. Un test de 40 descendances biparentales a été installé dans la pépinière de Duchesnay en 1991 avec les plants issus de bouturage. Un autre test, cette fois composé de 37 descendances a été installé en 1992 dans la pépinière de Saint-Modeste.

Les greffes de 25 clones sélectionnés de mélèze d'Europe et de mélèze du Japon ont été installées dans une serre-tunnel et serviront pour la poursuite de programme d'amélioration des deux espèces et pour la production d'hybrides sous abri. Un test de 60 descendances de mélèze de Sibérie obtenues de l'INFP a été installé dans la pépinière de Normandin en 1992. Il sera transféré sur le terrain en 1994.

Les éclaircies sélectives ont été pratiquées dans deux plantations expérimentales en collaboration avec la compagnie PFCP ltée dans le but de les aménager en vergers à graines pour la production de semences de mélèze hybride (*L.* x *eurolepis*) et de mélèze du Japon.

Avec l'épinette de Norvège, quatre tests de descendances ont été installés dans nos arboretums en Gaspésie en 1991 et 1992. Au total, 222 descendances sont représentées. Elles ont été récoltées sur des arbres sélectionnés dans quatre plantations expérimentales transformées en sources de graines améliorées. Deux autres tests ont été mis en marche dans les pépinières de Duchesnay et de Saint-Modeste en 1991. Il s'agit de 98 descendances dont 4 sont biparentales, qui ont été multipliées par bouturage.

En collaboration avec Forêts Canada, Région du Québec, nous avons installé sur le terrain trois tests de descendances dans chacun des trois domaines écologiques de la zone C d'amélioration de l'épinette de Norvège au Québec. Il s'agit de 269 descendances récoltées sur les arbres sélectionnés soit dans les peuplements naturels de Pologne soit dans les plantations (tests de provenances ou plantations commerciales) du Québec et de l'Ontario. Deux nouveaux tests de cette expérience ont été installés dans les pépinières de Grandes-Piles et de Duchesnay en 1992. Ils seront transférés sur le terrain de la zone d'amélioration A en 1994. Le but de ces tests est de délimiter avec plus de précision les zones d'amélioration de l'épinette de Norvège au Québec et d'obtenir du nouveau matériel nécessaire à la poursuite du programme d'amélioration. En ce qui concerne les croisements dirigés avec l'épinette de Norvège, le Service de l'amélioration des arbres évalue la qualité des hybrides obtenus lors de croisements effectués par le Service de la production de plants. En 1991 aucun croisement n'a été effectué à cause de faible floraison. Par contre en 1992, 33 combinaisons avec les arbres sélectionnés de provenances recommandées pour la zone C ont été pratiquées dans l'arboretum de Lac Saint-Ignace et à la Station forestière de Valcartier (Forêts Canada). Plus de 14 800 semences ont été ainsi obtenues. L'ensemencement et le bouturage de ce matériel se fera à la bouturathèque de Saint-Modeste. Les plants obtenus seront destinés au reboisement et à des tests de descendances.

Les travaux de sélection des arbres pour la résistance au charançon (*Pissodes strobi*) se sont poursuivis : 45 nouveaux arbres ont été sélectionnés dans la région de la Beauce où cet insecte fait beaucoup de dégâts.

Dans le cadre de transformation des anciens tests en source de graines améliorées, nous avons commencé les travaux d'éclaircie dans une plantation expérimentale composée surtout de descendances d'épinette de Norvège de Bulgarie. Les coupes d'éclaircie sélective seront réparties sur les quatre à six prochaines années.

# SÉLECTION DE CLONES ET AMÉLIORATION GÉNÉTIQUE DU PEUPLIER PAR GILLES VALLÉE

Mots-clés : *Populus,* sélection de clones, tests de provenances et de descendances, test clonal, croisements.

Depuis 1988 les travaux sont surtout réalisés dans les régions du Saguenay — Lac-Saint-Jean et de l'Abitibi-Témiscamingue où des programmes de populiculture sont en voie d'accomplissement.

En résumé, à ce jour, un total de 3 689 clones ont été sous observation au SAA, dont 575 ont été sélectionnés dans les peuplements naturels au Québec et 2 355 dans des plantations comparatives et des pépinières; 759 autres clones ont été introduits surtout d'Europe et d'Ontario. De plus 73 dispositifs de tests clonaux, 23 plantations de collections de clones, 38 dispositifs de tests de provenances et descendances ont été mis en place. Quelque 2 834 croisements ont été faits dont 377 ont donné des semis. Ajoutons l'obtention de pays étrangers de 257 lots de semences et de la récolte au Québec de 529 lots de semences sur 29 espèces ou hybrides.

En 1992, les croisements suivants ont été faits : *P. x Rollandii x P. maximowiczii* et *P. trichocarpa, P. x Jackii x P. maximowiczii* et *P. trichocarpa.* Ces croisements ont produit plus de 3 000 semis qui seront soumis à une procédure de sélection pour la production de clones. Les génotypes de *P. x Rollandii* et *P. x Jackii* utilisés dans les croisements proviennent de régénération naturelle dans la région du Saguenay — Lac-Saint-Jean. Tandis que ceux de *P. maximowiczii* et *P. trichocarpa* sont des sélections dans des populations introduites. En 1991-92 quelque 16 tests clonaux, 4 tests de descendances, 11 plantations de collection de clones et 7 plantations de collection de provenances-descendances ont été établis. L'analyse statistique des tests clonaux de 10 ans et plus a été effectuée permettant la recommandation de clones pour différentes régions du Québec.

## AMÉLIORATION DE L'ÉPINETTE NOIRE PAR MICHEL VILLENEUVE

Mots-clés : Test de descendances, épinette noire, croisements dirigés, bouturage, embryogenèse somatique.

Depuis 1991, deux tests précoces en pépinière et trois tests de descendances de première génération ont été établis. Le réseau de vergers et de tests de première génération est maintenant complété. Le nombre de dispositifs expérimentaux pour l'amélioration génétique de l'épinette noire s'élève maintenant à 81.

Deux variétés multifamiliales sont disponibles pour le reboisement. Les croisements dirigés nécessaires pour leur perpétuation sont dorénavant faits par le Service de la production de plants du ministère des Forêts, qui est responsable de l'approvisionnement de semences pour le Centre de bouturage de Saint-Modeste. Nous effectuons seulement les pollinisations requises pour nos propres expérimentations. L'étude de l'aptitude au bouturage des familles qui composent les variétés multifamiliales a démontré qu'un classement fiable des familles est très difficile à obtenir. Par contre, lorsque les conditions de bouturage sont uniformes, il semble possible de classer correctement des clones.

Les lots de boutures racinées destinées au reboisement ont toujours été produits en vrac, sans identification des familles. On ne connaît pas la diversité génétique des variétés multifamiliales livrées sur les sites de reboisement. Dans ce but, un protocole de suivi démographique des populations familiales de boutures racinées est à l'essai; 7 560 boutures de 21 familles ont été identifiées et seront suivies jusqu'à l'expédition des plants livrables.

Dans le cadre d'un mémoire de fin d'études, un étudiant en foresterie a analysé les résultats d'un test d'hybrides interprovenances après 20 saisons de croissance. La différence entre les provenances pures et les hybrides est minime : 5 % de différence pour la hauteur à 20 ans. Toutefois, la corrélation entre la hauteur à 20 ans et celle à 10 ans est très élevée (r = 0.96).

Un guide photographique pour l'évaluation du débourrement chez l'épinette noire est en préparation. Les sept stades reconnus semblent permettre de bien distinguer les familles dans un verger à graines de l'Outaouais. Un examen approfondi des résultats de ce printemps devrait confirmer l'utilité du système.

Des subventions ont été attribuées pour l'étude de l'embryogenèse somatique et pour la réalisation de la transformation génétique «in vitro». L'embryogenèse somatique a permis de produire plus de 3 500 plants de 25 clones provenant de 6 familles. Ces plants sont présentement en contenant et serviront à établir diverses plantations expérimentales. La transformation génétique a pour but précis d'introduire le gène de la toxine du *Bt* dans des génomes d'épinette blanche. La technique de biolistique («gene gun») est utilisée.

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# FOREST GENETICS RESEARCH AT LAVAL UNIVERSITY

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The past three years at Laval University have seen several positive developments in the field of forest genetics and tree improvement. In 1991, Dr. Louis Parrot has returned full time to his research projects on the improvement of hardwood species after six years at director of Département des sciences forestières. Jean Bousquet joined The Centre de recherche en biologie forestière (CRBF) and the Département des sciences forestières in 1990 to develop a research program in the area of molecular markers and their use in forest genetics, tree improvement, and phylogenetics. Dr. Francine M. Tremblay from Petawawa National Forestry Institute moved her research activities on tissue culture and somatic embryogenesis to the CRBF in 1990 following her assignment to Laval University through the interchange Canada Programme between Forestry Canada (FORCAN) and Laval. She has been supervising over the last two years one postdoc, three graduate students and two research assistants. Dr. Armand Séguin from Salk Institute joined the CRBF in 1992 to develop a research program on gene expression and gene transformation of conifers. He is supervising two Ph.D. students and one research assistance, in collaboration with Drs. F. Tremblay and J. Bousquet. This report will focus more specifically on research activities involving the three co-authors. Our group has been composed over the last two years by PDFs Dr. Peng Li (Ph.D. in forest genetics from Oregon State Univ., NSERC fellow) who has moved at the level of senior research associate in 1992, Dr. Laurent Maggia (Ph.D. in molecular biology from Univ, Paris VII - Nogent, NSERC fellow), Dr. Nathalie Frascaria (Ph.D. in population genetics from Univ. Paris XI - Orsay, AUPELF fellow), Ph.D. students Louise Savard (who completed her Ph.D. in September 1992, now at Labatt Research Division, London), Khasa Phambu (who completed his Ph.D. in June 1993), Nathalie Isabel (who joined the group in 1992, and co-supervised by Dr. Francine Tremblay), Carmelle Beaulieu (who joined the group in 1992, and co-supervised by Dr. Peng Li), M.Sc. student Martin Perron, and research associates Martin Michaud (biochemistry, M.Sc.), and Alice Roy (agricultural scienes, M.Sc.).

#### MOLECULAR PHYLOGENETICS

Several projects have been completed in the past two years. In collaboration with Dr. Steve Strauss (Dept. Forest Science, Oregon State Univ.), we have completed a review on the biosystematics of forest trees using molecular and biochemical markers (Strauss et al. 1992) and two major phylogenetic studies involving sequences of the chloroplast gene *rbcL* (Bousquet et al. 1992a, 1992b). Using procedures previously established for the amplification of specific genes (Bousquet et al. 1990), we have developed protocols for rapidly sequencing *rbcL* in tree species,

resulting in the determination of a large number of sequences for conifer and hardwood species. We have estimated phylogenies at the family level for conifer species and hardwood species, we have estimated phylogenies among major groups of plants, found that woody species evolved at a slower rate than annual species, and found complete agreement between phylogenies estimated using sequence data from the rbcL ORF, from the downstream flanking region of rbcL, and morphological characters in the Betulaceae. Using rbcL sequences, we are in the process of analyzing the higher Hamamelidae, which includes several major families of hardwoods such as the Juglandaceae, the Betulaceae, and the Fagaceae (Frascaria et al. 1993, Maggia and Bousquet 1993). Dr. Louise Savard and Mr. Martin Michaud have also developed the use of nuclear internal transcribed spacer sequences (ITS) for the estimation of family and genus structure in the Betulaceae, and show that the rate of substitution of this gene is about more than ten times those of the gene *rbcL* or the nuclear gene coding for 18S rRNA (Savard et al. 1993), making it very useful for deciphering relationships among closely related taxa. In collaboration with Steve Strauss and Dr. Mark Chase from Univ. of North Carolina, Drs. Louise Savard and Peng Li have 🗉 also amplified by PCR and determined 18S rRNA gene sequences for conifers, estimated plant phylogenies, and derived rbcL and 18S molecular clocks to estimate the time for the diversification of seed plants, which includes conifers and angiosperms (Savard et al. 1993). In collaboration with Drs. Maurice Lalonde and Roger Lévesque from Laval University, Mr. Luc Simon, a Ph.D. student, has also used similar strategies and estimated, using 18S molecular clocks, the origin of endomycorrhizae fungi. He found that it coincides with the origin of land plants (Simon et al. 1993). Dr. Li has also developed an improved statistical test for molecular clocks (Li and Bousquet 1992). Dr. Peng Li is currently completing a study on the evolution of mitochondrial genes in plants, including their use as molecular clock, and Dr. Maggia is completing a study on RNA editing in the Betulaceae, by developing PCR-based strategies of amplification of both genomic DNA of particular genes such as CoxI and their mRNA. We are also developing strategies to amplify more variable regions of the mitochondrial genome for phylogenetic studies at the family and genus levels. In collaboration with Drs. Ben Lepage and James Basinger from the Dept. of Geology of the Univ. of Saskatchewan, and financial help from the Polar Shelf Project, Dr. Louise Savard and Mr. Martin Michaud are also completing a study involving the isolation of fossil DNA and the sequencing of rbcL genes from Eocene trees (45 million years) of Axel Heiberg Island. Our phylogenetic studies are funded by NSERC and Fonds FCAR of Québec.

#### MOLECULAR POPULATION GENETICS

In the past two years, we have developed random amplified polymorphic DNA markers (RAPDs) for black spruce (Isabel et al. 1993) and yellow birch (Roy et al. 1992), which was made possible by grants from the Québec Ministry of Forests (MFOQ) and Fonds FCAR. In the first case, collaborations have been developed with Dr. Francine Tremblay and Mr. Michel Villeneuve (from Service d'amélioration des arbres (SAA) of the MFOQ), and in the second case, with Mr. John Mackay (also from SAA-MFOQ). For both species, we have described the mode of inheritance for a number of markers, and use them for various purposes such as fingerprinting of cell lines, follow up of the genetic stability through the process of somatic embryogenesis, certification of crosses, and detection of contaminants. Mrs. Nathalie Isabel has undertaken with Dr. Jean Beaulieu (FORCAN) a comparison of diversity estimates derived from allozymes and RAPD markers among and within five natural populations of black spruce in Québec. Mrs. Isabel is also pursuing in collaboration with Drs. Francine Tremblay and John Carlson (from Biotechnology Lab, UBC) the development of DAF markers for the evaluation of the genetic integrity in black spruce and white spruce clonal populations derived from somatic embryogenesis. She is also developing PCR-based strategies in collaboration with Dr. Francine Tremblay to estimate DNA methylation levels and their patterns of variation in cell lines of black spruce. Mrs. Alice Roy is completing two studies, 1) on the effect of bulking individuals and populations and the competition effects in PCR on population and species RAPD profiles, and

2) on the use of species-specific RAPD markers for the estimation of the phylogeny of the genus *Betula* and the certification of interspecific crosses. This project has been funded by the MFOQ, and Mr. André Rainville from SAA-MFOQ is actively involved in the project by performing the crosses. In collaboration with Dr. Bill Cheliak (formerly at PNFI, now at the Pest Management Institute of Forestry Canada), Dr. Khasa Phambu has completed the development of allozyme markers for a number of tropical species (Khasa et al. 1993), and using some of these allozyme markers, he has reassessed the population genetics of two Acacia species, found an ancestral-descendant relationship between them, and evaluated their mating system (Khasa et al. 1993a,b,c,d). Our isozyme studies on alder species conducted in collaboration with Dr. Maurice Lalonde are now completed (Bousquet and Lalonde 1991).

#### GENECOLOGY AND TREE IMPROVEMENT

In collaboration with Mr. John Mackay from SAA-MFOQ, Dr. Peng Li has completed a review of the population genetics and the genecology of hardwoods in Canada, and their implications for gene conservation (Li et al. 1992a). In collaboration with Dr. Louis Parrot and funds from MFOQ, he has also completed a study on the adaptation and performance of 21year-old biparental progenies of black walnut derived from parents introduced in Québec more than a hundred years ago (Li et al. 1992b). In collaboration with Dr. Jean Beaulieu from FORCAN, Dr. Li has completed a study on early testing and the genecology of white spruce in Québec (Li et al. 1993). He is currently completing a study on the delineation of breeding zones for white spruce in Québec using multivariate approaches, in collaboration with Dr. Jean Beaulieu. Dr. Khasa Phambu has completed his studies on the genetic variation associated with rooting ability of cuttings of Acacia species, in collaboration with Dr. Gilles Vallée from SAA-MFOQ (Khasa et al. 1993e). A project funded by the program "Essai, expérimentation et ransfert technologique" of FORCAN has been initiated last year in the Bas St-Laurent - Gaspésie region, in collaboration with Jean Beaulieu and Corporation Agro-forestière Transcontinental. The goals of the project are to select for superior materials adapted to this region from local sources and 2nd generation materials derived from crosses. Family and clonal tests are currently being established on several sites. In collaboration with Dr. Louis Parrot, SAA-MFOQ and Mr. André Rainville, we are also initiating a genetic improvement program for sugar maple. Also in collaboration with SAA-MFOQ and Mr. Michel Villeneuve, we are initiating a program aimed at developing red black spruce hybrid varieties more resistant to early competition in commercial plantations. Both precedent programs involve conventional breeding and the development and use of RAPD markers. Dr. Peng Li, Mrs. Alice Roy, and Mr. Martin Perron, a new graduate student, will be the most active researchers in these programs for the next three years.

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# RECHERCHE ET DÉVELOPPEMENT SUR LES SEMENCES ET LE POLLEN À LA DIRECTION DE LA RECHERCHE DU MINISTÈRE DES FORÊTS DU QUÉBEC

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**Mots-clés:** Pollen, conservation, test de germination, forçage, contamination pollinique, pollinisation de masse, semence, maturation, physiologie, triage, étêtage, dormance.

L'un des mandats de la Division de R-D sur les semences, boutures et plants du Service de l'amélioration des arbres (MFO) est de planifier et de réaliser des travaux de R-D sur la production, l'utilisation et la protection des semences et du pollen forestiers dans le but de répondre aux besoins du secteur des opérations régionales. Ce mandat est divisé en trois projets: la production et l'utilisation du pollen, la biologie des semences des arbres résineux et la biologie des semences des arbres feuillus.

#### PRODUCTION ET UTILISATION DE POLLEN

Le MFO a mis sur pied une banque de pollen qui permet de maintenir la viabilité de grandes quantités de pollen des principales espèces résineuses durant une période de 3 à 5 ans. Parallèlement à cette banque, des tests de germination ont pu être également développés afin de valider chacune des entrées et des sorties de pollen de cette banque. Par ailleurs, nous avons mis au point une technique qui permet de réduire le temps de développement des cônes mâles de *Pinus banksiana* d'environ deux semaines sans nuire à la qualité ni à la quantité de pollen produit. La contamination pollinique a également été évaluée dans quatre vergers à graines afin de réduire le taux de contamination en-deçà du seuil fixé à 20%. Enfin, nous travaillons actuellement sur des techniques de pollinisation de masse qui permettrait de réduire la quantité de pollen utilisée et éventuellement, d'augmenter le rendement en graines des cônes.

#### PRODUCTION ET UTILISATION DE SEMENCES

Les principaux travaux réalisés ont porté sur l'étude morpho-physiologique de la maturation des semences de *Picea glauca*, de *Pinus strobus* et de *Pinus resinosa*. Par ailleurs, des travaux d'étêtage (étêtage, éducation et rabbatage) ont également été entrepris dans un verger à graines de *P. glauca*. Enfin, un germoir de masse permettant de faire germer plusieurs kilogrammes de semences à la fois a également été développé.

L'étude de la dormance des graines du *Prunus serotina* a été entrepris afin de mettre au point une technique de quantification de l'état de dormance d'un lot ainsi que pour lever cette dormance au printemps. Des travaux sur la synchronisation de la germination des glands de *Quercus rubra* ont été réalisés afin d'identifier l'origine de l'étalement de la germination de ces semences.

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# TREE GENETICS AND IMPROVEMENT AT FORCAN QUEBEC

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Keywords: *Pinus strobus, Picea glauca, Picea abies,* tree improvement, genetic structure, isozymes, flower induction, tissue culture, white pine blister rust, white pine weevil, clonal test, tree selection.

Research activities on the genetics and improvement of white spruce (*Picea glauca* (Moench) Voss.), Norway spruce (*Picea abies* L. Karst) and white pine (*Pinus strobus* L.) have increased during this period through the addition of three professionals, Mrs. Marie Deslauriers (biotechnology), Dr. Richard Hamelin (pathologist) and Mr. Robert Lavallée (entomologist) to our team. Our efforts continue to be focused on genetics and improvement, but special attention is now directed toward the two main pest problems on our conifer species in Quebec, i.e. blister rust (*Cronartium ribicola* J. C. Fisch). and white pine weevil (*Pissodes strobi* Peck). The project is also increasing its activities in the use of biotechnology as it offers useful tools to study the genetics of our species.

#### WHITE SPRUCE

Research objectives on the genetics and improvement programme initiated in 1976 have been pursued as planned. The genetic variation distribution among and within provenances, the extent of genetic control in growth and bud phenology traits and potential of early testing for height growth were investigated in a provenance-progeny test involving 285 open-pollinated families from 57 provenances from Ontario and Quebec (Li et al. 1993). Eight open-pollinated half-sib family field tests have been established at the beginning of the 1980s. They were assessed for 13-year survival, height and damage. Bud phenology was also assessed in one of the tests where growth conditions are the most unfavourable. Data analysis was initiated and provisional seed transfer zones will be established. The best families and superior trees will be selected to complete our breeding population for which 100 superior trees have already been selected and grafted.

Flower induction techniques were routinely used with success to increase strobile production. Stem injections of  $GA_{4/7}$  and a bumper seed crop in 1992 allowed us to initiate more than 50% of the controlled crosses from our breeding plan and to establish a pollen bank. Our studies of grafts in the soil-based clonal bank revealed that, even in a good seed crop year, grafts pruned on a regular basis and maintained to a maximum height of 2 m produce female flowers only when they are stimulated by  $GA_{4/7}$ . The male flower production on that material was very low and only a few clones that were treated produced. In the spring of 1993, a year in which natural flowering was poor, the use of  $GA_{4/7}$  helped us to make some controlled crosses and to add many clones to our pollen bank. We have also investigated the effects of flower induction on segregation and gene linkage using isozyme analysis. Hormone injection as well as heat stresses did not affect allele segregation. Furthermore, we were not able to show any

significant effect on gene linkage. These results convinced us that flower induction is a safe technique to increase cone production and speed up the completion of our breeding programme.

A progeny test was initiated in 1992 and will be established at three different locations in the Gaspesian and Lower St. Lawrence regions. These tests will include 24 openpollinated half-sib families from local sources and 76 full-sib families from selected trees in genetic trials located in western and central Quebec. Our objective is to demonstrate that progenies from selected trees are well adapted to regional conditions and may give higher yields than local sources.

The genetic structure and variability of two natural populations from southern Quebec were compared, using allozyme variants, to those observed in three provenances from the Great Lakes - St. Lawrence region. Furthermore, genetic variability observed in a number of superior trees selected within these provenances and integrated into our breeding population was compared to the one found in the two former groups. Results showed that levels of genetic diversity were similar in both natural populations and provenances and that selection of a restricted number of individuals within the latter did not result in a loss of alleles. The level of heterozygosity observed in the phenotypically selected trees was higher than in the natural populations or provenances suggesting that selection in favour of individuals demonstrating a strengthened vigour favoured the most polymorphic genotypes.

## NORWAY SPRUCE

In 1992, three progeny tests, including 266 half-sib families, were established in the Appalachian breeding zone. A test was located in each of the three forest associations (balsam fir - white birch, balsam fir - yellow birch and sugar maple - yellow birch) found in this zone. The main objective of these tests is to provide information to more precisely delineate and, if necessary, subdivide the breeding zone. The experimental design used is adapted from the modified augmented design proposed by Lin and Poushinsky (Lin, C.-S. and G. Poushinsky. 1983. Biometrics, 553-561.) Material for the tests in the Laurentian breeding zone was also transplanted in 1992 to our research nursery at the Valcartier Forest Experiment Station (V.F.E.S.) and to the provincial nurseries at Duchesnay and Grandes-Piles. They will be established in 1994. All of these progeny tests were planned, produced and established in collaboration with the Service d'Amélioration des Arbres du Ministère des Forêts du Québec.

Clonal tests were also initiated from family and individual selections made in the progeny test raised at the V.F.E.S. More than 350 clones representing more than 60 families were propagated during the summer of 1992. The selected families represent early-stage fast growing stock from the old Proulx, Smith and Lachute stands and fast-growing stock, at a later stage, originating from Latvia, Byelorussia and Northeastern Poland. These clones will be used to set up two tests, the first one aiming to demonstrate the effectiveness of early selection in families coming from recommended provenances; the other, to study the genetic variability in the selected material grown under similar ecological conditions to those of the progeny tests in the Appalachian breeding zone.

The study of wood quality of the 22 provenances from Central Europe has been completed. In addition, data from a 70-year-old plantation have been collected and analysed to evaluate wood quality radial variation from the pith to the bark and also to show the relationship between juvenile and mature woods. Results have been presented (Blouin et al. 1993) and will be published in the near future.

The combined treatments of root pruning, heat and  $GA_{4/7}$  applied to 2 m-high containerized grafts gave us poor results, only a few clones gave male and/or female flowers. In

the soil-based clonal bank, the clones treated in 1991 and those treated in 1990 (carry-over effect) produced more flowers than the control. The statistical analysis will be done this year. In 1992, but not in 1993, the  $GA_{4/7}$  was very useful in stimulating male flowering on selected trees in a provenance trial that was established in 1969.

As the white pine weevil has become the most significant pest in plantations in Quebec, especially on Norway spruce, it was decided to significantly increase our research efforts on this problem. Research was ongoing on the biology and the dynamics of the insect in plantations as well as on the influence of site-related factors on plantation susceptibility and weevil biological performance. Since 1992, when Mr. Robert Lavallée joined our project, much effort is being invested in the study of tree resistance or tolerance to the insect. First-year results are presented in the poster section.

#### WHITE PINE

The inheritance of allozymes of 15 polymorphic loci was demonstrated using haploid megagametophytic tissues of viable seed from a total of 300 eastern white pine trees sampled in ten natural populations in Quebec. Except for two allozymes of the second locus of the phophoglucose isomerase, allozymes segregated as expected from a Mendelian control. Linkage analysis for 69 pairs of loci showed independency. The study of the population genetic structure revealed a slight excess of homozygotes overall due to population subdivision considering that subpopulations were in Hardy-Weinberg equilibrium. The degree of differentiation among populations was low (2%), as expected from a coniferous species with a wide natural range. The estimated outcrossing rate was nearly 1.0, suggesting that there was a very small frequency of selfing in the two populations studied. However, there was an excess of homozygotes in the filial populations while there was either equilibrium or a slight excess of heterozygotes in both mature populations (Beaulieu 1993). These results give a good idea of the genetic diversity of this species over its distribution in the province of Quebec. To complete the study, we need to sample natural populations in the Gaspesian and Mauricie regions and at the limits of its distribution, i.e. in Abitibi and on the North Shore. Based on these results, we will review our selection programme to insure that our breeding population contains most of the diversity found in Quebec.

From 1986 to 1988, 5 genecological tests were established under aspen and birch cover in the moderate rust incidence zone of southern Quebec. Five-year growth measurements, survival and damage assessments were made in 1991 in 2 of the 5 genecological tests. The last 3 tests will be evaluated this year. The tests were established under cover to increase the reliability of genetic information by reducing tree damage caused by the white pine weevil.

The selected trees in the Cap Tourmente breeding orchard have produced few flowers over the last period. Crosses have been made using pollen from exotic pine species from the tree collection at Maple, Ontario. As some of the blister rust resistant trees from Maple have been grafted, we will increase our hybrid production as flowers are produced and select the progenies for resistance to blister rust. To do so, a special project has been set up to study the genetic variability of the rust over its range. A previous study using isozyme analysis showed little evidence of variability (Bérubé and Plourde 1992). To confirm these results, a large spore collection was gathered this spring and summer to study the variability of the fungus using RAPD's markers. Dr. Richard Hamelin joined our team in November 1992 and is responsible for this activity. Interesting progress has been achieved in *in vitro* spore production of the rust as monosporal cultures have been obtained. Efforts will be directed toward developing an artificial inoculation technique. Embryogenic lines of white pine have been obtained and maintained. Research is ongoing to find the *in vitro* requirements for embryo maturation and germination.

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# POLLEN DISPERSAL STUDIES

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Keywords: Pollen dispersal, seed orchards, contamination, Picea mariana, Pinus banksiana.

Studies in pollen dispersal have been on-going since 1989 at the Island Lake seed orchard near Chapleau, Ontario. The orchard, established in 1982, includes jack pine (*Pinus banksiana* Lamb.) approximately 10 years old, and black spruce (*Picea mariana* (Mill.) B.S.P.) approximately 5-7 years old. The aim of the study is to model the dispersal of wild pollen into and across seed orchards. The model will help management decisions regarding anti-contamination methods intended for seed orchards.

#### POLLEN DISPERSAL MODEL

At present, a Lagrangian Markov-chain model developed for forestry-spraying applications (Picot et al. 1987) is being modified for pollen dispersal across seed orchards. Once developed and tested, the model will be made user-friendly and used to answer management questions about the use of physical anti-contamination methods such as buffer zones or wind breaks at various seed orchard sites.

#### SOURCES OF CONTAMINANT POLLEN

Seasonal liberation of pollen is being investigated for jack pine and for black spruce. The work is in cooperation with Prof. G. Caron (Univ. Moncton). The aim is to develop a heat-sum model to predict the release-date of pollen and may be useful for anti-contamination methods such as water-spaying or SMP.

The diurnal variation of atmospheric pollen is being investigated. Preliminary data suggests most pollen is liberated between approximately 10 a.m. and 4 p.m. local time.

The variation in concentration with height at the upwind edge of the seed orchard will be measured in 1993. As well as providing input data for the model, some light will be shed on the amount of conifer pollen high above the orchard.

#### DISPERSAL OF POLLEN

Pollen dispersal was measured using sticky slides from 1989-91. However, Rotorod pollen samplers were used from 1992-93. These were set-up in a 5x5 grid pattern across the seed orchard and at a height of 3 m. Exotic pollen of known aerodynamic characteristics, oil palm (*Elaeis* sp.) and *Lycopodium* sp., has been released into the seed orchard from a point source during the pollination season. This will be used to verify the dispersal model because with-in orchard production of jack pine pollen occurs. Preliminary results using 15 m releases of these 'light' pollen indicate conifer pollen entering at this height would certainly deposit within 400-800 m under the conditions encountered so far.

#### DEPOSITION OF POLLEN

Pollen deposition to the ground, on to vegetation and on to seed cones has been measured in the field. Wind tunnel studies will also be conducted to quantify the effectiveness of the seed cones and the vegetation to capture airborne pollen.

Preliminary work on the effect of rain has shown that more pollen is deposited on to seed cones by rain, under certain conditions, than by direct impaction of airborne pollen. These conditions are likely to occur in the 'afternoon thunderstorms' caused by instability during high pressure conditions.

### GENETIC MEASUREMENTS OF CONTAMINATION

Isozyme measurements of contamination are being made within the seed orchard as a whole, and at a selected points within the seed orchard. This data will be compared with the contamination estimates predicted by the dispersal model.

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# FOREST GENETICS AT LAKEHEAD UNIVERSITY

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Keywords: Genecology, focal point seed zones, Larix laricina, Pinus banksiana, Picea mariana, Populus, Abies lasiocarpa var. bifolia.

As part of a recent curriculum revision, forest genetics and tree improvement are now taught as a required one-semester second year course in the four year H.B.Sc.F. programme. Two graduate courses are also offered on demand.

Research directions are changing: two of the faculty (REF and PHT-K) are no longer actively pursuing research in forest genetics; and the third (WHP) has changed directions from phenotypic studies of conifers to 1) short-term genecological trials to provide improved means for seed transfers in jack pine and black spruce and 2) the interaction of genetic and spacing factors in determining form in jack pine.

#### POPULATION GENETICS

Peggy Tripp-Knowles has shut down her isozyme laboratory and ceased further work in population genetics. Her new areas of interest are Feminist Science Critique and the History and Philosophy of Science.

#### GENETIC STUDIES OF TAMARACK AND BALSAM POPLAR

Rob Farmer has recently completed manuscripts based on long-term studies in tamarack and balsam poplar. He is now terminating his genetics research and concentrating on seed physiology beginning with a study of white pine germination. He is also writing an undergraduate text on seed ecophysiology of North American forest trees. He is currently serving as a consultant for poplar breeding projects in India and China.

#### FOCAL POINT SEED ZONES

Bill Parker has developed a new site-specific approach to defining seed zones in North American conifers based on short term provenance field trials, greenhouse trials and comparative frost hardiness trials. Using focal point seed zones, an individual site to be reforested becomes the focal point, and a unique seed zone is established for that site as needed. This approach depends upon (i) obtaining good comparative data in adaptive characteristics from throughout the range to regenerated and (ii) graphic analysis of multivariate summary scores by geographic information systems software to delimit boundaries of unique seed zones for any location to be reforested. A database has been established for jack pine and black spruce to the east and west of Lake Superior, and additional collections and tests are underway to expand this area westward to the Manitoba border.

Three graduate students have been working on short term-testing projects to provide the data base for focal point seed zones. Annette van Niejenhuis has conducted field and greenhouse trials of jack pine; Maria Davradou completed comparative frost hardiness trials of jack pine; and, Sun Hongnian has completed frost hardiness trials of black spruce. Annette has assumed a position as research associate in charge of continuing collections and trials of jack pine and black spruce.

## COMPARATIVE PHENOTYPIC VARIATION STUDIES

Lynn Palmer completed her M.Sc.F. studies investigating phenotypic variation in Yukon populations of subalpine fir. Multivariate analyses indicated geographic differentiation of coastal and interior subalpine fir populations and an affinity between northern and interior subalpine fir populations. Results indicate a common Rocky Mountain refugium for the northern Yukon and more southerly interior populations. Varietal status is proposed for subalpine fir populations occurring east of the Coast and Cascade mountains of Washington and British Columbia. The new name for this interior variety is *Abies lasiocarpa* (Hook.) Nutt. var *bifolia* (Murray) Palmer & Parker.

Maddie Maley completed her M.Sc.F. studies investigating phenotypic variation in northwestern Ontario populations of jack pine. Discriminant analysis of cone and needle data indicated that the sampled populations generally varied longitudinally with a steep cline in the Nipigon area (ca. 88°15'W) apparent for cone data. These results may reflect separate lineages for east and west groups although the observed pattern may simply be the result of local adaptations to a variable environment.

#### GENETIC X SPACING FACTORS DETERMINING JACK PINE FORM

In cooperation with Dave Morris, OMNR, Centre for Northern Forest Ecosystem Research, a short term field trial has been established as a test of wind-pollinated progenies from parent trees having good, average and poor form grown under different initial spacings (0.25 to 3 m) in a common plot on two contrasting sites. This trial will be used to determine (i) narrowsense heritabilities and genetic correlations of the traits that determine jack pine form and (ii) changes over time to these parameters at the different spacings.

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# **GENETICS AND GENECOLOGY** *PICEA* L. (SPRUCE)

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Keywords: *Picea*, genecology, genetic variability in spruce forest ecosystems, interspecific hybridization, gene pool conservation.

The objectives of these studies are: (1) to quantify genotype x environment interaction as part of a breeding strategy for selecting desirable spruce genotypes for maintaining genetic variability and diversity in spruce forest ecosystems, and for developing valuable hybrids for 'new forests' under changing environmental regimes; (2) explicitly, to investigate G x E interaction by determining genetic variation in efficiency, growth and nutrition, and productivity as related to site regions, breeding zones, etc., utilizing the *Piceta* as outdoor laboratories; (3) to create long-term genetic banks for gene pool and population studies, breeding and assessment of genetic parameters to elucidate the breeding system and structure of the genus *Picea*; (4) to produce, test and select the best hybrids for propagation; (5) to utilize the arrays of spruce species and genotypes located in the *Piceta* for long-term monitoring of forest health as related to possible airborne pollution and/or climatic change.

#### HYBRIDIZATION

The hiatus in flowering of spruce noted in 1990 (following two years 1988, 1989 of massive flowering) continued in 1991. Only 64 tree x pollen parent crosses were made utilizing 140 ramets. There were 30 interspecific crosses involving 18 species and one tri-hybrid cross.

Results, in keeping with the extremely low level of flowering, were mostly negative. Many crosses, including controls, resulted in hollow seeds. Even open pollinated collections had very low germination. Several confirmatory crosses were obtained and three new-to-science crosses: *Picea jezoensis hondoensis* x *P. maximowiczii*; *P. orientalis* x *P. glehnii*; and *P. omorika* x *P. smithiana*; all with low crossability. Seedlings of the latter cross, however, are not yet confirmed and must await development of greater biomass sufficient for evaluation.

The much awaited flowering in 1992 was massive and extended through several districts in three site regions. Tragically, due to management constraints funds for this work were not released until the breeding window in *Picea* was well advanced. Since female strobili were already opening we were forced to abandon breeding designs embracing all the early and mid season flowering types. Only small numbers of late-flowering types were therefore captured.

Twenty-eight tree x pollen parent crosses were made, replicated with 62 ramets. There were 20 interspecific crosses and two tri-hybrid crosses made, utilizing 18 species and one form. The breeding, unfortunately, constituted about one half of what might be expected in a <u>poor</u> flowering year. Confirmatory crosses were with *P. mexicana* x *P. bicolor* (Gordon 1990), all with very low crossability. An expected cross was that of the closely related species *P. shirasawae* x *P. koyamai*. Crossability was moderately high. Seedlings germinated from several crosses exhibited evidence of contamination, presumably due to late borderline bagging, and were discarded. Pollen collections were also much reduced: only six species were collected.

#### FAST GROWING HYBRIDS FOR THE BOREAL FOREST

Breeding was finally completed in 1991 sufficient to satisfy the remaining unfilled compartments of a factorial design of the interspecific cross *P. omorika* x *P. mariana* (Gordon 1976, Fowler 1980). This work was commenced several years ago (Gordon 1989) but natural vicissitudes (pollen failure, flowering periodicity, squirrel depredation, etc.) have long delayed the completion of breeding. The seed is currently banked but germination will be initiated when time permits and suitable areas for the experiment will have been selected.

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# PHYSIOLOGICAL GENETICS AND PLASTICITY

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# Keywords: Climate change, drought tolerance, elevated CO<sub>2</sub>, G x E, *Picea mariana*, provenance, water use efficiency, within-stand variation.

#### INTRODUCTION

The Physiological Genetics and Plasticity Project examines genetic variation in adult tree and seedling physiological, morphological and phenological traits and the relationship of such traits with productivity and/or survival. This approach is being applied toward research aimed:

- (1) to assess the "plasticity" of natural and breeding tree populations in response to predicted climate change factors;
- (2) to understand and exploit currently observed genetic x environmental interactions;
- (3) to increase selection accuracy in tree improvement programs.

#### DROUGHT TOLERANCE RESEARCH

1993 represents the third year of intensive physiological assessment of putative drought tolerant and intolerant full-sib families of black spruce. The families are part of a 7 x 7 diallele established on three sites at PNFI in 1973 by Dr. E.K. Morgenstern. The sites range in productivity apparently largely due to differences in water availability. Two of the families studied have maintained high productivity on all three sites (stable) while the other two (unstable) have had high productivity on the two wetter sites but not the dry site. Gas exchange measurements conducted over two seasons have shown the stable families to have higher net photosynthesis rates on both wet and dry sites and during both a very dry and a very wet year. As the families did not differ in stomatal conductance, this indicates differences in water use efficiency. Work done in cooperation with Dr. Larry Flanagan (Carleton University) using stable isotopes of carbon corroborated the gas exchange results. Furthermore, over three years the families have exhibited very similar water relations. All physiological evidence supports that the site with lower productivity is drier than the other sites. Thus, the family differences in growth on the dry site appear due to differences in carbon gain under carbon limiting conditions. Under wetter conditions, where carbon gain is not limiting growth, family differences in photosynthesis are not translated into growth differences. Grafting was conducted to increase ramet numbers so that original parents can be used to recreate original crosses and make new crosses for further field/greenhouse/growth room research.

#### BASIC CO<sub>2</sub> RESPONSE WORK

Two controlled-environment studies examined growth and ecophysiological responses of black spruce (*Picea mariana* (Mill.) B.S.P.) seedlings to elevated  $CO_2$  under varied water and nutrient additions. The purpose was to identify important traits to measure in subsequent genetic studies. Growth under elevated  $CO_2$  (700 ppm) increased final seedling dry weights by 20 to 48% compared with seedling growth under ambient  $CO_2$  (350 ppm). Percent increases in seedling dry weight were greater under drought versus well-watered conditions and higher versus lower nutrient additions. Seedlings grown under elevated  $CO_2$  displayed higher water use efficiency than seedlings grown under ambient  $CO_2$ . This was apparent based upon instantaneous gas exchange as well as XPP measurements. Elevated  $CO_2$ - induced stimulation of relative growth rate was greatest shortly after seedling emergence and decreased with increased seedling size. Acclimation of net photosynthesis (P<sub>n</sub>) was observed and was reversible.

#### WIDE-RANGE PROVENANCE WORK

John Seiler, a tree physiologist from Virginia Tech, joined the Project for the summer of 1993 to cooperate on a physiological experiment using a wide range black spruce provenance test. The purpose is to examine the effects of varied source and sink strengths on photosynthesis in black spruce. In the field component of the test, gas exchange of four diverse sources has been measured every two weeks since April 6. Measurements will be continued until early winter. In the greenhouse component, chambers have been constructed so that the seed sources can be grown in factorial combinations of varied photoperiod and  $CO_2$  concentrations. Both growth and physiology are being examined.

#### NARROW-RANGE PROVENANCE WORK

A series of growth room experiments are being conducted to examine the responses of Ontario black spruce provenances to climate change factors. Chamber conditions are being manipulated via a host computer to simulate both diurnal and seasonal weather. The current experiment is examining provenance responses to combinations of varied  $CO_2$  concentrations and water availability. A subsequent experiment will examine responses to drought and high temperature stress. Seed sources have been provided by Dr. Dennis Joyce (OMINR) and represent more cold and warm seed origin locations as well as dry and wet sites. Growth, gas exchange and stable isotope ratios are being measured. A custom built multiple plant, automated, computerized gas exchange system is being developed for this work to increase the precision, accuracy as well as the time-scale of gas exchange measurements. Plans are underway to collect seed and create clones to examine within-stand variation to climate change factors.

#### INHERITANCE OF ACQUIRED TRAITS

Black spruce seedling stock is being grown to be outplanted in a 1994 field trial to test hypotheses regarding the inheritance of acquired traits. Seed was the result of controlled crosses using seed sources moved from both northern and southern natural stands and planted at PNFI in 1964 (pollination utilized mother trees and pollen from distant provenances grown at PNFI as well as in the original stands).

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# **GENETIC DIVERSITY AND REPRODUCTIVE SUCCESS**

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Keywords: Conservation biology, genetic diversity, reproductive success and ecology, conservation strategies.

In 1992, the National Forest Genetic Resources Centre (NFGRC) was established at the Petawawa National Forestry Institute (PNFI) within the Forest Genetics and Biotechnology Program at PNFI. The NFGRC has been partially funded by the federal government's Green Plan, and has in turn funded a number of conservation-oriented research projects across Canada (British Columbia, Alberta, Quebec, New Brunswick and Newfoundland). A new research project entitled "Genetic diversity and reproductive success" (PI-62) was also established within the NFGRC to help address public concerns with the effects of land use practices, and particularly forestry practices, on the genetic diversity and reproductive success of forest populations. Initially, research will focus on (i) the effects of declining population numbers and population fragmentation on genetic diversity and reproductive success in trees, (ii) the identification of rare, vulnerable, and genetically unique tree populations for conservation purposes, and (iii) the development of strategies and recovery plans for conservation of rare and vulnerable tree species in collaboration with provincial land management agencies.

Red pine (*Pinus resinosa*) and white pine (*P. strobus*) will be the initial focus of research within project PI-62. Genetic diversity and reproductive success in disjunct populations of both species from Newfoundland have been studied over the past 4 years. These Island populations are declining rapidly in number due to excessive exploitation without proper forest management controls, and are being threatened with the introduction of diseases such as the white pine blister rust (*Cronartium ribicola*) and the scleroderris canker (*Gremmeniella abietina*).

Ecological studies on reproductive success, natural self-pollination, genetic variation, and gene flow in red pine have been conducted. In 1992, an Island-wide and rangewide sample of the red pine gene pool were established at two locations in central Newfoundland to preserve the gene pool. However, molecular genetic studies were unable to differentiate Island from mainland populations using isozymes and random amplified polymorphic DNA (RAPD) markers (Mosseler et al. 1991, 1992).

Similar studies have recently been initiated with white pine. Cones were collected in 1991 and 1992 from approximately 20 trees from each of 6 natural populations in Newfoundland and from 3 populations from the Ottawa Valley for comparative ecological studies on genetic diversity and seed production. Red pine and white pine are also at the centre of conservation efforts in Ontario where a strategy is being developed by the Ontario Ministry of Natural Resources (OMNR) to preserve "old growth" populations of both species.

The NFGRC, and especially PNFI's new project on genetic diversity and reproductive success, are also collaborating with geneticists and ecologists from OMNR in the

development of recovery plans for the conservation of rare, threatened, and vulnerable tree species in Ontario. Initial efforts will focus on the trees of the Carolinian Forest zone.

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# RESEARCH ON SEED SCIENCE AND REPRODUCTIVE DEVELOPMENT

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# Keywords: Sex expression, gibberellins, soil nitrogen, seed production, seed maturation, postharvest ripening, dormancy, storage, cryogenics, upgrading, germination, prechilling, moisture content.

The National Tree Seed Centre Project continued to carry out research on tree seed science and reproductive development and provide seed bank and associated services until April 1, 1992. After reorganization, research and development on seed and reproductive development has been assigned to a project carrying the title of this report and the seed bank and associated service functions have been assigned to another project. Both are components of the newly established National Forest Genetics Resources Centre. The project lost the valuable services of science contractor Bruce Downie, who started a Ph.D. program with Derek Bewley at the University of Guelph, and technician Moe Anderson who retired in September, 1991.

## RESEARCH AND DEVELOPMENT

## Seed Ouality

Post-harvest ripening: Results of 1988 were different from the 1984 findings. Nonetheless, a combination of post-harvest ripening of seeds in cones for 6 weeks and prechilling extracted seeds for 3 weeks to break dormancy appears to be a good practice for white spruce.

Upgrading seed quality: The IDS (incubation-desiccation-separation) technique proved to be effective for upgrading the germinability of jack, lodgepole, and eastern white pine seeds. Modifications of the technique are required before it can be applied to white spruce.

Effect of moisture content on prechilling treatment: Findings from collaborative research with the Alberta Forest Service on moisture content effect on prolonged prechilling of white spruce seeds indicated that the threshold moisture content for effective prechilling is over 20% (fresh weight), with the optimum around 25-27% (fresh weight). For laboratory germination tests, 3-week controlled moisture prechilling in aerated plastic bags was the best amongst all prechilling techniques used.

Prechilling effect on black spruce seeds: Subcellular electron microscope studies with Professor Berjak of the University of Natal demonstrated that the beneficial effect of prechilling on non-dormant black spruce seeds following accelerated aging was due to membrane repair. A report is in preparation.

## Seed Storage

Results of a recent study on the effects of seed moisture content (4.2-11.4% fresh weight) and storage temperature (+4° to -196°C) on the germination of white spruce seeds revealed that germination was affected significantly by all levels of seed moisture content except 11.4% at different storage temperatures after 6 months storage. The reduction in germination was due to induced dormancy.

In cooperation with Pierre Charest and Bruce Downie, Ben Wang authored a review of "*Ex situ* storage of pollen, seeds, and *in vitro* cultures of perennial woody plant species" for FAO.

## Seed Production

Seed orchard soil fertility: Application of  $NH_4NO_3$  to white spruce seed trees did not promote sexual development whereas application of carbofuran, a systemic insecticide, to soil to protect foliage and seeds was associated with increases in seed cone bud production. In experiments with jack pine, sexual reproductive responses were not always observed as a result of applying  $NH_4NO_3$  to soil and spraying foliage with  $GA_{4/7}$  even though soil and foliar nutritional responses were consistently observed.

Containerized seed trees: Pollen cone production in 6-year-old containerized jack pine trees was very sensitive to modification of the soil nitrogen supply but relatively insensitive to foliar sprays of  $GA_{4/7}$ ; the opposite was observed for seed cone production. Pollen cone production was suppressed by optimum to high levels of soil nitrogen and stimulated by low levels. Seed and pollen cone production was depressed by low soil moisture tension. Intact trees outside polythene shelters did not display sexual responses to nitrogen or  $GA_{4/7}$  treatments. In another experiment, growing conditions were found that allowed seed cone production on up to 80% of trees less than 2 years from seed and pollen cone production on up to 30% of these trees. Assessment of treatment effects for two years revealed that the sexual response to soil nitrogen supply persisted whereas the response to  $GA_{4/7}$  application did not.

Stem incorporation of  $GA_{4/7}$ : Stem injections and implants of  $GA_{4/7}$  were evaluated for promotion of sexual reproductive development of white spruce, Norway spruce, and jack pine seed trees. Seed strobilus production was promoted by injections and implants in white and Norway spruce. Treatments were less effective in promoting development of pollen strobili and vegetative shoot production was reduced when pollen strobili were promoted. In jack pine, pollen but not seed strobilus production was promoted by treatments.

*Crown pruning:* Observations for four years following stem pruning of jack pine seed trees revealed that production of seed strobili was not reduced by light pruning, whereas significant reductions were observed on severely pruned trees. Trees reacted to topping by increasing production of seed strobili on the uppermost branches remaining on the trees at the expense of pollen strobili. The senescence of old branches was reduced by both levels of topping to about half of that on control trees.

Other: Allocation of resources to sexual development competes with allocation to growth and defensive functions. Observations on jack and white pine indicate that defensive functions display high degrees of intra- and inter-tree variability and comparison of several alders revealed that fast-growing species can have high levels of resistance to herbivory. Information on the physiology and genetics of tradeoffs among allocations to growth, sex, and defensive functions is required.

## TECHNOLOGY TRANSFER

Ben Wang provided technical consulting services to CIDA's seed projects in S.E. Asia and the southern African countries. He also attended the 23rd ISTA Congress held in Buenos Aires, Argentina, and the IUFRO/CNSF Symposium and Workshop in Burkina Faso. Hugh Schooley continued to act as the editor of the CTIA Tree Seed Working Group News Bulletin. Current ongoing cooperative research with universities, provincial forest services, and industrial agencies continues to provide excellent opportunities for technology transfer.

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# GENETIC RESOURCES MANAGEMENT IN ONTARIO OPERATIONAL PROGRAMS AND SCIENTIFIC SUPPORT

# Cathy Nielsen, Joan Wild, Randy Ford, Paul Charrette, Bob White, Frank Schnekenburger, Jim Hood, Rong Ho, Dennis Joyce, and Peter Nitschke

## OPERATIONAL PROGRAMS

## The Ontario Tree Improvement Board

Intensive tree improvement, seed and stock deployment, and genetic diversity concerns are coordinated through the Ontario Tree Improvement Board. The Board, which was legally incorporated in April of 1993, provides an umbrella organization which embraces the entire province. The province has been divided into 6 zones, each of which receives the support of a specialist. Operational programs are carried out by zonal cooperative associations which currently involve the Ministry of Natural Resources (MNR) and local forest industries. The current structure includes former Ontario Tree Improvement Council members, the North Shore Tree Improvement Cooperative, and many new industry partners.

Each of the zonal associations is responsible for developing their own program. Program priorities, objectives, and work plans are developed jointly by the zonal members. All zones are currently drafting Memoranda of Understanding and have set up Steering Committees to provide program direction and Operational or Technical Committees to coordinate implementation. The Ministry of Natural Resources provides scientific and operational program advice and support to the zones.

## Zone 1: Lake of the Woods - English River

This Association manages 24 seed orchards of 3 species along with their associated tests. The current emphasis of our program is on maintenance and management of existing sites and operational seed collection.

## Black Spruce

Eleven black spruce seed orchards were established in 1982 and one was established in 1989. Measurements have been completed on eight of the 1982 orchards and family tests. Analysis is being completed this winter with rogueing to be completed in 1993. An additional three orchards and six family tests are scheduled for measurement and analysis in 1993 for rogueing in 1994.

Significant cone harvests are occurring in the orchards. A Cone Monitoring System developed by Forestry Canada is being used in Fort Frances, Sioux Lookout and Red Lake Districts to improve cone crop prediction and protection programs in the orchards. Some of the orchards are scheduled for crown pruning after rogueing this fall.

A black spruce genecology study has been funded by the Association and will be initiated by Lakehead University in 1993. This study will be used to determine focal point seed zones for this species.

## Jack Pine

Six jack pine orchards and 17 family tests have been established (1989-1991) and are being maintained. Two family tests are scheduled for first measurement this fall.

Two crown pruning trials have been established, and preliminary results from one have been analyzed. Fertilizer calibration trials have been established in Red Lake, Sioux Lookout and Fort Frances Districts orchards.

A genecology study was initiated in 1992.

## White Spruce

Six clonal orchards have been established and are being maintained.

# Zone 2: LAKE NIPIGON WEST

The cooperative tree improvement programs in jack pine and black spruce have shown excellent progress and have established all first generation seed orchards and associated family tests. The members are now concentrating their efforts on intensive management of existing sites and activities towards the second generation.

#### Black Spruce

In the breeding zone to the west of Thunder Bay (i.e. 4400 breeding zone), a 12 ha seed orchard (MNR) and three family tests (2 Canadian Pacific; 1 MNR) were established in 1991. All three tests suffered low levels of mortality and damage after planting but now appear to have stabilized and are growing well. Survival and growth have been very good in the orchard.

The three family tests (2 Abitibi-Price; 1 MNR) in the breeding zone north of Thunder Bay (i.e. Lake Nipigon West), will be measured later this summer. An additional eight ha seed orchard will be established in the spring of 1994, at the Thunder Bay Forest Nursery (MNR).

#### lack Pine

In the spring of 1992, one 10 ha seed orchard (MNR), and 3 family tests (2 Canadian Pacific; 1 MNR) were established in the 4400 breeding zone. Stock losses during over winter cold storage will require a fill-in planting in the orchard in 1994.

Results from fifth year measurements of 3 jack pine tests (2 Canadian Pacific; 1 Abitibi-Price) in the Lake Nipigon West breeding zone were quite favourable, and the first rogueing in the seed orchard (Canadian Pacific) is scheduled for the spring of 1994.

#### Breeding Orchard

Work is on-going in establishing the breeding orchard (MNR) for jack pine and black spruce from each breeding zone with clones of the parent selections. The breeding orchard also serves as a clonal archive for older selections of black spruce and white spruce. Deer browsing of jack pine clones during the winter of 1991-92 caused considerable mortality. The breeding orchard was treated this past fall with a browse repellant which appears to have reduced the incidence of browsing.

# Zone 3

Work was initiated to form a local Seed Management Association of Forestry Industry companies and the Ontario Ministry of Natural Resources. Company and Government representatives have met to develop a Memorandum of Understanding and prepare a draft Seed Management Plan.

Recently, activities have been directed towards seed collection programs for the major commercial tree species. However, in 1956, an intensive selection program of both black spruce and white spruce plus trees was initiated that was continued until 1980. This material has been established and maintained in orchards and is available for use in future intensive programs.

## Zone 4

The focus of our intensive tree improvement efforts is currently on the maintenance of existing sites and production of orchard seed. In addition, emphasis is now being put on the deployment of seed and seedling stock to avoid maladaption. The Association has 8 members and includes members of the former Ontario Tree Improvement Council.

#### Black Spruce

Fertilizer calibration trials were put in place in the seed orchards to determine acceptable nutrient levels for the trees during the establishment through cone production stages of the orchard. Some preliminary results have been obtained from the analysis of the first year's data from one test.

In an attempt to moderate the soil temperature and improve moisture availability, poplar chip mulch was placed around 30,000 trees in an orchard with poor soil conditions. Subsequent temperature measurements showed that the mulch had a dramatic beneficial effect.

A genecology study has been planted in two locations; one representing a severe climate and one representing a moderate climate. The trees have been assessed once and a cursory investigation of the summarized data shows trends which appear to correlate with some climate parameters.

One farm field test was planted in 1991 and another in 1992 to supplement data obtained from conventional field tests. Family heritabilities for total height are .76 or higher in the 1991 test.

A male flower induction study has been initiated in cooperation with Dr. Rong Ho at the Ontario Forest Research Institute. First results in the spring of 1993.

#### lack Pine

The seed orchards for Breeding Zones 1 and 3 have been rogued; approximately 30% of the trees were removed from each orchard. The rogued trees were removed from the orchard and chipped.

The Breeding Zone 4 orchard has been rogued and the remaining trees were later topped to facilitate cone collection and to encourage the crowns to grow laterally. The first cone crop to develop since rogueing has been picked.

Two farm field tests have been planted and will be assessed for the first time since planting in the fall of 1993.

A cone crop monitoring system has been implemented in three orchards and is expected to predict cone and seed production and identify their damaging agents.

The climate, pollen flow and contamination study at the Island Lake Tree Improvement Area is completed; the finished model and associated computer software is expected to be completed in the winter of 1993/94.

A genecology study has been planted in the same two locations as the black spruce study and is to be measured for the first time in the fall of 1993. This study is a cooperative effort with the North Shore Tree Improvement Association and the Ontario Forest Research Institute.

## Zone 5: NORTH SHORE

The North Shore Tree Improvement Association was formed eight years ago. The members include 4 companies and 5 MNR Districts. Activities in the past two years have concentrated on maintenance of first generation orchards and tests. We have also begun the first rogueing in jack pine orchards.

## lack Pine

A genecology study was started in 1991 in cooperation with Zone 4. Information from this study will be used to realign breeding zone boundaries and determine appropriate seed transfer rules. In conjunction with this study, 5-year measurements were taken on 8 stand tests in the fall of 1992.

A cone crop monitoring program has been implemented in 2 of the orchards. Six family tests and one orchard were measured in the fall of 1991. This information was used to carry out a 30% rogueing in 2 seedling seed orchards in 1992. Another 8 family tests and one orchard were measured in the fall of 1992, and will be used to rogue 3 orchards in 1993. Most jack pine family tests have suffered damage from white pine weevil and pine shoot borer.

A crown management study is underway in one of the orchards to determine a suitable height for topping, assess possible physical damage to topped trees, and evaluate the effect on cone production.

## Black Spruce

Two seedling seed orchards and five family tests are in place. Initial survival in these orchards has been less than expected. Mulching in the second year however, increased survival considerably. Activities in the last two years have concentrated on general maintenance including tending, fertilizing, and mulching.

## White Pine

Three clonal seed orchards are planned. Two of these have been established with the third set for initial planting in the fall of 1993. Activities have included fertilizing, mulching, irrigating and vegetation management. A genecology study is planned in cooperation with Zone 6. Cone collection for this will begin in 1993.

## White Spruce

The clonal seed orchard for breeding zone 4200 is almost complete. An irrigation system has been installed. Armillaria continues to be a problem in this orchard. A fertilizer calibration trial was established in 1992.

## Zone 6

As a result of reorganization within the MNR and the establishment of OTIB, Zone 6 was formed to encompass all of Southern Region as well as four districts in Central Region. Responsibility for coordination of the forest genetics program in Zone 6 lies with the Southern Ontario Forest Genetics Group based out of Brockville.

One of the main activities of the group has been to realign the scope of the forest genetics program to encompass gene conservation activities as well as the traditional intensive tree breeding programs that have been pursued in the past. In conjunction with realignment of program direction, the establishment of a Forest Genetics Cooperative has been initiated. Potential partners for all components of the program have been contacted. Establishment of the cooperative and development of specific program goals and objectives will continue to be the highest priority for Zone 6 in 1993.

#### Intensive Tree Improvement

#### White Pine

White pine is the highest priority species for tree improvement in Zone 6. One thousand and three hundred grafts were planted in the Kemptville seed orchard in 1992, with an additional 900 grafts to be planted in 1993.

An archive will be established in the spring of 1993 in cooperation with the National Capital Commission who have provided a site within the greenbelt surrounding Ottawa and will assist in maintenance of the grafts.

Breeding for white pine during 1991 and 1992 has concentrated on completing polycrosses for clones represented in production orchards. A total of 241 crosses were completed across 4 site regions. In addition, 131 open pollinated collections were made from clones in the Grattan seed orchard for testing purposes.

#### Norway spruce

A total of 3190 Norway spruce grafts were completed in 1992 and 1993 to complete grafting of 200 selections which make up the breeding population for the southern Ontario program. The grafts will be grown in the breeding hall as potted stock to be used for test crosses and eventually production crosses to supply a vegetative propagation program.

## <u>Larch</u>

Breeding has continued in the exotic larch program. The objective is to test the Japanese and European clones to determine which clones have the highest breeding values within each species. The best performers will then be used in hybrid production crosses for a hybrid larch vegetative propagation program. Ninety-one and one hundred and ten crosses were completed in 1991 and 1992, respectively. Cone crop monitoring was conducted in the hybrid larch seed orchard in 1991 and 1992.

## Red Spruce

Planting of the Rideout Township red spruce seed orchard in Bancroft District was completed in 1991. The orchard contains 116 clones.

## Hybrid Poplar

Material resulting from ten years of eastern cottonwood breeding was converted to cuttings which are now being rooted in a greenhouse. The rooted cuttings will be planted in Kemptville nursery for future use. The breeding strategy for hybrid poplar is now undergoing a review to determine the most efficient way of meeting the hybrid poplar tree improvement objectives.

## Gene Conservation

Gene conservation activities in Zone 6, are supported by Genetic Heritage, a program under MNR's Sustainable Forestry initiative. Activities in Zone 6 under this program include the completion of six status reports on rare Carolinian species, the completion of mapping of remaining white pine stands in southern Ontario, grafting for the completion of a white spruce gene bank and ninety-six seed collections from stands across Zone 6 to be utilized in a white spruce genecology study. Information generated through the projects noted above will serve as background information for gene.conservation strategies which are being developed by a provincial committee under the leadership of Dennis Joyce, Provincial Geneticist for MNR.

# SCIENTIFIC SUPPORT

Over the last three years, the Ontario Ministry of Natural Resources has been reorganizing to meet the challenges of forest management in the future. As part of this initiative, the role of the Ontario Forest Research Institute within the ministry is to provide science-based information and tools which are timely, relevant, and useable, and which promote the scientific integrity of forest policy and management. As a result, strong ties are being forged between research and operational program.

The Institute's Genetic Resource Management Program provides scientific expertise and leadership in the continuing development of the operational tree improvement program and in the development of policy and guidelines for management of genetic diversity and gene conservation.

The current program is divided into 5 projects:

- \* Data Management,
- Quantitative Genetics,
- \* Reproductive Biology,
- \* Genecology,
- Genetic Diversity.

#### Data Management Project

Tree improvement data management in the province is currently focused on the following areas: 1) analyses of family test data for the purpose of rogueing first-generation production seed orchards, 2) development of a comprehensive, provincially-networked tree improvement database management system, 3) development of supplementary data management systems, such as the Cone Crop Monitoring System for seed orchards, and

4) development of decision support systems, such as the seed allocation support system "Seed where".

## Quantitative Genetics Project

Studies in the Quantitative Genetics Project, some of which were carried over from the old spruce breeding project, deal with the estimation or prediction of genetic parameters such as: genetic components of variance, genotype by environment interaction, genetic correlations, and genetic gain. Four field studies used pedigreed families of white and black spruce and three of these also employ clones. Tenth-year growth data will have been collected from all trials by the end of 1993. One farm-field trial of white spruce investigating the inheritance of growth rhythms of shoot elongation and radial growth is undergoing final analysis. The remaining study uses computer simulation models to investigate the probable results from different tree breeding and selection schemes, the magnitude of effects of different genetic parameters and the sensitivity of different breeding alternatives, including breeding, selection and testing procedures, to departures from underlying genetic assumptions.

## Reproductive Biology Project

The goal of the Reproductive Biology Project is to develop operational coneinduction technology and a crown-management procedure for potted and land-based orchard trees of economically important conifers, and to understand the mechanisms of sexual reproduction.

## Genecology Project

The goal of the genecology project is to identify the adaptive strategies of native forest tree species and develop models to describe patterns of adaptive variation. The information being developed in this project will be used to develop biologically sound seed transfer guidelines; refine breeding zone boundaries, and for developing management plans addressing climate change concerns. The oldest study (black spruce in northeast Ontario) is three years old and has one more year of data collection. A jack pine study (north of Lake Huron) is in its second year, and a white spruce (southern Ontario) was sown in the spring of 1993.

## Genetic Diversity Project

Under the Sustainable Forestry Initiative, the Genetic Heritage program has three goals. The first goal is to establish a process for developing recovery plans for tree species that are vulnerable to extirpation forces because of small population sizes. The focus of this effort is to work with species before they require expensive, labour intensive efforts for their conservation. Candidate species are being identified and status reports are being written for priority species. A process for developing recovery plans based on these status reports has been identified and the first plans should be completed in 1994. The second goal is to develop management guidelines for managing genetic diversity in the landscape. Work toward developing guidelines is drawing together relevant scientific principles from the fields of forest genetics, population genetics, quantitative genetics, and conservation biology. A set of guidelines should be available by the conclusion of the Sustainable Forestry Initiative in 1995. The final goal is to develop scientific information on the patterns of adaptive variation in trees. State-of-the-art climatic models are being developed in collaboration with Forestry Canada. Studies of genetic variation in growth rhythm and cold hardiness among populations of trees are in progress and will be related to climatic variation. This information will be used for management concerns such as growth and yield and climate change.

# FOREST GENETICS RESEARCH AT THE FACULTY OF FORESTRY, UNIVERSITY OF TORONTO

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The Faculty of Forestry at the University of Toronto offers undergraduate courses in forest genetics and tree breeding at the second and fourth year level. A graduate course in forest genetics is offered as well.

Research at the Forest Genetics Laboratory (FGL) is coordinated by L. Zsuffa, Professor of Forest Genetics. The laboratory currently has a graduate student complement of two Ph.D. students, and one Master's student. During the past two years, three Master's candidates and four Ph.D. candidates defended their theses. Also contributing to studies are an Assistant Professor (W.A. Kenney), a Research Associate and Adjunct Professor (R.L. Gambles), and a Research Technician (B.J. Vanstone).

Under the direction of Dr. Louis Zsuffa, research in the FGL includes the genetic improvement, characterization and evolutionary studies of willow, poplar, white pine, and black spruce.

Research at the FGL, has involved: progeny and clonal testing; inter- and intraspecific hybridization; biomass quality studies; assessment of species and clonal variation in nutrient requirements and uptake; resistance/susceptibility to diseases and pests; plant regeneration from callus cultures; isozyme and cp-DNA studies; and linkages and inheritance patterns. Graduate and postdoctoral studies contribute considerably to this research.

This report outlines some of the current research being undertaken at the FGL.

## SHORT ROTATION INTENSIVE CULTURE (SRIC) AND BIOENERGY RESEARCH

Willows are important in forest genetics research because of their fast growth, wide distribution, richness in genetic diversity (almost untouched natural gene pool), adaptability, and ease of propagation by stem cuttings. The advent of short rotation, intensively managed biomass plantations has resulted in an interest in the potential of willows to supply part of the world's future demand for wood fibre, energy, chemicals, and food.

The FGL is active within the International Energy Agency (IEA). The Bioenergy Agreement of the IEA was developed to coordinate an international cooperative research programme. L. Zsuffa, with the assistance of R. Gambles, coordinates the IEA/BA Task VIII "Efficient and Environmentally Sound Biomass Production Systems". As a part of Task VIII, the Genetic Improvement Activity is run by the FGL. Its objective is to develop high yielding and environmentally acceptable stock for short rotation forestry biomass production systems.

The FGL is also active with IUFRO working groups on integrated research into biomass, poplar breeding and provenances, and white pine breeding and provenances. We have contributed significantly to the work of the Poplar Council of Canada, and the International Poplar Commission of the Food and Agriculture Organization.

# Improvement of North American Salix L. for Biomass Production by Hybridization and Clonal Selection

The main objective of this research is to develop superior clonal varieties of North American willow species for short rotation biomass energy plantations via hybridization and testing.

In the early 1980's, selections of parental species and genotypes were made from across the range of willows in North America. Inter- and intraspecific hybrids were made and valuable species combinations were identified; the hybrids were established in family/clonal screening trials and research was performed on the identification and evaluation of the best clonal selections, based on biomass production, feedstock quality (moisture content, specific gravity), growth habit, disease resistance, frost hardiness and the presence of genetic x environmental interaction. Relationships between biomass qualities and growth yield were determined and provided selection criteria for multiple traits important for clonal selection and utilization.

## **Demonstration Farms**

The logical next step in the development of short rotation bioenergy plantations, was to establish a demonstration farm. As the interest in the production of energy feedstocks through SRIC has grown, so has the need for larger scale plantations to: demonstrate the concept of SRIC; provide information for SRIC financial analyses; determine overall performance; provide a venue for mechanization of trials; etc. This need has been recognized both internationally and nationally.

A five-year contract to propagate the highest yielding clones in order to develop demonstration energy farms was initiated in 1991 with joint funding, from: Ontario Ministry of Energy (MOE); Ontario Ministry of Agriculture and Food (OMAF); Energy, Mines and Resources, Canada (EMR); Agriculture Canada; and Forestry Canada, with in-kind contributions from the Ontario Ministry of Natural Resources (OMNR). Two demonstration farms, of about 2 hectares each, planted at a spacing of 15,000 stools/hectare, were established in the spring of 1993. The design of these farms is similar to production plantations in Sweden, so that comparisons can be made with operational scale plantations, and so that mechanization of harvesting can be facilitated.

The establishment of these prototype farms represents the culmination of several years of research into the testing and screening of willow clones for energy production. However, as with the development of any "domesticated" crop, it is essential that continued breeding be carried out to introduce new varieties into the production population to ensure continued gains in yield, quality, and pest resistance, and to develop clones for specific site conditions.

# Disease Resistance/Susceptibility

Resistance to disease is an important characteristic to observe in any plant breeding programme. In North America, willow breeding for increased biomass production is in its early stages, and it is important to establish the degree of susceptibility, and the variability in that susceptibility, to diseases. Disease surveys, carried out on our field trials, have included observations of *Melampsora* Theum. leaf rust. Rust can become a serious problem in willow biomass plantations, causing premature leaf fall, reducing growth and weakening the host. Currently we are participating in the IEA Rust/Clone Interaction Trial. Identical trials were planted in Europe and North America, using common European and North American clones, in order to compare disease (particularly *Melampsora* rust) and pest resistance/susceptibility and growth.

Another study, undertaken by B. Beatson as her M.Sc.F. thesis, investigated genetic resistance to disease by establishing the degree of variability in willow species, family and clonal response to attack by the pathogen.

## **TREE-FORM WILLOWS**

Research was initiated on tree-form willows in 1990. Tree-form willows are fastgrowing and promising for a variety of industrial products. They are easily harvested and processed using current industrial technology. Domtar Inc. demonstrated its interest in this research by offering land and maintenance assistance for tree-form willow trials.

The two major goals of the project are to study tree-form willows by: 1) establishing demonstration trials of cloned plus trees from a natural stand and superior trees selected from arboreta; and 2) by characterizing them genetically, using DNA and electrophoretic techniques.

Two demonstration trials were established in the spring of 1991 in Iroquois, Ontario (in cooperation with Forestry Canada, PNFI); in 1992 in Berwick, Ontario, in cooperation with OMNR. The FGL has been awarded a contract to take over responsibility for the entire 1991 Iroquois trial, which contains poplar, silver maple and alder, as well as willow. This will allow growth comparisons within and between the genera.

A study, entitled "The effect of ectomycorrhizae on the growth and macronutrient uptake of some clones of *Salix alba* L. and *S. nigra* Marsh." was completed in 1991.

# RELATIONSHIP BETWEEN BASIC DENSITY AND WOOD ANATOMICAL CHARACTERISTICS IN SRIC PLANTATIONS OF SALIX

This study is being undertaken by P. Bhojvaid as his Ph.D. thesis. Basic density (specific gravity) is important in wood quality analyses. High specific gravity is correlated with wood strength properties and pulp yields. Its importance is further elevated because it can be modified by silvicultural operations and genetic manipulation. An energy feed stock with high specific gravity is expected to be more suitable for energy conversion. Charging a digester with a feedstock of high basic density will result in more substrate present per unit time, making the conversion process more efficient.

Specific gravity has generally been shown to be negatively correlated with growth rate. However, evidence does exist for the presence of genotypes which exhibit both these traits. Basic density is a complex trait and is a function of the combined effect of many

other chemical and physical traits in tissues, however, it has been shown to exhibit a high heritability. Little is known about what chemical and physical properties govern this trait. A better understanding of these relations would aid in: selecting species and clones with superior specific gravities; explaining relationships between growth and specific gravity; and understanding the processes involved in the development of genotype by environment interaction.

The objectives are to: determine relationships between wood density and wood anatomical and chemical characters in *Salix* coppice; and to determine the role of moisture stress on the basic density of wood.

# GENETIC CHARACTERIZATION OF SALIX

The progress of breeding research in willows is hindered by the lack of knowledge on the basic genetics of *Salix* species. The following studies outline research undertaken in the FGL in the area of genetic characterization.

# Identification of poplar and willow clones using DNA fingerprinting

This inquiry is being coordinated by L. Zsuffa, under the IEA/BA Genetic Improvement Activity. Several researchers have worked on this project. The study was initiated because the development of molecular and biochemical markers is necessary for reliable and efficient genetic identification, certification and preservation of poplar and willow clones, and for effective genetic improvement.

The work is progressing well, and has received further support from the IEA/BA.

## A review and assessment of genetic improvement and a strategy proposal for Salicaceae

This comprehensive review is being undertaken by L. Zsuffa, to review past research in genetics and breeding of Salicaceae (poplars, aspen, and willows), evaluate results and needs, and recommend a strategy for further genetic improvement.

# Dynamics of isozyme electrophoretic spectra in families of Salix exigua Nutt. and S. eriocephala Michx. and their implementation in willow breeding

This Ph.D. thesis was completed by F.A. Aravanopoulos in the spring of 1992. The objectives of this research were: to study inheritance and linkage of isozymes in *Salix exigua* and *S. eriocephala*; to investigate tissue differentiation in these species by comparing isozymes of root tips and fresh leaves; and to compare their allozyme heterozygosity with growth parameters and biomass production.

Fifty-two isozyme genes and 70 alleles were revealed in 26 enzyme systems. Mendelian inheritance and co-dominant allelic expression were verified in 11 variable loci of *S. eriocephala* and 13 variable loci of *S. exigua*. Two linkage groups were discovered in *S. eriocephala* and three in *S. exigua*. Map distances and chiasma interference estimates were derived. Relationships between alleloenzyme heterozygosity and biomass production were investigated. Positive associations were revealed in *S. eriocephala* but not in *S. exigua*.

The discovery of the linkage groups will assist in locating other genes of interest. The positive heterozygosity-biomass associations in *S. eriocephala* and their possible theoretical explanations could affect future breeding approaches and bridge biochemical genetics with breeding efforts.

# <u>Phylogenetic relationships of willow species based on allozyme variation and chloroplast DNA</u> <u>diversity</u>

This study, completed in 1992 by K.X. Chong as his Ph.D. thesis, investigated: genetic structure, similarity and diversity of congeneric *Salix* species based on allozyme variation and cpDNA diversity; phylogenetic relations and evolution of willows using isozymes and cpDNA markers; and the identification of isozymes and cpDNA as genetic markers for the species used. Nine taxa, including eight North American and one European species were studied.

Levels of isozyme genetic variation differed greatly among species. Tree-type willows generally maintained higher genetic variation than shrubby willows. High genetic identity between *S. amygdaloides* Anderss. and *S. nigra* suggested a progenitor-derivative species pair which has undergone a rapid diversification.

# Quantitative genetic parameters in seven characters of Salix eriocephala

This study, undertaken by L. Jingzhong, as his M.Sc.F. thesis, investigated quantitative genetic parameters in twenty clones, four from each of five full-sib families from two test sites in eastern Ontario. The characters evaluated were stem height, stem basal diameter, number of stems, number of branches, moisture content, specific gravity and plant biomass.

Characters fell roughly into three heritability classes. Stem height, stem basal diameter and biomass weight were in the low heritability group; number of stems and number of branches were in the medium group; and moisture content and specific gravity demonstrated high heritabilities. Analysis of covariance revealed that biomass weight was positively correlated with number of stems, number of branches and moisture content. Specific gravity was negatively correlated with stem height, stem basal diameter and moisture content.

# Variation in organogenic capacity within and among three willow species under *in vitro* tissue culture

This project, undertaken by F. Liu as his M.Sc.F. thesis, investigated *in vitro* cultures of stems and calli, and analyses of chromosomes and isoenzymes in three willow species, in order to examine organogenic capacity and somaclonal variation.

Significant variation within and among species was found. Organogenic capacity was affected mainly by genotype and interaction between genotype and medium. A positive correlation existed between callus initiation capacity and cell enlargement versus division ability. Somaclonal variability varied with genotype and culture age, and decreased regeneration capacity. Genetically stable yellow calli had a potential for organo- and embryogenesis. Genotypes of nine clones investigated were identified. Each genotype had its own optimal treatment combination and concentration range of hormones for best expressing its organogenic capacity.

# GENETIC CHARACTERIZATION OF SOME WHITE PINE SPECIES AND HYBRIDS AND ITS RELATIONSHIP TO BLISTER RUST RESISTANCE

This research was undertaken by E. Chagala as her Ph.D. thesis. The results of this study aid in the proper identification of breeding stock, combination of desired traits and

certification of controlled crosses; they lead to a better understanding of the genetics of white pine, and may divulge information on the genetic control of blister rust resistance and other important silvicultural traits.

The genetic structure, linkage relationships and genetic variation of five white pine species were investigated by isozyme analysis using starch gel electrophoresis, based on tissue from mature needles and megagametophytes. Methods for using isozyme analysis for the identification of these species and interspecific hybrids were also investigated. Thirteen enzyme systems were analyzed, most of which were shown to be under multiple gene control. Thirty-six loci coded for the 13 enzyme systems in each of the species. Inheritance studies showed that most of the isozyme variants were under Mendelian single-gene control.

Up to three linkage blocks were determined in each species, with most of the linked genes being on the same chromosome. The mean values of average heterozygosity, proportion of polymorphic loci, and number of alleles per locus were calculated and found to be comparable to those of other conifer species. Genetic similarities and distances, and divergence times, based on allele frequencies, were calculated. These results, along with results from a cluster analysis, showed that *P. strobus* L., *P. monticola* Lamb. and *P. griffithii* McClell. were genetically the closest species while *P. peuce* Griseb. and *P. koraiensis* Sieb. & Zucc. each formed a separate cluster. Several alleles were identified as markers for all of these species and their interspecific hybrids.

# GENETIC MARKERS FOR GROWTH, DROUGHT TOLERANCE AND RESPONSE TO ANTIOXIDANT ACTION IN BLACK SPRUCE

This research, undertaken by K. Vishnevetskaia as her Ph.D. thesis, investigates black spruce (*Picea mariana*), one of the most commercially important forest tree species of Ontario. Extensive research has been directed toward the production of seedlings that are morphologically suited for drought-prone sites and that contain genetically determined drought tolerance. Recent innovations in biochemical and molecular biology techniques used in the study of the genetics of forest species have led to major advances in forestry breeding. Since the selection of individuals is often based upon traits which are not expressed until the plant is mature, one must rely on genetic markers. Polymorphic proteins (enzymes) and restriction fragment length polymorphisms (RFLPs) could play a role as such markers.

In order to use antioxidants for improving the growth and drought tolerance of conifers, the interrelationships between seedling development, physiology and genetics need to be elucidated.

The objectives of this research are to: determine fast growing and drought tolerant genotypes of black spruce; investigate the relationship between heterozygosity and growth; and heterozygosity and drought tolerance; determine genotypes with the most significant increase in growth and drought tolerance as a result of antioxidant application; and, using the information derived from the above, to indicate the potential use of heterozygosity as an early selection criterion and as a marker for antioxidant response.

Further objectives are to: investigate linkage relationships among allozyme loci affecting growth and drought tolerance using enzyme markers; construct restriction fragment length polymorphism maps; and to identify loci that code for drought tolerance and growth.

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# MANITOBA'S TREE IMPROVEMENT PROGRAM

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Keywords: Jack pine, black spruce, white spruce, seed orchard, co-operative.

Tree improvement programs have been established for jack pine (*Pinus banksiana* Lamb.), black spruce (*Picea mariana* (Mill.) B.S.P.) and white spruce (*Picea glauca* (Moench) Voss) in the most active breeding zones of Manitoba. These programs were established through co-operatives between the Manitoba Forestry Branch and Forestry Canada, Abitibi-Price Inc. and Repap Manitoba. New co-operative programs are being established for the second priority zones and supportive research is being completed by Forestry Canada through the Canada-Manitoba Partnership Agreement in Forestry. This report highlights the progress which has occurred from 1991 to 1993.

## JACK PINE

The 20-year measurement of the Eastern Breeding District family test was completed in 1991. Forestry Canada is analyzing the data to identify the single-pair matings for establishing the second generation improvement program.

The first roguing of the Interlake Region mass selection orchard was initiated in 1992 with 50% of the trees removed based on their on-site performance. The second roguing of the Northern Region mass selection orchards will be completed in the fall of 1993. Collectable cone crops are expected from these orchards in the next two to three years.

# BLACK SPRUCE

Tending existing family tests and seed orchards has been the major priority during the past two years. A new co-operative program was initiated between Repap Manitoba and Manitoba Forestry Branch for black spruce in the Nelson River Breeding Zone. Three black spruce family test plantations were established in 1993 and a 7.0 hectare seedling seed orchard site selected for planting in 1994.

## WHITE SPRUCE

Repap Manitoba and Manitoba Forestry Branch crews selected 175 white spruce plus trees in the Saskatchewan River Breeding Zone. Scions from the trees have been grafted to establish a container seed orchard.

Species	Breeding Zone	Plus Trees Represented	Family Tests Established	Seed Orchards Established	Clone Bank
Jack Pine	Eastern Mountain Interlake Northern	209 214 320 320	4.5 ha (4 sites) 1.0 ha (1 site)	6.0 ha 5.0 ha 4.4 ha	0.1 ha
Black Spruce	Southeast Lk. Wpg. East Interlake Sask. River Nelson River	400 384 171 445 423	14.3 ha (3 sites) 10.9 ha (3 sites) 13.8 ha (4 sites)	7.2 ha 3.8 ha 7.2 ha (7.0 ha cleared)	0.6 ha 0.8 ha
White Spruce	Mountain Sask. River	355 175	9.0 ha (4 sites) 11.1 ha (3 sites)	(4.5 ha cleared)	0.5 ha
Total		3,416	64.6 ha	33.6 ha	2.0 ha

# Table 1. Manitoba's Tree Improvement Program Summary

Supportive research projects have been initiated by Forestry Canada through the Canada-Manitoba Partnership Agreement in Forestry. Projects include the development of a container seed orchard program for jack pine and white spruce, the establishment of a jack pine gain test and the vegetative propagation of jack pine rooted cuttings at the Morden Research Station.

# TREE IMPROVEMENT UNDER THE CANADA-MANITOBA PARTNERSHIP AGREEMENT IN FORESTRY

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Keywords: Pinus banksiana, Picea mariana, jack pine, black spruce, seed orchards, mass selection, pair mating, potted seed orchards, rooted cuttings, best linear prediction, wood density, stem quality, western gall rust.

Enhancement of Forestry Canada's tree improvement activity in Manitoba under the Canada-Manitoba Forest Renewal Agreement of 1984-89 will continue under the Canada-Manitoba Partnership Agreement in Forestry (PAIF) signed in March 1991. Albert Nanka, who coordinated the establishment of jack pine (*Pinus banksiana* Lamb.) seed orchards in Manitoba under the 1984-89 agreement, has returned to Edmonton. Jerry Klein and Paul Chapman were relocated to the Manitoba District Office (MDO) to implement tree improvement activities under the new agreement. These activities are intended to (1) continue establishment and development of jack pine seed orchards, (2) develop more efficient delivery of genetic improvement, especially by controlled pollination and vegetative propagation, and (3) initiate a second generation of jack pine breeding.

# JACK PINE SEED ORCHARDS

Mass selection seed orchards were planted in the Northern and Interlake regions of Manitoba in 1986 (Nanka 1991) and 1987. Plots planted with 48 or 25 trees from one stand at 1m spacing will be thinned in four mass selection steps to 1-3 trees. A sample of trees are measured at each thinning for assessment of relative genetic quality of the 32 source stands for each seed orchard, and of the phenotypic effect of mass selection. The first thinnings were carried out by Manitoba Forestry Branch with assistance from Forestry Canada in 1991 and 1992. Genetic assessment of these seed orchards, or of their constituent trees, can be initiated when the mass selection process is completed in 1997-98.

Controlled pollination was successfully completed in 1992 on eastern breeding district family test trees in four plantations, selected by analysis of 15-year measurements. Careful planning and allocation of a sizable combined federal-provincial force overcame the expected and unexpected obstacles. Twenty-eight unrelated pairs were mated. Progenies from these matings will expand the seed orchard planted in 1988 with progenies of family-test trees selected on 10-year measurements (Klein 1986). A report is available describing multiple-trait combined selection and mating for this seed orchard and one in Saskatchewan from western breeding district family test parents (Klein 1992).

A potted grafted seed orchard is being established for western Manitoba using parent clones selected from 10-year results in the central breeding district family test, which includes sources and test plantations in eastern Saskatchewan and western Manitoba. Sixty parent clones grafted in 1992 and 1993 will be re-assessed when 17-18 year observations are analyzed.

## APPLIED TREE IMPROVEMENT RESEARCH

Much effort in applied tree improvement research in Manitoba is directed toward facilitating use of vegetative propagation of control-pollinated seedlings to expedite delivery of genetic gain for jack pine. The difficulty of vegetative propagation of jack pine has impeded use of such a system with this species. That part of the system is being addressed by a PAIF contract with Dr. C. Davidson and Dr. R. Browne, at Agriculture Canada, Morden Research Station. In the first year of work, rooting of better than 90% has been achieved with cuttings from seedlings under 5 years of age. Current efforts are being focused on increasing the numbers of cuttings from each donor plant.

Experiments were initiated on response of jack pine flowering and seed production to nitrogen level and a variety of crown management treatments. These experiments seek to maximize efficiency of controlled pollination, as well as contributing to management of conventional open-pollinated seed orchards. Grafts of jack pine are being maintained in container culture for experimental application of treatments intended to accelerate and increase flower and seed production, for seed orchards or breeding of advanced generation progenies.

Other applied research activities include a vegetation management trial in a young black spruce (*Picea mariana* (Mill.) B.S.P.) seedling seed orchard, early testing in jack pine, and a test of realized gain in the first generation of the eastern breeding district jack pine breeding program.

# INITIATE SECOND GENERATION

Measurements and other observations in the eastern breeding district jack pine family test to 21 years from planting are currently being analyzed to select genotypes to use as parents of second generation progenies. "Best linear prediction" scoring is being applied to assess breeding value of family test trees. Traits included in the scoring are height, diameter, wood density, stem quality, and incidence of western gall rust (*Endocronartium harknessii* (J.P. Moore) Y. Hiratsuka). Some preliminary selections were grafted in February 1993 in the hope of expediting production of some second generation progenies. A final choice has not yet been made between the polycross - pair mating and blocked diallel systems of producing second generation populations.

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# WEYERHAEUSER CANADA - TREE IMPROVEMENT IN SASKATCHEWAN

## Diane Roddy & Spencer McDougald

# Weyerhaeuser Canada Ltd., Saskatchewan Timberlands Box 1720 Prince Albert, Saskatchewan S6V 5T3

Keywords: Pinus banksiana, Picea glauca, Populus tremuloides.

In a jack pine tree improvement program, a grafted first generation orchard can now meet all operational seed requirements, the establishment of cross pollinated tests of the orchard parents is nearing completion, and second generation selections will soon be made from a larger base of trees established in 10 to 13 year old open pollinated progeny tests. In a white spruce program, a grafted orchard is not yet meeting all seed requirements, breeding work for cross pollinated tests of the orchard parents is underway, and the establishment of a larger base of selections in open pollinated progeny tests will be completed this fall. Aspen tree improvement is receiving a lower priority, a new tree improvement greenhouse/headerhouse facility is nearing completion, and two research projects with other agencies are underway.

## JACK PINE PROGRAM

A grafted first generation seed orchard containing 31 clones now produces enough seed to meet all reforestation requirements, although unimproved seed is still used where the loss of valuable seed and seedling quality are concerns.

A series of open pollinated progeny tests for 221 selections will be remeasured over two years, in the fall of 1993 and 1994. The trees will range in age from 10 to 13 years, and the data will be analyzed in one group by staff from Weyerhaeuser's Southern US Tree Improvement Program. Although it was reported earlier that there <u>was</u> significant genotype by environment interaction in these tests based on some initial 10 year measurements, there was an error made in the data analysis, and the interaction is not expected to be found in the new data results. Second generation selections for a breeding program will be made from the tests using an index which weights individual and family performance. A light roguing of the production orchard will also be done based on the resulting data, with a heavier roguing to follow based on 15 year data.

A series of cross pollinated progeny tests for the orchard trees is being established over four years. The last tests were sown in the greenhouse this season, and will be outplanted in 1994.

### WHITE SPRUCE PROGRAM

A grafted first generation seed orchard containing 40 clones is now producing some seed for operational planting, but frost severely damaged the 1992 flower crop, and moderately damaged the 1993 flower crop. Seed from a seed production area is used, when available, to supplement orchard production, and unimproved seed is also sown to meet our planting needs.

Breeding work for a series of cross pollinated progeny tests of the orchard parents is underway, and the first tests could be sown in the greenhouse as early as 1994.

The establishment of a series of open pollinated progeny tests for 236 selections is nearing completion, and the last set of tests will be outplanted in the fall of 1993.

# ASPEN PROGRAM

As members of the Aspen/Larch Co-operative at the North Central Experiment Station, University of Minnesota, we are selecting aspen plus trees, and collecting seed from check trees in preparation for an aspen provenance trial.

# OTHER PROJECTS AND DEVELOPMENTS

A new tree improvement greenhouse facility should be completed by the fall of 1993, and the first crop grown in 1994. An attached headerhouse will provide a place for filling and seeding containers before they go into the greenhouse, as well as a facility for processing seed and pollen, and storing supplies and chemicals.

There are currently two co-operative research projects underway. Techniques for rooting cuttings of jack pine are being refined by staff at Agriculture Canada's Research Station in Morden, Manitoba, and we are growing white spruce that have been treated with microbial seed inoculants by Esso Ag. Biologicals in Saskatoon, Saskatchewan.

# **PFRA SHELTERBELT CENTRE - TREE IMPROVEMENT SUMMARY**

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Keywords: Shelterbelt, windbreak, progeny test, seed orchard, provenance test.

The primary objective of the tree improvement programme at the PFRA Shelterbelt Centre is to develop genetically superior trees and shrubs for shelterbelt planting in the prairie provinces of western Canada. From 1991 to 1993 the programme has focused on poplar (*Populus* spp.), Scots pine (*Pinus sylvestris* L.), Siberian larch (*Larix sibirica* Ledeb.), green ash (*Fraxinus pennsylvanica* Marsh. var. subintegerrima (Vahl.) Fern.), and bur oak (*Quercus* macrocarpa Michx.).

#### SCOTS PINE

A major collection of Mongolian Scots pine (*Pinus sylvestris* L. var. mongolica) from its native range in Northeast China (Heilongjiang province and Inner Mongolia) was completed. This collection, in co-operation with the Northeast Forestry University, Harbin, China includes 34 individual trees from 24 locations. The seed is being propagated for test planting in 1995. A thirteen-year-old provenance of 60 Scots pine origins was evaluated in 1992. Trees from the Ukraine and West central Russia and southern Siberia performed best. The slowest growing trees originated from north of 58° latitudinal. Two half-sib progeny tests were established, one each in Saskatchewan and Manitoba. All Scots pine seed grown for shelterbelt plantings in 1993 originates from a one hectare seedling seed orchard.

## SIBERIAN LARCH

A Siberian larch progeny test was planted at three Saskatchewan and Manitoba locations in 1993. Seed for the tests was obtained from Forestry Canada - Petawawa National Forestry Institute.

Roguing of a 1987 seedling seed orchard is under way. Phenotypically superior trees were selected and vegetatively propagated. These trees will be planted in the seedling seed orchard.

## **GREEN ASH**

Detailed studies investigating the water relations of green ash are under way. Screening methods to identify drought resistant sources of green ash are being developed. To date 15 sources have been screened. Three sources have been identified as having superior drought resistance. A three hectare first generation clonal seed orchard has been completed. The orchard will be coming into production within the next two to three years. Progeny tests initiated in 1989 will be evaluated in 1993.

## POPLAR

A clonal field test of 100 *Populus* hybrids developed at the Shelterbelt Centre were evaluated after five growing seasons. Several hybrids with superior growth, hardiness and insect and disease resistance were identified. The best performing clones will be used to make up a clonal package, the objective being to increase the diversity of poplar shelterbelts which will help to maintain healthy populations that have the genetics to accommodate the movement of a disease or virus through the shelterbelt.

In 1992, a new male poplar P. x 'Manitou' selected from an open pollinated population of P. x 'Walker' was introduced for planting in prairie shelterbelts. 'Manitou' is a vigorous tree resistant to the major insects and diseases found on the prairies.

# **BUR OAK**

In 1993, range-wide provenance tests of bur oak were planted at two sites in Saskatchewan and one in Manitoba. Sixty families are included in the test. This is a co-operative project with the USDA, Forest Service, USDA Agriculture Research Service, and USDA, Soil Conversation Service. Objectives of the project are to: 1) determine the nature and extent of genetic variation present among open-pollinated progenies of bur oak from selected sources in the Great Plains; 2) identify best adapted sources of bur oak for planting in the Great Plains; and 3) provide germplasm that can be used for selection and trait improvement as well as advanced generation breeding.

# CONSERVATION PLANT MATERIAL DIRECTORY

The Shelterbelt Centre is co-ordinating the compilation of a Conservation Woody Plant Material Directory. The world wide directory will provide information on institutions and agencies involved in development of conservation woody plant material, as well as lead the user to sources of conservation woody plants and facilitate their exchange or access to information about them. It is hoped the directory will be completed by 1994.

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# GENETICS AND TREE IMPROVEMENT RESEARCH AT THE UNIVERSITY OF ALBERTA

# Bruce P. Dancik, Om P. Rajora, and Associates

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Keywords: Molecular, population, and evolutionary genetics; molecular markers for disease resistance; speciation and biosystematics; chloroplast, mitochondrial and nuclear DNA; isozymes; tree breeding; tissue culture micropropagation.

Om P. Rajora joined the Department of Forest Science, University of Alberta on June 1, 1992 as a Assistant Research Professor. During the past two years, our research activities continued in the areas of molecular, population, and evolutionary genetics, speciation, biosystematics, biotechnology and tree improvement. In 1992 and 1993, we also initiated and conducted research projects on molecular genetics of disease resistance in jack pine (*Pinus banksiana* Lamb.) and aspen (*Populus tremuloides* Michx.), and on the impact of silvicultural practices on maintaining genetic diversity in white spruce (*Picea glauca* (Moench) Voss).

Since the last report one Ph.D. student completed his degree program in forest genetics.

# MOLECULAR, POPULATION AND EVOLUTIONARY GENETICS, SPECIATION, AND BIOSYSTEMATICS

## Isozyme Genetics

Om Rajora and Bruce Dancik completed and published studies on genetic characterization and relationships of *Populus* species, interspecific hybrids and their clones, and on allozyme variation and inheritance in leaves of *Populus* species and hybrids in comparison to those in root tips. Data analyses and manuscript preparation are in progress on our (Om Rajora and Bruce Dancik) study completed earlier on population genetic structure, variation and evolution of *Picea engelmannii* Parry, *P. glauca* and their putative natural hybrid complex in Alberta. Isozyme studies of *Populus* species with Dr. Burton V. Barnes, University of Michigan, continue.

## Molecular Genetics

John Barrett completed his Ph.D. studies on "Molecular characterization of a single *cab* gene in lodgepole pine" under the joint supervision of Bruce Dancik and Curtis Strobeck (Department of Zoology). Om Rajora completed and partially published his studies on chloroplast and mitochondrial DNA variation and inheritance and genetic relationships of *Populus* species. Dr. Rajora continues to examine variation and inheritance of nuclear genes involved in major metabolic pathways, in *Populus* species and hybrids. Technologist Mary Aleksiuk, Bruce Dancik, and Francis Yeh completed studies on extraction and variation of purified chloroplast DNA in 10 conifer species. Former Research Associate Keith Eggar accepted a position as Assistant Professor at Memorial University.

# Variation and Differentiation

Barbara Thomas (B.Sc., M.Sc., University of British Columbia) began a Ph.D. program under the supervision of Ellen Macdonald and Bruce Dancik. Barb is studying variation and differentiation of trembling aspen utilizing material collected from several Alberta populations grown under controlled environment (phytotron) conditions.

Rob Wright is nearing completion of a Ph.D. study of jack pine under the supervision of Ross Wein and Bruce Dancik. Christine Hansen and Leonard Barnhardt began M.Sc. programs under the supervision of Bruce Dancik.

## MOLECULAR GENETICS OF DISEASE RESISTANCE, AND GENETIC FINGERPRINTING

Om Rajora and Bruce Dancik have initiated and continue to conduct the following two research projects on determining the molecular genetic basis of disease resistance in two tree species: (1) *Populus tremuloides* - developing technology, probes and guidelines for DNA fingerprinting of individual clones that are resistant and susceptible to decay and stain; (2) *Pinus banksiana* - identifying molecular genetic markers for differentiating genotypes and families that are resistant and susceptible to western gall rust (WGR) caused by *Endocronartium harknessii* (J.P. Moore) Y. Hiratsuka, and developing guidelines for molecular marker-assisted early testing and selection of jack pine for WGR resistance.

# IMPACT OF SILVICULTURAL PRACTICES ON MAINTAINING GENETIC DIVERSITY

Om Rajora and Bruce Dancik have undertaken studies on examining the impact of silvicultural practices on maintaining genetic diversity in the regenerated white spruce (*P. glauca*) forest by using molecular genetic techniques.

#### TREE IMPROVEMENT

Dr. Sally John completed her assignment as tree improvement specialist/ research assistant professor and joined her spouse, Dr. Jean Brouard, on an assignment in Zimbabwe.

Om Rajora, while working as a consulting geneticist for the Alberta Forest Service, developed a long-term program plan for genetic improvement of trembling aspen and other poplar hardwoods in Alberta. Also, he made seed source collections of plains cottonwood (*Populus deltoides* Marsh. var. occidentalis Rydbg.). Ruichuan Zhang began a Ph.D. program under the supervision of Bruce Dancik and Dick Pharis (U. of Calgary) on very early evaluation of seedling performance.

## Breeding

Om Rajora has made reciprocal controlled crosses between WGR-resistant and WGR-susceptible jack pine genotypes for western (Saskatchewan) and eastern (Manitoba) breeding districts.

# TISSUE CULTURE MICROPROPAGATION

# Om Rajora streamlined techniques for operational tissue culture micropropagation of *P. tremuloides*, for the Alberta Forest Service.

#### ACKNOWLEDGEMENTS

Support for the various studies above was provided by NSERC research grants and postgraduate scholarships, Canada-Alberta, Canada-Saskatchewan, and Canada-Manitoba Partnership Agreements in Forestry, and Forestry Canada

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# GENETICS AND TREE IMPROVEMENT PROGRAMME ALBERTA LAND AND FOREST SERVICES

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Keywords: Tree breeding, genetic improvement, provenance studies, species testing, seed orchards, progeny testing, white spruce, lodgepole pine, aspen.

This report summarizes the progress of the Alberta Land and Forest Services (L.F.S.) genetics and tree improvement programme for the period 1991-1993.

# PROGRAMME DEVELOPMENT

Due to Departmental restructuring, Alberta Forest Service and Alberta Lands Division were merged to become Land and Forest Services. Various projects were reviewed in view of tightening budgets and it was decided to discontinue the recently started hardwoods genetics and improvement project. The Genetics and Tree Improvement Centre facilities at Pine Ridge Forest Nursery were renovated and expanded to provide additional seed storage, laboratory and office space. The programme emphasis during the report period continued on expanding existing seed orchards to meet substantially increased seed production requirements and to achieve larger involvement of industry in ongoing tree improvement projects.

## GENETIC IMPROVEMENT

# Assembly of Breeding Stock

Field selection of superior trees to provide material for L.F.S. as well as L.F.S./Industry cooperative projects continued. Selections for L.F.S. projects consisted of a total of 55 lodgepole pine (*Pinus contorta var latifolia*), tamarack (*Larix laricina*), black spruce (*Picea mariana*), Douglas-fir (*Pseudotsuga menziesii var glauca*) and Scots pine (*Pinus sylvestris*) trees. As part of the L.F.S./Industry cooperative projects, a total of 76 white spruce (*Picea glauca*), black spruce and aspen (*Populus tremuloides*) trees were selected. Industry participants in these cooperative projects were Alberta Pacific Forest Industries, Blue Ridge Lumber (1981) Ltd., Canadian Forest Products Ltd., Weyerhaeuser Canada Ltd., Daishowa-Marubeni International Ltd. and Slave Lake Pulp Corporation.

## Genetic Testing

Stock was seeded for two series of white spruce progeny tests that will be outplanted in 1994. The trials for breeding region 'E' are comprised of 64 open-pollinated half-sib families and those for breeding region 'H' are comprised of 54 families. A total of five field trials will be planted in the two regions. Stock production is also in progress for lodgepole pine progeny trials for breeding region 'K'. Forty-three open-pollinated half-sib family seedlots collected from selected superior trees were seeded. Outplanting of the field trials is scheduled for 1995.

Early screening of lodgepole pine half-sib families for resistance to western gall rust (*Endocronartium harknessii*) was started in cooperation with Dr. Y. Hiratsuka of Northern Forestry Centre by inoculating young seedlings in a greenhouse. A set of 126 families from breeding region 'C' were screened in 1992. Another set of 101 breeding region 'B1' families is being screened in 1993. Large differences were found among families for infection by western gall rust in 1992 screening trials. Approximately three percent of the families showed zero percent infection. A portion of resistant seedlings from selected families are being outplanted at a high infection field site to confirm their field resistance to the disease.

## Seed Orchards

Regions 'E' and 'H' seed orchards started in 1988-1989 were reviewed and redesigned due to insufficient grafts in many clones needed to complete the orchards within the next two years. The new design required tree spading some established grafts into reassigned positions. Grafting from scions collected from already established ramets is being done to provide additional grafts for expanding these seed orchards in order to meet original seed requirements. Expansion orchards will be located adjacent to the existing orchards. Region 'F' Douglas-fir and western larch seed orchards were started near Brooks, Alberta. Planting of these is expected to be completed in 1994.

Stock was seeded for a lodgepole pine seedling seed orchard for breeding region 'C'. One hundred and fourteen of half-sib seedlots were selected to make up the orchard based on a 9 year field performance of the families in field trials. Outplanting of the seed orchard will take place in 1994 cooperatively with Blue Ridge Lumber. Production of grafts was also commenced for the region 'K' lodgepole pine clonal seed orchard which is expected to be outplanted in 1995. Expansion of existing black spruce and tamarack clonal seed orchards is also continuing as these species are becoming increasingly important for reforestation in Alberta.

Weyerhaeuser Canada has selected a seed orchard site in west central Alberta for establishment of a white spruce clonal seed orchard for breeding region 'J'. Development of this site is underway.

The first commercial flower crop in a small operational Siberian larch (*Larix sibirica* var. *raivola*) seedling seed orchard is being realized in 1993. Based on an ocular rating system it is estimated that a light cone crop can be harvested from the 14 year old orchard in the fall. It is estimated that between 25 and 75 cones can be harvested from 90 percent of the orchard trees. Because pollen production in the orchard was very good, seed set is also expected to be good.

A system for rating seed orchard cone crop production was developed to assist in projecting seed orchard cone production and planning cone collections. The system assesses cone production based on the quantity of cones, distribution of cones on the tree and tree size. Each tree is rated on a 0 (nil) - 6 (very heavy) scale. Information is summarized to correlate with the realized production level of various orchards.

The first commercial cone crop from three white spruce seed orchards will be collected this August. Region 'H' clonal seed orchard located at Pine Ridge has the best cone crop among the three orchards. It consists of 338 ramets. Of these 70% are bearing cones. Of the cone-bearing ramets, 50% have a cone crop rating of 3 or more indicating that the crop is fair to very heavy. A yield of approximately 2 kilograms of seed is expected.

## GENETICS AND TREE IMPROVEMENT RESEARCH

# Species Testing

Analyses of 11 year data were completed for a pine species trial containing promising red pine (*Pinus resinosa*) seed sources. The performance of the red pine compared favourably to lodgepole pine and jack pine (*Pinus banksiana*). Mean heights for the three species were 182 cm, 267 cm and 221 cm respectively. Mean survival of the red pine, however, was 53 percent. Sixty-three percent of the surviving red pine trees showed little or no winter damage. In 1992 two trials were established in central Alberta with three red pine seed sources that performed well in the original pine species trial. These seed sources originated from northwestern Ontario and northern Minnesota. Post planting assessment of the trials in spring of 1993 showed extensive winter and animal damage. The seedlings were extensively browsed by white tail deer.

Twelve year results from two U.S.S.R. Scots pine seed source trials established in central and northern Alberta corroborated results observed at 10 years of age. The best Scots pine seed lot in central Alberta and northern Alberta test sites outperformed the local lodgepole seed source by 18% and 13% respectively in height growth. Mean heights were 492 cm and 340 cm respectively for the best Scots pine seed source at central and northern Alberta sites. This compares to 404 cm and 296 cm respectively for control lodgepole pine seed sources. The top five performing Scots pine seedlots at each site originated from latitudes similar to those of the test sites. Scots pine at the central Alberta site suffered moderate porcupine damage. The animals, however, did not browse lodgepole pine. An additional U.S.S.R. Scots pine provenance/progeny test was established at Pine Ridge Forest Nursery in 1991. It was the last of three such trials to be outplanted, two having been established in 1990 at separate test sites in Alberta. Thirty-one single tree seedlots were included in each of these trials.

#### Provenance Studies

Fifteen year assessment of a Canada-range wide white spruce provenance trial established in central Alberta was completed. At age 12 measurement in 1989, the top ten fastest growing provenances originated from southern Manitoba, Ontario and Quebec. The same ten provenances were still ranked among the top ten at the 15 year assessment. The fastest growing seed source was from Angle Inlet in southern Manitoba. Mean height of this seed source was 3.32 m which was 22 percent better than the best local white spruce seed source. In light of these results, a new series of white spruce provenance trials were established in 1992 and 1993. Six large block plantings were established as part of operational reforestation projects in central Alberta. Planting stock for these plantings was grown from operational white spruce seedlots from southern Manitoba. The performance of the Manitoba seedlots will be compared to the local reforestation seedlots on these sites. Four additional trials were established in central and northern Alberta with better performing seedlots from the original Canada range-wide trial. In addition to these seedlots, new seedlots corresponding to similar geographic origins were added. Each of these trials contains 46 non-local white spruce seed sources and four local seed sources as controls.

Field assessment and data analyses continued on Alberta-wide provenance trials of white spruce, tamarack, lodgepole pine, and jack pine with the assistance of Dr. E.K. Morgenstern of the University of New Brunswick. Results from 17 different field trials are being compiled to develop a new seed zonation system to replace the present seed source movement guidelines for reforestation in Alberta.
Due to the expansion of the greenhouse complex at Pine Ridge Forest Nursery, several white spruce and lodgepole pine research seed orchards and seed production trials were relocated to a new research field using large tree spades. The material moved included seedlings and grafts that ranged in age from seven to eleven years. A study was set up to monitor the effect of relocation of the trees in research orchards on growth, flowering and seed production. At the time of the move, the orchards were eleven years old. Part of each orchard remained in the old field and was used as a control to compare performance of relocated trees. Growth performance of the lodgepole pine seed orchard in terms of height and crown width did not differ significantly between the two treatments one year after the relocation. Two years after the relocation, however, noticeable differences were observed. Mean heights and crown widths for the relocated trees were 4.3 m and 3.3 m respectively compared to 4.7 m and 3.8 m for the control trees. Diameter growth did not differ significantly between the treatments. Significant differences in cone production, seed production and seed viability were also observed. Mean number of cones per tree, seeds per cone and percentage germination for the relocated trees were 24, 6.5 and 70 percent respectively compared to 37, 19 and 94 percent for the control trees. It was clear that relocating the lodgepole pine trees adversely affected their growth and seed production characteristics.

In the case of white spruce, relocating trees did not appear to affect growth performance but significantly improved flowering and cone production two years after moving the trees. The relocated trees on an average, produced 80 female strobilii, 77 cones and 0.6 seeds per cone in comparison to 7 female strobilii, 4 cones, and 0 seeds per cone for control trees. The poor seed set was most likely due to very poor pollen production in the orchard.

In 1983 a study was started to compare the grafting success and seed production of greenhouse versus field grafting in white spruce. Grafting success was 22 percent better for the greenhouse grafts. The grafts were established in a field trial in 1985. To date there has not been any significant flowering. Growth performance in terms of height and crown development has, however, differed significantly. The field grafts performed more vigorously initially. By 9 years of age, height growth differed slightly but there were still noticeable differences in crown development. Mean height and crown width for the field grafts nine years after grafting was 100 cm and 70 cm respectively compared to 108 cm and 55 cm for the greenhouse grafts. The field grafts generally have a denser, branchier crown and may be expected to produce better cone crops.

In 1992 work commenced to develop an ocular flower/cone production rating systems for seed orchards in Alberta. Two years of data have been collected for white spruce and lodgepole pine seedlings and grafts. The data is being analyzed to develop models for predicting cone production in commercial seed orchards.

#### PLANT PROPAGATION, WOOD AND SEED TECHNOLOGY

#### Plant Propagation

During the report period stock production consisted of 62,266 seedlings, 4248 grafts, 5183 potted rootstock trees, and 1680 stecklings. The grafting program consisted mainly of white spruce and lodgepole pine with a total of 2990 grafts and 806 grafts respectively being completed for these species. A 65 percent success rate was achieved with white spruce, and a 90 percent success rate with lodgepole pine. Other species grafted were Douglas-fir, western larch, tamarack, black spruce, blue spruce (*Picea pungens*), Scots pine and jack pine.

Clonal propagation of aspen was commenced in 1991 in support of the hardwoods genetic improvement program. A total of 859 propagules were produced by rooting root suckers. In addition, 728 tower poplar propagules were produced using the same technique to provide planting stock for shelterbelts to be established on genetics experimental sites.

#### Wood Technology

Relative density and fibre length measurements of wood samples collected from selected parent trees continued. Species tested were Douglas-fir, lodgepole pine, black spruce and trembling aspen (*Populus tremuloides*). A total of 93 relative density and 113 fibre length assessments were completed. A technical review of the wood technology program was completed with the assistance of Dr. Lars Bach from the Alberta Research Council. A comprehensive report on the work completed to date and the results, is in preparation.

#### Seed Technology

A total of 122 seedlots were added to the genetics seed bank. Of these seedlots, 39 percent were white spruce, 58 percent were lodgepole pine, and 8 percent were from miscellaneous species. The seedbank presently contains 3471 seedlots.

Quality of seedbank seedlots continues to be monitored annually by testing a set of reference seedlots representing about 2 percent of seedbank entries. New seedlots are added to the testing program every few years to replace depleted seedlots and sample new seedbank entries. In order to lessen the work load, it was decided that in future, reference seedlot testing for coniferous seedlots will be carried out every second year. Deciduous reference seedlots, however, will continue to be tested annually because of their faster decline in viability. Mean germination of lodgepole pine seedlots in the seedbank has declined from 87 percent in 1981 to 77 percent in 1992. Germination testing of aspen, plains cottonwood (*Populus deltoides var occidentalis*), and balsam poplar (*Populus balsamifera*) showed germination decline of 5 to 10 percent in one year.

The ultra-low temperature seed storage study started cooperatively with the National Tree Seed Centre at Petawawa, is now in its fifth year. Average germination for white spruce after 5 years storage at -80°C was found to be 79 percent compared to a baseline mean germination of 82 percent at commencement. For lodgepole pine, mean germination at -80°C storage was 88 percent compared to an average baseline germination of 84 percent at -20°C storage. The study will be continued on white spruce for one more year although from results to date it is evident that -80°C storage temperature does not offer any advantage over conventional -20°C storage.

## EARLY EVALUATION OF INHERENTLY SUPERIOR GROWTH IN FOREST TREES & ITS PHYSIOLOGICAL BASIS

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Over the past several years we have used "retrospective testing" to evaluate methods of assessment of early family and genotype performance (e.g. ages 1-24 mos). This has involved growing progeny under near-optimal temperature, nutrients, water, and light in glasshouses, and assessing a variety of growth parameters, and as well endogenous hormone levels. These early phenotype characteristics can then be used to assess phenotypic and genetic correlation with performance in field progeny trials. The work has most recently been accomplished for each of (a) F1 half-sib Alta. lodgepole pine (*Pinus contorta* Dougl.) crosses and (b) various Radiata pine (*Pinus radiata* D. Don) crosses, including full-sib and F2 half-sib families.

In addition to determining whether early family selections might be practical, we are also interested in investigating the physiological (functional) basis of inherently rapid early growth in conifers and its relation to the gibberellin (GA) class of hormones. This is not only of basic importance in tree physiology, but may also be important in improving the magnitude of phenotypic correlations between early test results and field performance. Such correlations can, in theory, be increased by increasing test precision and/or the use of highly heritable seedling traits with high and consistent genetic correlations with field performances.

Results to date include the development of necessary genetic and statistical equations to complete the theoretical investigations of genetic gain from indirect early seedling selection under near-optimal glasshouse conditions for field tree performance under five early selection approaches:

- (1) indirect early selection for single seedling trait,
- (2) indirect early selection for single and multiple seedling traits,
- (3). multi-generation indirect early selection for single and multiple seedling traits,
- (4) index selection combining both early seedling and tree field traits
- (5) two stage selection, first on early seedling traits, then by tree field traits.

We have now completed a retrospective study on the genetic relationships between 28 seedling traits for 2 growth periods [equiv. of years 1 and 2] in the glasshouse and 9-year tree field tree heights (ht) for 120 half-sib OP families of lodgepole pine. Year 15 ht will be known in 10/93. Genetic differentiation among families for all seedling traits was well developed during each growth period, suggesting that an "inherent ability to grow rapidly" can be detected at early ages. This supports results and views put forward by Bongarten and Hanover (1985) and Pharis et al. (1990) that under optimal growth conditions, trees express their inherent genetic variation earlier! Early genetic differentiation of families is a necessary condition for early selection in lodgepole pine. Of the 28 seedling traits, 24 (86%) exhibited significant genetic correlations ( $P \le 0.0312$  to 0.0001) with 9-year field tree ht. This high level of genetic correlation between glasshouse and field performances was in part the result of the large number of families and the use of an unique experimental layout (minimizing seedling position effects on benches), and adjustments of individual seed weights (wt) in early seedling performances. Our high estimates of individual heritabilities and small S.E. (0.543±0.103 to 0.949±0.137), relative to other reports for lodgepole pine would result in higher early selection efficiency (Kang, 1985).

Substituting genetic parameters derived from glasshouse and field studies into 5 sets of early selection equations, the following are the 5 most significant results for lodgepole:

(1) Shoot-root biomass ratio, seedling height and seedling diameter were the 3 early traits most efficient for indirect selection of 9-year tree hts.

(2) Indirect selection for 9-year tree ht based on two early seedling traits was, on average, 34% more efficient than indirect selection based on a single early seedling trait.

(3) Two generations of early selection based on seedling ht after 2 growth periods in the glasshouse would result in expected indirect genetic gain in 9-year tree ht identical to that obtained from direct selection for 9-year tree hts.

(4) Index selection based on 1 early seedling trait and 9-year tree ht was, on average, 40% more efficient than selection based on 9-year ht alone.

(5) Under 2-stage selection, 20% of the families in the glasshouse could have been culled on the basis of basal diameter at lifting with no negative impact on the genetic gain in 9-year ht (e.g. gain expected if all families had been field tested).

These theoretical and experimental results of early selection in lodgepole pine reinforce the view that under carefully designed conditions (e.g. large number of families, effective experimental layout, near-optimal growing conditions, adjustment for seed wt, proper selection of early seedling traits, and proper use of appropriate genetic and statistical equations) very early selection is an effective tool for tree improvement in general, and lodgepole pine in particular.

Within the last 12 months new genetic information for field performance (Best Linear Prediction [BLP] developed by Dr. Tim White [U. Florida] for the Southern Tree Breeding Association [Australia] has become available [via Dr. David Boomsma] on >100 families of Radiata pine across Australia, including the 9 full sib and 5 half-sib families used in our earlier Phytotron early growth studies. With the BLP method the Breeding Values for Diameter at Breast Height (field) can be predicted, and genetic gain quantified. We have thus replotted our earlier experimental results for several of the best early growth parameters (stem dw and seedling ht), relative to Percentage Genetic Gain. Because the earlier trial used applied gibberellin A4/7 [GA<sub>4/7</sub>] as a research tool to promote shoot growth, we have been able to compare the Percentage Genetic Gain for each of the families. An example for height growth at age 6 months for these families was given in Table 6 in Pharis et al. (1991).

Now, using the BLP method, we can show our early (ages 3-4 mos) measurements of phytotron-grown seedling stem volume or stem dw (the best early growth parameter), or even height to be significantly related to Tree Breeding Value (% Gain) for Diameter at Breast Height in the field across Australia. For example, for full-sib families, the correlation of family means (r) was 0.77 for age 4 month stem dry wt and 0.69 for age 6 month stem dry wt. Interestingly, for older (thereby larger) plants which were becoming pot-bound, a gibberellin A<sub>4/7</sub> soil drench increased the significance of the relationship of early growth measurements to Breeding Value % Gain in the field (e.g. for full-sib families, age 6 mo. stem dry wt,  $r = 0.69 \text{ w/o GA}_{4/7}$ , but r increased to 0.81 when GA<sub>4/7</sub> was applied).

#### Relationship Between Levels of Endogenous Gibberellins and Seedling Growth

Because of the "history" of applied  $GA_{4/7}$  stimulating both elongation growth (ht) and wood growth of conifer seedlings, we proposed to explore this relationship -- namely is endogenous GA level related to rapidity of growth of seedlings [genotypes] within a family, and to inherently rapid or slow growth between families. The latter was answered in a positive manner for GA4+GA7+GA9 levels in 8 half-sib OP families of Idaho-origin lodgepole pine [supplied by Dr. G. Rehfeldt] (Zhang, 1990) and for GA9 levels in needles of Radiata pine (see Table 2 in Pharis et al., 1991).

We have also been exploring this question further with tissue harvested from an F2 half-sib, 16 family "retrospective trial" of Radiata pine (seed provided by Dr. R. Burdon, NZ For Res Instit., Rotorua, seedlings grown at U Alta).

Preliminary results show that endogenous GA9 (a key intermediate in the non-hydroxylation GA biosynthesis pathway) is significantly and positively related to early (age 3-6 months) growth rank of seedlings (genotypes) within each of the 16 families, needles of fast-growing seedlings having significantly more GA9, and needles of slow-growing seedlings significantly less.

Unfortunately, unlike the earlier full-sib 5 family comparison for Radiata pine (Pharis et al., 1991), and the half-sib OP 8 family comparison for Idaho origin lodgepole pine (Zhang, 1990), the relationship of GA9 levels to family growth rank for the F2 Radiata pine half-sibs is complex. For example, among the 16 families, there are ca. 2 slow growers with very high GA9, and 3 fast growers with low to very low GA9. Thus, the earlier pattern [see Zhang, 1990 and Pharis et al., 1992], where needle GA9 was significantly related to both early glasshouse and later field progeny trial rank fits for only 10 to 12 of the 16 families from our more recent F2 half-sib OP early progeny trial (Zhang, Pharis, Dancik, unpubl.). It is thus possible that what we are seeing in these F2 crosses is segregation among families for genetic blocks in GA biosynthesis [or very rapid GA biosynthesis], <u>even though</u> within any one family, rapid growth and high GA9 are still positively and significantly correlated. This aspect will be explored further over the next several years, not only with Radiata and lodgepole pines, but also with other commercially important conifer species.

#### ACKNOWLEDGEMENTS

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## FOREST GENETICS ACTIVITIES AT THE UNIVERSITY OF BRITISH COLUMBIA

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Keywords: Population genetics, genetic diversity, genome mapping, molecular genetics.

Forest genetics at the University of British Columbia was reinstated to a place of importance when Gene Namkoong joined the Faculty of Forestry in October, 1992, as Head of the Forest Sciences Department. Dr. Namkoong's contributions to population genetics, breeding of forest trees and germplasm conservation during his career with the U.S.D.A. Forest Service are well known to members of the Canadian Tree Improvement Association. Dr. Namkoong's expertise brings many opportunities for expanding interactions with the B.C. Ministry of Forests tree improvement programs. He will also play a key role in exciting developments at UBC in the area of biodiversity, including the recent successful establishment of the Centre for Applied Conservation Biology in the Faculty of Forestry and the emerging Biodiversity Research Centre in the Faculty of Science. Dr. Gene Namkoong continues to work with and consult for colleagues in the USDA Forest Service. He also remains very busy in international functions. Dr. Namkoong's research at UBC will commence shortly when renovation of the forest genetics laboratory is complete. Please refer to the list of publications for an update on Dr. Namkoong's recently completed projects. The remainder of this report will deal with research activities underway in Carlson's group.

### SEED ORCHARD DNA FINGERPRINTING

This project is being conducted in collaboration with Joe Webber and Mike Stoehr of the B.C. Ministry of Forests and Ben Sutton of B.C. Research. We are developing DNA fingerprints to identify all 95 trees in an operational interior spruce seed production orchard using RAPD (Random Amplified Polymorphic DNA or single-primer DNA amplification) markers. These DNA fingerprints will be used for paternity analysis, to monitor parental balance in supplemental pollinations, to monitor pollen contamination, and to certify seed lots. Fingerprinting the clones was accomplished by post-doctoral fellows Yong-Pyo Hong and Robin Davidson. This study has now expanded to include a study on effective population size, using the DNA fingerprints, that will be conducted by a new post-doctoral fellow Filipos Avaradopolis, in collaboration with Gene Namkoong.

#### GENOME MAPPING

We are constructing genetic linkage maps using RFLP (Restriction Fragment Length Polymorphism) and RAPD markers. We plan to use these maps to identify markers linked to traits important in tree improvement, for use by breeders in early selection. We pioneered the approach of constructing genetic linkage maps for single-trees using haploid megagametophyte DNA. To increase access to the RAPD technology, we have distributed, at cost, 700 RAPD primers to over 200 research groups around the world. Dr. John Carlson was also appointed to the scientific advisory committee for the U.S.D.A Forest Service's tree genome mapping program. The first review of the project was held at the U.S. Forest Service's Pacific Southwest Research Station at Albany, California, on June 25, 1993.

#### White spruce genetic linkage maps

Our B.C. Science Council funded white spruce mapping study is drawing to a close. Post-doctoral fellows Yong-Pyo Hong and Robin Davidson and PhD student Jeff Glaubitz constructed genetic linkage maps with RAPD markers for four parent trees in the interior spruce tree improvement program in collaboration with Gyula Kiss at the B.C. Ministry of Forests Kalamalka Research Station. With these maps we are searching for DNA markers linked to weevil resistance.

## Douglas-fir genetic linkage maps

We have initiated an NSERC strategic research project to construct genetic linkage maps for Douglas-fir. An undergraduate honours student, Jason Broome will soon complete two single-tree maps with RAPD markers for parent trees from Jack Woods' B.C. Ministry of Forests coastal Douglas-fir tree improvement program. When a new post-doctoral fellow Xiaming Wu joins the group in October, the next phase of the project will begin with construction of a map for an F<sub>1</sub> progeny population from the two mapped parent trees. The F<sub>1</sub> map will be used to identify QTLs for wood quality and other segregating traits that Woods has documented phenotypically.

#### POPULATION GENETICS

## Analysis of Genetic Diversity in Douglas-fir

Bundit Ponoy, a PhD student from Thailand supported by CIDA and the Canada-ASEAN Tree Seed Project, has completed a study on genetic diversity in Douglas-fir in British Columbia using DNA markers, with the assistance of post-doctoral fellow Yong-Pyo Hong. Using RFLP analysis of the two organelle genomes and RAPD analysis of the nuclear genome, we compared diversity within and between 18 populations of Douglas-fir. The pattern of genetic variation in Douglas-fir observed in this study was the typical high variation within population and low variation among populations. As a trial study, only a minimum sample size was used, nevertheless it provided a clear demonstration that differences in the degree and distribution of diversity and apparent modes of evolution may be obtained from the three genomes.

#### Quantitative genetics of IUFRO Douglas-fir Progeny Trial

Bundit Ponoy, co-supervised by professor emeritus Oscar Sziklai, also evaluated quantitative traits in 100 families of 17-year old coastal Douglas-fir progenies in progeny tests at three locations (Caycuse, Courtenay, and Gold River) on Vancouver Island. The progeny tests used a systematic single tree plot-design and included four family types --full-sib, half-sib from clone bank (C), half-sib from original plus tree (P) and control. Genetic gains were estimated for height, diameter, and volume at different selection intensities. About 18-30% gain for volume was obtained when selection was made at 10% of the best parents in each family type. Age-age correlations showed that field performance at years 5-6 could predict height growth at 17 years.

#### Analysis of DNA Diversity in Western Red Cedar

A PhD student, Jeff Glaubitz, is assessing genetic variation in western red cedar in the chloroplast and mitochondrial genomes by DNA amplification and DNA sequencing and in the nuclear genome using RAPD and RFLP markers. We hope to assemble an extensive RFLP and RAPD genetic diversity data base from sampling of populations in coastal B.C., interior of B.C., the Charlotte Islands, Utah, Washington, Oregon and northern Călifornia to encompass the entire range of western red cedar. This will serve as a valid comparison of isozyme and DNA markers in population genetics. This study is a collaboration with Y. El-Kassaby, of Canadian Pacific Forest Products and is supported by the Forestry Canada, National Forest Genetic Resources Centre.

#### Population Genetics of the Eastern white pine weevil, Pissodes strobi

In collaboration with John McLean, forest entomologist at UBC, and with support from the Green Plan, MSc student Kornelia Lewis is using RAPD markers to assess the extent of genetic variability, population subdivision, and host specificity in weevil populations in British Columbia. We hope that this information will assist both pest management and breeding for weevil resistant spruce.

#### Population Genetics of Endocronartium

We are also using RAPD markers and RFLPs to assess the extent of genetic variability, the existence of races, the mode of inheritance (life cycle), and the spread of infection in the western gall fungus *Endocronartium harknessii*. Forest pathology PhD student LiJuan Sun (Bart VanderKamp, supervisor) is sampling spores from individual galls from across the province.

### MOLECULAR CYTOGENETICS

Garth Brown, a forest genetics PhD student, has developed a fluorescence in situ hybridization (FISH) technique for physically mapping DNA on conifer root-tip chromosomes. With FISH, we have localized the major ribosomal DNA (rDNA) loci to 14 chromosome sites in white spruce - one rDNA site on each of 7 separate chromosome pairs (2n = 24). A preliminary karyotype for spruce was constructed based on the rDNA hybridization sites and chromosome morphology. This karyotype is sufficient for the unambiguous identification of all 24 spruce chromosomes. We are now working on a detailed molecular karyotype for white spruce by mapping newly characterized repetitive DNA elements, using FISH. We are interested in using FISH to study the organization of the conifer nucleus and for correlating genetic linkage maps with metaphase chromosomes.

#### LIGNIN MOLECULAR BIOLOGY

Carlson and PhD student P. Dharmawardhana, are collaborating with Brian Ellis in the Plant Science Department and Carl Douglas in Botany on an NSERC Strategic Research Grant on "Biotechnological modification of lignin in poplar." We hope to learn enough about the genes controlling lignin synthesis and biochemical process of lignification during wood development to devise a strategy for breeding trees with decreased lignin or improved wood density.

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## SEED CERTIFICATION AND BIODIVERSITY RESEARCH PACIFIC FORESTRY CENTRE

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Keywords: Official certification and testing, biodiversity, genetic diversity, *ex-situ* conservation, simulated aging, protein matrix, thermal gradient system.

The seed testing and source certification services, and the forest biodiversity research program at the Pacific Forestry Centre are outlined.

### OFFICIAL CERTIFICATION AND TESTING OF TREE SEEDS

## F.T. Portlock

As the Certifying Authority under the OECD scheme for the Pacific and Yukon Region, 65 certificates of provenance for 216 kg of seeds in the source-identified category were issued in 1991. The majority of the source-identified seed certified was *Picea sitchensis* (Bong.) Carr. (111 kg). Small amounts of *Pinus contorta* Dougl. (54 kg), *Alnus rubra* Bong., *Thuja plicata* Donn, and *Pinus ponderosa* Laws. were also certified. Under the ISTA seed testing rules, 26 certificates of seed quality were issued in 1991; these represented 225 kg of seeds from 9 species, with *Picea sitchensis* accounting for 50% of the total weight.

In 1992, 1255 kg of source-identified category seeds were certified. In addition, 1 kg of *Picea sitchensis* seed was certified under the untested seed orchard category. The sourceidentified category seed included *Picea sitchensis* (958 kg), *Abies lasiocarpa* (Hook.) Nutt. (23 kg), *Picea glauca* (Moench) Voss (14 kg), *Pinus contorta* (16 kg), *Thuja plicata* (26 kg), *Tsuga heterophylla* (Raf.) Sarg. (7 kg), and *Alnus rubra* (3 kg). In addition 208 kg of *Pseudotsuga menziesii* (Mirb.) Franco were also certified. Concurrently, 91 ISTA certificates of seed quality, for 1507 kg of seeds of 10 species were issued; *Pinus contorta*, *Abies grandis* (Dougl.) Lindl., *Pinus ponderosa*, *Picea sitchensis* and *Pseudotsuga menziesii* were the major species.

The registration of seed orchards in the untested seed orchard category of OECD was continued with two additional orchards registered in 1991, bringing the total in British Columbia to 12.

## **BIODIVERSITY RESEARCH**

#### D.G.W. Edwards

Following termination (1991) of the seed research program, a program of forest biodiversity research was initiated, the main focus being genetic diversity and its conservation. Two collaborative projects are underway with Dr. M. Meagher (PFC) and Dr. Y.A. El-Kassaby (Canadian Pacific Forest Products). Specific portions of the work are being assisted by Dr. C.L. Leadem and Dr. S. L'Hirondelle (B.C. Ministry of Forests), and Dr. J. Maze (Univ. B.C.).

The first project, "*Ex-situ* conservation of forest biodiversity in B.C.", is funded under FRDA II, and is aimed at studying what happens to the genetic base of a tree seed crop when the seeds are stored. By means of simulated long-term (accelerated) aging, this project challenges the assumption that the genetic integrity of stored seeds does not change over time. All the coniferous species under domestication in this region, plus a minor species, mountain hemlock (*Tsuga mertensiana* (Bong.) Carr., and a representative broad-leaved species, red alder (*Alnus rubra*) will be investigated.

The objectives are to determine if commonly-used forest tree-seed storage practices result in a reduction in genetic diversity; to relate any reductions to specific genotypes; to identify specific changes (gene markers) in the protein matrix with changes during storage; and to propose alternative approaches to current seed storage procedures to circumvent any changes in the genetic composition of stored seedlots.

Genetic differences in germination parameters and dormancy have been found in *Pseudotsuga menziesii* (El-Kassaby et al. 1992), *Picea sitchensis* (Chaisurisri et al. 1992) and *Thuja plicata* (El-Kassaby et al. 1993). Evidence of strong genetic (maternal) control ( $h_b^2 > 0.5$ ) has been found for *Pseudotsuga menziesii*, *Picea sitchensis*, *Thuja plicata* (El-Kassaby et al. 1993) and *Tsuga mertensiana* (in preparation). The unique structure of coniferous seeds dictates this strong maternal effect (4:1 maternal:paternal) (El-Kassaby et al. 1992). A novel interpretation of germination parameters has also been reported (Thompson and El-Kassaby 1993).

Since strong genetic control over germinability/dormancy has been established, similar differences in rate of seed deterioration can be expected. Genotype-specific differences in rate of deterioration under simulated aging have been documented for *Picea sitchensis* (Chaisurisri et al. 1993) and in *Pseudotsuga menziesii* (in preparation); work has begun on *Tsuga heterophylla*, *T. mertensiana* and *Pinus contorta*. These results indicate that the genetic makeup of a seedlot before/after simulated aging will be different.

Biochemical analyses are underway to establish the protein matrix for fresh, untreated seeds of *Pseudotsuga menziesii* and *Pinus contorta*, and to determine changes in the matrix caused by simulated aging.

The second project, "Genetic diversity in mountain hemlock", is funded under Green Plan. With current timber harvesting extending further into the sub-alpine elevations, it is important to obtain and understand the traits and characteristics of high-montane species, their range, diversity, regeneration potential and reproductive success, and to relate this information to the harvesting method that best maintains the species.

The objectives are to assess the state of existing knowledge on *Tsuga mertensiana* and to produce an annotated bibliography; to examine reproductive success and germination ecology; to determine adaptive and quantitative seedling attributes which will be analysed for intra- and inter-population variation in morphology and field performance; to determine the

mating system; to establish frost-hardiness patterns/environmental-adaptation patterns with a view to preparing seed-transfer guidelines; to determine genic variation; to analyse morphological variations in foliage, buds, cones and seeds; and to determine genetic relations to other species.

A literature search has located about 5 dozen articles, but only scattered information on reproductive morphology, reproductive method, hybridization and genetic variation in *Tsuga mertensiana* has been found; the bibliography is to be published in 1993.

Using a thermogradient system (courtesy Dr. C.L. Leadem, B.C. Min. of For.) that allows the study of several temperature regimes in combination with several dormancy-breaking treatments on several seed sources all at the same time, we are finding that germination in this species is very temperature sensitive. The early results indicate that temperature combinations such as 20° during the day and 15° during the night, i.e. around 340-degree-days, must be achieved for at least a three-week period during the early part of the season if rapid and complete germination is to occur. Unless climatic conditions conducive to rapid germination coincide with a seed crop shed the previous fall, natural reproduction in *Tsuga mertensiana* will fail. Harvesting methods may need to be adjusted, therefore, to ensure that there is an adequate seed supply into cut blocks on an on-going basis.

As part of a research program on the sustainable development of Pacific yew, *Taxus brevifolia* Nutt., from the bark of which the anti-cancer agent taxol has been isolated, we are also investigating seed dormancy (supported in part by the B.C. Min. of For.). Of approximately 100 seed pretreatments tested so far, only long-term stratification produced any germination.

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## CANADIAN PACIFIC FOREST PRODUCTS LIMITED TREE IMPROVEMENT PROGRAM AND FOREST GENETICS ACTIVITIES

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Keywords: Seed orchards, Douglas-fir, Sitka spruce, western hemlock, yellow-cedar, western red cedar, white pine.

#### SEED ORCHARDS/TREE IMPROVEMENT

The tree improvement program was reviewed during the development of a strategic plan for Saanich Forestry Centre. The seed orchard program was assessed in light of the level of genetic testing for every species, future seed demand, and present seed supply. As a result, a consolidation plan was proposed and its implementation is in progress. At the completion of this plan, seed supply for Douglas-fir, western hemlock, yellow-cedar, and western red cedar will be met from 6 conventional and one hedge seed orchards. The emphases for western white pine and Sitka spruce will be directed towards the establishment of small seed orchards with putatively resistant stock.

Canadian Pacific Forest Products joined the newly formed Regionalized Cooperative Tree Improvement Program for Western Hemlock in the Pacific Northwest (HEMTIC) and continues to cooperate with the B.C. Ministry of Forests and Forestry Canada.

#### RESEARCH

Several research projects were completed/initiated during the period covered by this report (see publications). These include: assessment of inbreeding and genetic variation in a western red cedar seed orchard, genetic evaluation of seed crop harvesting, handling and seedling production practices, assessment of supplemental mass pollination success rate in Douglas-fir seed orchards, *ex-situ* conservation of forest biodiversity, genetic diversity and seed biology of Pacific yew, domestication and biological diversity in Coastal Douglas-fir, genetic diversity in mountain hemlock, and reproductive-cycle plasticity in yellow-cedar. These projects are being conducted in collaboration with scientists from the University of British Columbia, B.C. Ministry of Forests, and Forestry Canada and funded by B.C. Ministry of Forests, the Science Council of B.C., the Forest Resource Development Agreement, and the National Forest Genetic Resources Centre (Green Plan), Forestry Canada

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## RESEARCH AT FORINTEK CANADA CORPORATION RELATING TO TREE IMPROVEMENT

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R.J. Barbour, a scientist with the Resource Assessment Group, Eastern Laboratory in Ottawa, and member of CTIA joined the U.S. Forest Service in Oregon in February, 1993. With his departure and with the shift in priorities at Forintek, studies related to tree improvement have been reduced. Since 1992, tree-improvement-related activity was limited to assessment of relative density of western larch plus trees.

## RELATIVE DENSITY OF WESTERN LARCH PARENT TREES IN B.C. TREE IMPROVEMENT PROGRAM

Forintek has evaluated the breast-height relative density of about 200 western larch plus trees selected by the B.C. Ministry of Forests. The increment cores were divided into juvenile (first 15 growth rings from the pith) and mature wood, and were analyzed separately for relative density. The mean densities were 0.493 gms/cu.cm. for the mature wood with a range of 0.378 to 0.588, and 0.468 gms/cu.cm. for the juvenile wood density, with a range of 0.378 to 0.588. Coefficients of variation were 6.0% and 7.0% for mature and juvenile wood density, respectively. The juvenile-mature wood boundary was made on the basis of an x-ray analysis of breast-height cores from 34 western larch plus trees which showed that wood density levelled out generally after the 10th-15th growth ring from the pith.

## GENETIC IMPROVEMENT OF INTERIOR DOUGLAS-FIR/WESTERN LARCH

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Keywords: Pseudotsuga menziesii var glauca, Larix occidentalis, tree breeding, genetic testing, seed orchards.

Tree breeding programmes for Interior Douglas-fir and western larch were initiated in 1982 and 1987, respectively. The objective of both programmes is to develop welladapted seed, selectively bred to produce trees with improved volume growth and quality, while maintaining acceptable levels of genetic diversity. The breeding strategies for both species are based on recurrent selection for general combining ability, wind-pollinated genetic testing and soil-based, clonal seed orchards.

## PARENT TREE SELECTION/PROPAGATION

For Interior Douglas-fir, selection of first-generation parents from wild stands in eight breeding zones was completed in 1990. Over 1700 trees were selected, propagated by grafting, and established in clone banks and separate breeding arboreta. For western larch, 383 first-generation parents have been selected from wild stands in two breeding zones, propagated by grafting, and established in seed orchards and breeding arboreta. The target number of firstgeneration western larch parents is 550.

## GENETIC TESTING/SEED ORCHARDS

Since 1983, wind-pollinated genetic testing of Interior Douglas-fir has proceeded rapidly. In spring, 1992 the final tests of this initial wave of genetic testing were established in the East Kootenay breeding zone. In total, 1661 families from eight breeding zones are included in tests across 39 sites. For western larch, the East Kootenay and West Kootenay/Shuswap Adams series of wind-pollinated genetic tests were established in 1991 and 1993, respectively.

First-phase clonal Douglas-fir seed orchards will be established for each breeding zone on the basis of parental breeding values for six-year height. In spring, 1993 field-grafting of the top 40 parents commenced for the establishment of the Quesnel Lakes and Cariboo Transition seed orchards. Both orchards will be located on Vernon Seed Orchard Company land in Vernon. In 1990, two clonal first-generation western larch seed orchards were established at the Kalamalka Forestry Centre.

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Keywords: Forest genetics, tree breeding, white spruce, Engelmann spruce, insect resistance.

The objective of this project is to produce genetically improved planting stock of white and Engelmann spruce (*Picea glauca* [Moench] Voss and *P. engelmannii* Parry).

Twenty-year diameter (dbh) measurements were carried out for the Prince George Selection Unit progeny trials. Due to high costs, height measurements were only carried out for a small sample to establish relationships between mean family heights and diameters. Results indicated a strong correlation (r=.91) between the two traits. Correlation between 15-year mean family height and 20-year dbh was also strong (r=.94). Thus, the less costly alternative of dbh assessments were justified. Evaluation of the data indicated that 6-year height measurements would provide very reliable data for determining families to be included in seed orchards and further breeding (Table 1.).

Table 1. Percent gain in 20-year ht, dbh, and volume associated with selection based on early<br/>height and dbh. Gain percentages represent the difference between the mean of the best<br/>25% (44) of families and the mean of the plantations (173 families).

SELECTION	HEIGHT (20-yrs)	DBH (20-yrs)	VOLUME (20-yrs)
CRITERION	%	%	%
Initial ht	7.1	7.7	26.3
3-year ht	10.4	10.3	35.7
6-year ht	11.6	11.5	40.0
10-year ht	12.9	12.6	44.0
15-year ht	13.2	12.4	44.0
15-year dbh	11.8	12.6	42.9
20-year dbh	12.6	13.3	45.4

Continued investigation of the white pine weevil (*Pissodes strobi* [Peck]) problem (Kiss and Yanchuk 1991) provided further evidence of genetic control of resistance. Parental clones of the progenies on which the above study was based have been attacked by weevil at the Kalamalka Forestry Centre. Although statistical analyses are incomplete, it appears that most of the attacked clones had highly weevil susceptible progenies. In order to obtain further insight into the genetics of weevil resistance, a number of crosses were made using putatively resistant and susceptible families. Field testing of the resulting seedlings will provide much needed information regarding the nature of genetic resistance.

Several researchers in a number of organizations are involved with white pine weevil research. In order to coordinate various activities related to weevil research and to create a linkage among researchers in this field, the establishment of a new network is being proposed by the Pacific Forestry Centre in cooperation with the B.C. Forest Service. Researchers and organizations will be requested to join this new network.

A working plan for second generation breeding of interior spruce has been completed. In 1993, an excellent flower crop permitted us to produce most of the required crosses for the second generation breed production. Field trials of these crosses will commence shortly.

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## WESTERN WHITE PINE IMPROVEMENT PROGRAM FOR BRITISH COLUMBIA

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Keywords: Cronartium ribicola, Pinus monticola, disease resistance proteins, DNA markers.

## "OPERATIONAL" PROGRAM OF SELECTION AND RUST SCREENING

Support for tree selection, seed collection, stock rearing and rust inoculation, plus grafting of selected trees, is provided by the B.C. Forest Service under a Memorandum of Understanding. Selection of parent trees toward the target of 300 trees per zone in the coastal and interior zones has nearly been completed: 318 from the coast and 289 from the interior. Of these, only 21 coastal and 8 interior trees were selected for exhibiting "bark" reactions to the rust, and 3 coastal and 3 interior trees exhibited "tolerant" reactions. All other trees were judged "canker-free" after careful examination. Final seed collections should be made in 1994.

Inoculation of 1+1 open-pollination family stock varied more than previously, due to poor rust survival on *Ribes* groves and to aberrant weather. For this reason, all stock inoculated in 1991 was re-inoculated in 1992. Surviving stock from previous inoculations was examined and seedlings exhibiting "useful" defenses from low-spotted (reduced susceptibility) and high-surviving families were selected for cloning for both re-testing and possible seedorchard use. To the end of 1992, 146 seedlings from 90 parent trees had been selected for such purposes.

## **RESEARCH PROGRAMME**

All plantations established 1987-1988 to assess field resistance to rust, or to root diseases, were evaluated. In general, rust infection varied widely by site, due mainly to nearby *Ribes* plants. Northerly plantations, some beyond the range of *Pinus monticola*, suffered more frost damage than those farther south; northern seed sources were damaged less, while exhibiting good vigour. Plantations in white pine's native range displayed less-clear results. Rust infection on Idaho F2 material planted in 1984 by the B.C. Forest Service was less than on untested local seedlots, while growth vigour was good, indicating potential for this seed in southern B.C. areas.

Two demonstration plantations have been established, using open-pollination seedlings from low-spotted vs. high-spotted (low vs. high rust susceptibility) parents. Further demonstration/field-resistance tests have been established using spare family stock from our rust-screening sowings. These may assist in rankings for seed-orchard selection, or for orchard rogueing. Controlled mating of parents judged superior from our operational rust inoculation began in 1992. Seedlings will be used in further inoculations and in field "demonstration" plantations.

Resistance to root diseases, a major factor favouring white pine over its associates in regeneration decisions, is being tested on nine sites. Mortality due to *Armillaria ostoyae* is increasing at two sites, but no damage due to *Phellinus weirii* or *Inonotus tomentosus* has been found.

Extraction and analysis of bark proteins from phenotypically resistant or susceptible trees found 29 proteins statistically different by resistance class. Amino-acid sequencing of two "resistant" proteins was attempted, but has not been completed, due to technical problems. Foliar chitinase in trees exhibiting resistance in leaves was contrasted to those judged susceptible. Slightly-more chitinase was found in the bark of resistant than of susceptible trees. Once proteins considered indicative of resistance to blister rust are isolated, they will be sequenced sufficiently to allow synthesis of peptides and production of antibodies. The antibodies will be used as "immuno probes" to test parents or their progeny for useful resistance, perhaps removing the need for rust inoculation of the families. As a demonstration of the technology's potential, a protein from sugar pine associated with overwintering has been found in western white pine by the anti-sugar pine antibody. Work on rust basidiospores has produced 15 monoclonal antibodies (MABs) specific to blister rust. These MABs will be used in further studies of the pine-rust pathosystem.

Studies on genetic diversity in the blister rust fungus show considerable genetic variation in aeciospores from individual rust cankers, indicating that sexual reproduction occurs, a point previously questioned by some pathologists. Since sexual reproduction could create races of the pathogen varying in virulence, studies were carried out to assess population variation using DNA markers. The ribosomal gene cluster was mapped and sources of restriction site variation and length heterogeneity determined. Variation was high among cankers in a stand. No evidence of geographic distinction was found.

Systems for studying the molecular basis of disease responses in five-needle pines, and for cloning and characterizing the genes involved in these responses, were developed. These methods are being used to identify and clone genes activated in western white pine in response to pathogen attack.

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## GENETIC RESEARCH AND IMPROVEMENT FOR YELLOW-CEDAR, REDCEDAR AND PACIFIC YEW

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Keywords: yellow-cedar, redcedar, Pacific yew, clonal forestry, genetic architecture, selfing, gene conservation.

#### YELLOW-CEDAR

A clonal forestry tree improvement strategy is in place for yellow-cedar (*Chamaecyparis nootkatensis*). Genetic tests, using offspring from parent trees mated in 8-clone disconnected, partial, circular diallels, will be clonally replicated and will serve the dual purpose of providing selections for an advanced generation breeding population and information for selection of improved clones for production. Twelve diallels have been completed (out of a total of 24 proposed) and the first series of genetic tests will be planted in 1994/95. The yellow-cedar short-term genetic architecture and adaptability study has been completed (Russell 1993). Eleven long-term provenance field tests, two with family structure, have been established.

#### REDCEDAR

Past research has indicated that redcedar (*Thuja plicata*) has minimal isozyme variability (Yeh 1988), a low outcrossing rate in seed orchards (El Kassaby unpubl. data), and no inbreeding depression in seed traits (Owens et al 1990). Experiments have been established to investigate extent and pattern of genetic variability in, and the effect of selfing on, morphological and physiological traits pertaining to adaptability and growth.

## PACIFIC YEW

In response to increased interest and subsequent harvesting of Pacific yew, a gene conservation program was established. Over 500 clones have been collected from throughout the range of yew in British Columbia, and are currently being rooted for establishment in clone banks at Cowichan Lake Research Station.

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## **SEED PRODUCTION RESEARCH - B.C. MINISTRY OF FORESTS**

## Joe Webber and Michael Stoehr

## Glyn Road Research Station 1320 Glyn Rd. Victoria, British Columbia V8W 3E7

# Keywords: Supplemental mass pollination, seed orchard management, alternate seed orchard designs, flower induction.

### In Memory of Stephen Douglas Ross

Since our last report, we lost a colleague and friend. Steve Ross died in November, 1991. In passing, Steve left a legacy of research accomplishments that we will all be using for many years to come. Steve was a senior research scientist with the Research Branch of the Ministry of Forests and acted as project leader for the Seed Supply group. His career included many milestones most notable was his contribution to the physiology of flowering in conifers, and the development of the container approach and alternate orchard designs for seed production. He authored or co-authored over 100 scientific publications and served as Associate Editor for the *Canadian Journal of Forest Research* and adjunct professor in the Forest Biology program at the University of Victoria. Steve will be sadly missed by all who knew him.

#### SEED PRODUCTION RESEARCH

Our former approach to solving seed production issues was divided roughly into two disciplines: flowering (Ross) and pollen (Webber) research. We have now joined these two and our focusing on seed production as a single research focus. Michael Stoehr has now joined our research team and brings with him new skills to bear on seed production research. Michael will take on research responsibility associated with seed orchard and wild stand production (see below). Joe will focus principally on barriers to reproductive success including his pollen management research as well as continuing much of Steve's flower induction work and container seed orchard program.

We are currently very active in developing *in vitro* viability assays for all of our important conifers. A recent publication (Webber and Bonnet-Masimbert 1993) details the approach for Douglas-fir (*Pseudotsuga menziesii*) and similar approaches for interior spruce (*Picea spp.*), lodgepole pine (*Pinus contorta*) and western hemlock (*Tsuga heterophylla*) are being developed. A pollen manual for interior spruce has been published and a similar manual for Douglas-fir is in print.

We are also continuing studies on the flower induction of western larch including the  $GA_{4/7}$ , girdling and root pruning effects on both potted and wild stand stock. Some limited data on pollen viability including a germination media for western larch has also been completed (Webber and Ross 1993). Horticultural and environmental effects on interior spruce container seed production are receiving considerable attention. In spite of reaching target production figures of about 50 seed cones per flowering ramet (about 1 m tall), filled seed per cone remains disappointingly low. Both shoot and root temperatures during seed cone

development are being investigated as an important deterrent to obtaining target values of about 50 filled seed per cone.

Future research includes continuing studies for the interior spruce CSO, western larch and seed production problems associated with ambalis fir (*Abies amabalis*).

### SEED ORCHARDS AND SEED PRODUCTION RESEARCH

In Douglas fir (*Pseudotsuga menziesii*) and interior spruce (*Picea* sp.) we have genotyped one clonal orchard each using DNA markers in cooperation with BC Research Corp. This will allow us to carry out paternal analysis to estimate the efficacy of supplemental mass pollination under a range of environmental conditions, selfing rates and outside contamination. In lodgepole pine (*Pinus contorta*), where DNA markers are not yet available, SMP efficacy with respect to timing during receptivity, will be evaluated as a function of filled seed per treated cone.

Established crown management studies will be continued in lodgepole pine and interior spruce to evaluate the effect of crown pruning on long-term seed production and selfing.

A progeny test was established to evaluate the effects of outside-orchard pollen contamination on early seedling growth rhythm and phenology. Six orchard clones were controlpollinated with five single-tree pollen lots from within the orchard and five single-tree contaminating pollen lots. This orchard is a coastal-interior transition zone Douglas fir orchard established on a coastal site. Field tests will be established to further evaluate the effects of coastal male parents versus transition zone male parents on a transition site on survival, frost hardiness and phenology.

## PUBLICATIONS

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## COASTAL DOUGLAS-FIR BREEDING AND GENETIC RESEARCH

## J. H. Woods

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Keywords: Douglas-fir, breeding, advanced generation, inbreeding, harvest index, selection.

A significant recent event in the Douglas-fir breeding program is the retirement of Chris Heaman. Chris made an outstanding contribution to the field of forest genetics and to forestry in B.C. with his work toward understanding the genetics of Douglas-fir. Chris's energy and dedication will be missed.

#### FIRST GENERATION BREEDING PROGRAM

The primary test group of the first generation Douglas-fir (*Pseudotsuga menziesii* (Mirb.) Franco) breeding program consists of 372 parents mated in 62 six-tree modified halfdiallels and tested across 11 sites per series, for a total of 88 sites established between 1975 and 1985. From this material, 65 clones have been selected and a second-generation orchard established at Bowser on Vancouver Island. This orchard will meet seed supply needs in the primary coastal breeding zone for approximately two decades. Selection was based on 12-year (from planting) stem volume, wood density and stem form.

Maintenance and periodic assessment of the diallel sites plus open-pollinated and other sites continues. This material is supplying advanced generation selections, and will provide a better understanding of seed movement and other genetic parameters.

## ADVANCED GENERATION BREEDING PROGRAM

Over 300 clones have been selected, to date, from the diallel series and other tests for a second generation breeding population. This material is being organized into small sublines (12 to 16 clones) for at least one generation, and mated using a complimentary design. A polymix is being used to estimate parental GCA values, and a circular (partial diallel) design with families planted in blocks is being used for forward selection of a third generation. Early-testing will be used, with intensive site preparation, one by one meter spacing and nearly complete weed control. The first poly-mix and circular matings were done in 1993 on young potted grafts induced with root pruning and  $GA_{4/7}$ .

Support research for the advanced generation program includes low-level inbreeding work, early testing, harvest index, wood density and quality (cooperation with the University of British Columbia) and freeze testing (cooperation with the University of Victoria). In addition, area-based realized gain trials were established to demonstrate volume gains per unit area and to link single-tree progeny tests with area-based yield. Design work is underway to develop more series of realized-gain trials.

## PROCEEDINGS

**OF THE** 

## **TWENTY-FOURTH MEETING**

**OF THE** 

# CANADIAN TREE IMPROVEMENT ASSOCIATION

Part 2

# THE FUITURE FORESTS: OPTIONS & ECONOMICS

Held in Fredericton, New Brunswick August 15-19, 1993

Editor:

J. Lavereau

## **COMPTES RENDUS**

## DE LA

## VINGT-QUATRIÈME CONFÉRENCE

DE

## L'ASSOCIATION CANADIENNE POUR L'AMÉLIORATION DES ARBRES

## 2<sup>e</sup> PARTIE

# LES FORÊTS DE L'AVENIR: LES OPTIONS ET L'ÉCONOMIE

Fredericton (Nouveau-Brunswick) du 15 au 19 août 1993

**Rédactrice:** 

J. Lavereau

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## The members of the 24th CTIA/ACAA executive were:



- Front Row: Greg Adams, Kathy Tosh, Judy Loo, Dale Simpson
- Back Row: Yill Sung Park, Don Fowler

## WELCOMING REMARKS — K. TOSH

Good Morning Honorable Minister, Invited Speakers, Ladies and Gentlemen. Welcome to New Brunswick, Bienvenue à Nouveau Brunswick. I would like to officially open the 24th biennial meeting of the Canadian Tree Improvement Association. Je voudrais ouvrir officielment la 24ième reunion de l'Association Canadienne pour l'Amelioration des Arbres. We are very pleased and honored to be hosting this international event and are fortunate to have such an excellent slate of speakers with us today.

The theme of this year's meeting is "*The Future Forests, Options and Economics*". This theme, I believe is especially appropriate to our situation in New Brunswick where first generation orchards are producing improved seed, and second generation orchards are currently being established. We are now at the point where long-term decisions must be made about the program's direction. There are more breeding strategy options available to tree improvement programs than ever was in the past. Today we will hear some of these options. The second part of the symposium is the economics, and I think in all aspects of forestry and not just tree improvement this is a major consideration. Again we have some excellent speakers who will address this issue.

Tree Improvement in New Brunswick has made considerable progress since the last CTIA meeting was held in Fredericton 20 years ago, and this is due in large part to the cooperative nature of tree improvement in New Brunswick. The New Brunswick Tree Improvement Council and its members made up of industry, government and university agencies have worked hand in hand for the future benefit of all users of the forests.

I would also like to take this opportunity to thank the sponsors without whom this meeting would not have been possible:

Canada/New Brunswick Cooperation Agreement on Forest Development Fraser Inc. J.D. Irving Ltd. Juniper Lumber Co. Ltd. Miramichi Pulp and Paper Inc. Natural Resources Canada - Maritimes Region N.B.I.P. Forest Products Inc. N.B. Dept. Natural Resources and Energy Stone-Consolidated Inc. Université de Moncton

Finally I would like to wish everyone a successful and productive meeting. J'espere que vous aurez un bon congres.