Plant Canada 2005
Schedule & Abstracts

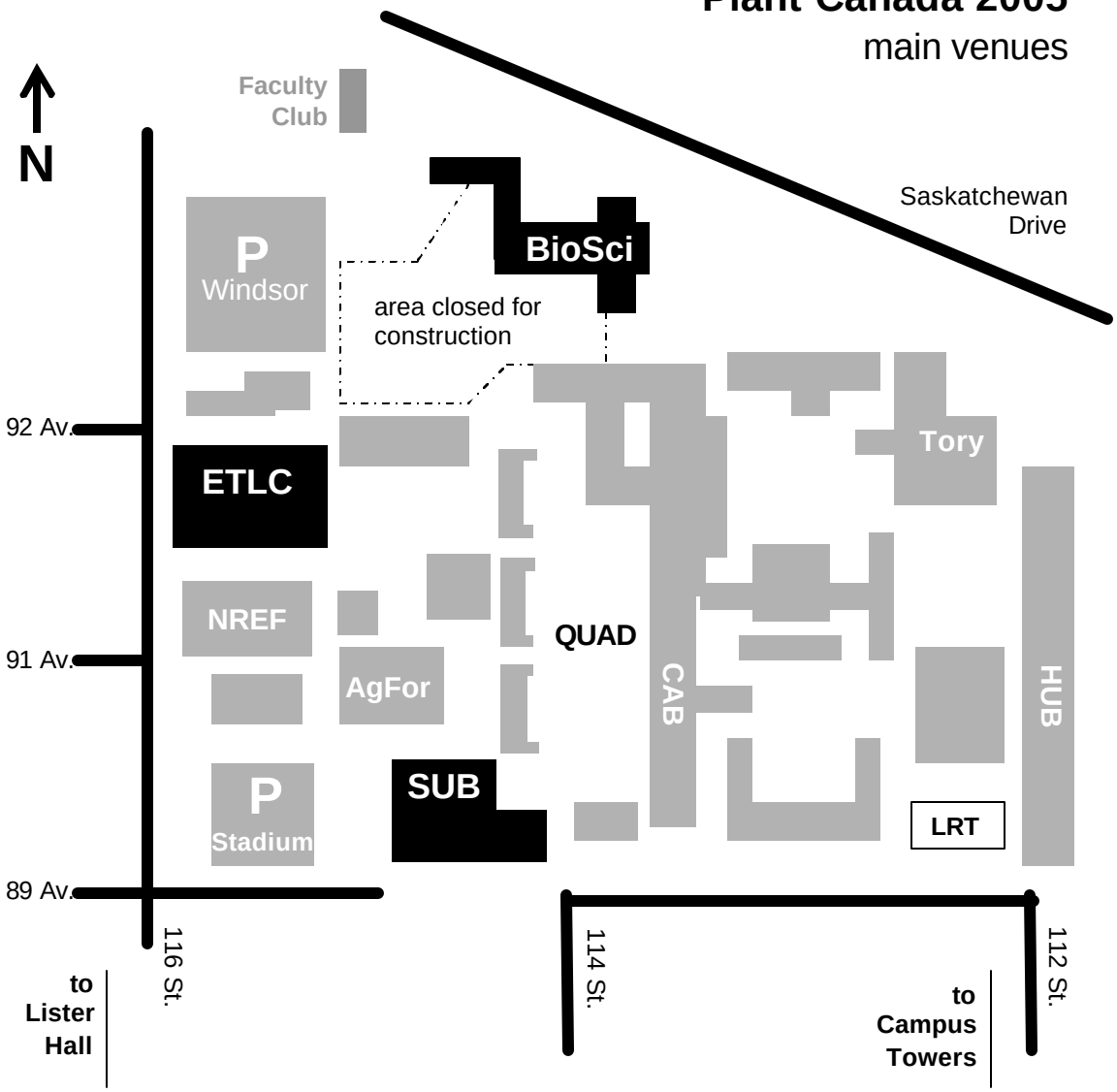
Edmonton, Jun 15-18

General Information

Daily Schedules

Abstracts

Cover artwork: Cow Parsnip, Annora Brown; Watercolour on paper. Gift of Emma Read Newton, University of Alberta Art and Artifact Collection, Museums and Collections Services.



pizza, italian, sushi, fast food and other restaurants along 109 St...

...and along 82 Ave.

SUB (Students Union Building; 2nd Floor)

- Registration desk
- Horowitz Theatre (HOR): plenary sessions and some symposia.
- Dinwoodie Lounge (DINW): exhibits & posters

ETLC (Engineering Teaching and Learning Center/ Maier Learning Center)

- Oral sessions in room numbers beginning with prefix "ET"

NREF (Natural Resources Engineering Facility)

- Oral sessions in room numbers beginning with prefix "NR"

Fast food and some other **restaurants** are available in SUB, HUB, and along 109 St., and along 82 (Whyte) Ave. After 5PM, most businesses on campus will be closed, with the exception of a few fast food restaurants in HUB.

Posters & Exhibits

The poster and exhibit hall in Dinwoodie Lounge (SUB, 2nd floor) will be open during the following hours:

Wednesday, June 15	9:00AM – 12:00PM	Open for set up
	12:00PM – 9:30PM	Viewing
		Poster Reception 7:30-9:30 PM Presenters with even-numbered poster abstracts (e.g. P100) please attend your poster from 7:30-8:30PM. Presenters with odd-numbered poster abstracts (e.g. P101) please attend your poster from 8:30-9:30PM.
Thursday, June 16	10:30AM – 7:30PM	Viewing
Friday, June 17	10:30AM – 2:00PM	Viewing
	2:00PM – 4:00PM	Open for take-down. All posters must be removed before 3:00PM

Oral Presentations

Please come to the session room at least 15min. before the session start time to load your presentation onto the projector computer. Your presentation should be in WindowsXP-compatible Power Point format, on a CD-ROM or a USB RAM drive/memory stick. You will not be able to connect your personal computer to the projector.

Special Lectures

CSPP Gold Medal Symposium Thu, Jun 16 7:30 – 9:00PM ET 2001
My long journey in plant research: contributions to flavonoid biochemistry
Ragai Ibrahim, Concordia University

CBA Luella K. Weresub Mycology Lecture Thu, Jun 16 7:30 – 9:00PM ET 2002
Rust fungi and a rusty mycologist: The morphology, life cycle and taxonomy of rust fungi
Yasu Hiratsuka, Canadian Forest Service, Edmonton

Special Plenary Lecture Fri, Jun 17 11:00AM – 12:15PM HOR
The challenge of the 21st century: setting the real bottom line
David Suzuki, David Suzuki Foundation
n.b. this lecture will be followed by a book signing at the Univ. of Alberta Bookstore

Special Meetings (open to all conference participants)

Can. J. Bot. name change discussion Wed, Jun 15 9:00 – 10:15 AM HOR
Come give your opinion about a potential change to the name of the Canadian Journal of Botany, and other issues related to this journal. Hosted by Larry Peterson.

NSERC information session Fri, Jun 17 12:15 – 1:45PM ET1003
Hosted by Brigit Viens, Program Officer, NSERC Research Grants.

Poplar discussion group Fri, Jun 17 7:00PM – 9:00PM NR2090
Opening the 'model tree' tool-box: A discussion of new opportunities and challenges for poplar research. Hosted by Brian Ellis.

Society General & Executive Meetings

Sandwiches are provided for all noon and supper-hour meetings (for large meetings, this is a box lunch). Coffee is provided for all executive meetings not scheduled over a noon hour.

Plant Canada			
outgoing board meeting	Tue, Jun 14	5:00PM – 7:00PM	SUB4-24
incoming board meeting	Fri, Jun 17	12:15PM – 1:45PM	SUB4-20
CBA			
outgoing executive meeting	Tue, Jun 14	3:00PM – 5:00PM	SUB4-24
incoming executive meeting	Sun, Jun 19	9:00AM – 12:00PM	SUB4-24
annual general meeting	Sat, Jun 18	12:30PM – 2:00PM	ET1003
CPS			
outgoing executive meeting	Tue, Jun 14	9:00AM – 5:00PM	off campus
business meeting	Sat, Jun 18	3:45PM – 5:15PM	ET1018
awards luncheon	Sat, Jun 18	12:00PM – 2:00PM	Lister Dining
incoming executive meeting	Sun, Jun 19	9:00AM – 5:00PM	off campus
CSA			
annual general meeting	Thu, Jun 16	12:30PM – 2:00PM	ET1003
CSHS			
annual general meeting	Sat, Jun 18	11:00AM – 2:00PM	ET1001
CSPP			
outgoing executive meeting	Wed, Jun 15	8:00AM – 10:15AM	SUB4-20
incoming executive meeting	Sat, Jun 18	12:00PM – 2:00PM	SUB4-20
business meeting	Thu, Jun 16	12:30PM – 2:00PM	ET1001

Awards

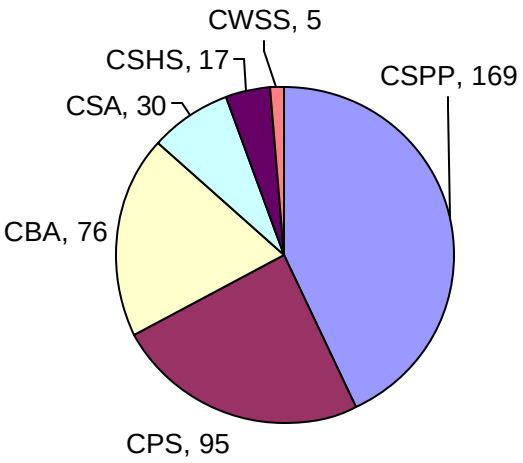
Some societies will adjudicate presentations by their student members, and present awards to these students at either their general meeting or at the closing banquet. Check with society representatives for details.

Conference Statistics

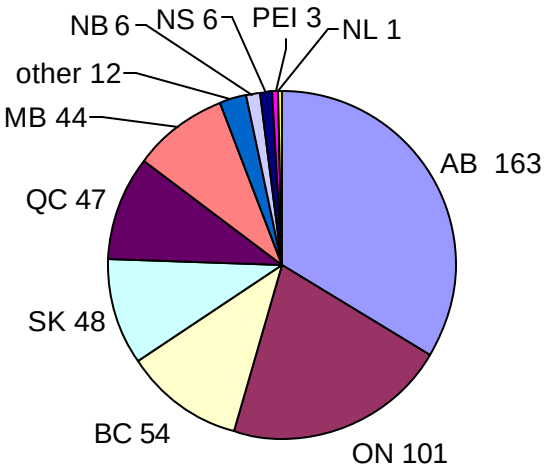
For the curious, and for future conference planners, here are some attendance and budget figures for Plant Canada 2005. This information is current as of May 2005 and is subject to change.

Attendance:

A total of 485 confirmed registrants are represented by the figures below. This includes paid registrants, plus invited speakers, sponsors, and volunteers. The figure on the left shows confirmed registrants classified by society membership (not all registrants are society members, and some are more than one society). The figure on the right shows all confirmed registrants classified by province of residence.



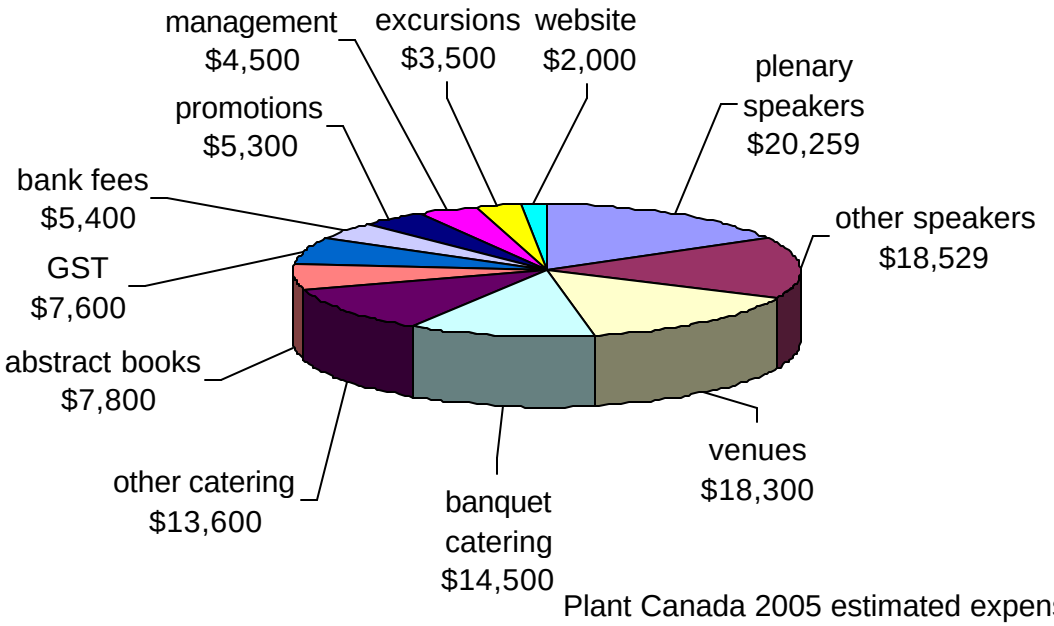
Attendance by society membership.



Attendance by province of residence.

Budget:

At the time of printing, $108,520 had been received from 423 paid registrations, and $15,000 had been received in paid sponsorships. Estimated expenses (detailed below) totaled $121,288. More revenues are expected from pending sponsorships and registrations. Any profit will be distributed among the participating societies in proportion to their members' attendance at the conference. Detailed financial information will be provided to the executive board of Plant Canada after the close of the meeting.



Plant Canada 2005 estimated expenses

plenary speakers & other invited speakers: Full or partial travel expenses for 26 invited speakers including plenary and special symposium speakers.
venues: Rental of: Horowitz Theatre, Dinwoodie Lounge, and other lecture theatres, board rooms and banquet rooms on U of A campus and Ft. Edmonton Park. Also includes rental cost of audio visual equipment.
banquet catering: Food and transportation to banquet at Ft. Edmonton Park.
other catering: Receptions, awards luncheons, box lunches, pizza lunches, coffee breaks and continental breakfasts.
GST: 7% GST collected on all registrations (exclusive of society membership fees)
credit card processing: 5% fee charged by bank and on-line transaction verification service
promotions: posters, mail-outs, tote bags
management: stipends and office costs for part-time conference manager, excursion leaders
excursions: transportation and admission charges
website: software, set-up, and server charges

Plant Canada (the federation)

Plant Canada 2005 is organized under the direction of Plant Canada, a federation of Canadian plant science societies. The following information about Plant Canada is also available at the federation's website: **www.plantcanada.ca**.

Purposes

- To organize and sponsor regular, effective scientific meetings and workshops under a national umbrella for plant science and related disciplines in Canada
- To operate and maintain a strong communication network among Member Societies and their individual members
- To be a strong and effective force for public education and advocacy in plant and related sciences

History

Prior to the formation of Plant Canada, the discipline of Plant Science or Plant Biology in Canada was fragmented into many small societies, each of which often met separately. In 1997, the Presidents of the CSPP and the CBA began discussing the possibility of merging the two societies. A committee to explore this idea was struck and was headed by Dr. Iain Taylor (UBC); this committee met several times during the next three years. It was soon established that a federation of existing societies was more agreeable than a merger and, at the 2000 joint meeting of the CBA and the CSPP, the Constitution and By-laws for the new federation were ratified in the business meetings of both societies. Since that time, we have been pleased to welcome four other societies as continuing members.

Executive & Board of Directors (as of Mar 22, 2005)

Carol Peterson, President
Paul Cavers, Secretary
Deep Saini
Rob D. Guy
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Christian Lacroix
Richard A. Martin
Bruce Gossen
Denise Maurice
Anne Légère
Yousef Papadopoulos
Gavin Humphreys
Yves Desjardins
Shahrokh Khanizadeh

Please direct suggestions or comments about Plant Canada 2005 to the Plant Canada board members. Contact information is available at www.plantcanada.ca

Next meeting: Plant Canada 2007, Saskatoon Lead organization: CPS

Plant Canada 2005 Participating Societies

Plant Canada 2005 is a joint meeting of the following six scientific societies:

- CBA (Canadian Botanical Association) http://www.uoguelph.ca/botany/cba/
- CPS (Canadian Phytopathological Society) http://www.cps-scp.ca/cps.htm
- CSA (Canadian Society of Agronomy) http://www.agronomycanada.com/about.html
- CSHS (Canadian Society for Horticultural Science) http://www.cshs.ca/
- CSPP (Canadian Society of Plant Physiologists) http://www.cspp-scpv.ca/
- CWSS (Canadian Weed Science Society) http://www.cwss-scm.ca/

Organizing Committee

National Organizing Committee

Mike Deyholos, Chair
Shaffeek Ali, CWSS-SCM
Randy Currah, CBA-ABC
David Ehret, CSHS-SCSH
Denis Gaudet, CPS-SCP
Roisin Mulligan, CBA-ABC
Jan Slaski, CSPP-SCPV
Dean Spaner, CSA-SCA

Local Volunteers

JoAnn Collins, Chair (and part-time Manager)
Adriana Arango
Belay Ayele
Disa Brownfield
Matt Bryman
Marie Davey
Melissa Day
Adrienne De Corby
Aaron Donahue
Jenna Francis
Matthew Gaudett
Matt Greif
Katherine Hunter
Terry James
Yuanqing Jiang
Abdul-Hameed Khadhair
Manjeet Kumari
Stefan Little
Frank Liu
Diana Lopez
Carla Medina
Yo Miyashita
Tara Narwani
Ryan McKenzie
Randall Mindell
Laura Paterson-Fortin
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Adrianne Rice
Melissa Roach
Aurea Siemens
Sam Skinner
Selena Y. Smith
Winnie Wang
Bill Yajima

Sponsors and exhibitors

Alberta Research Council
www.arc.ab.ca

Alberta Innovation and Science
www.innovation.gov.ab.ca

University of Alberta Conference Fund
University of Alberta, Faculty of Science
University of Alberta,
Department of Biological Sciences
www.ualberta.ca

Li-Cor Biosciences
www.licor.com

Canada Institute for Scientific and Technical Information (NRC Research Press)
cisti-icist.nrc-cnrc.gc.ca/cisti_e.html

Agricultural Institute of Canada
www.aic.ca

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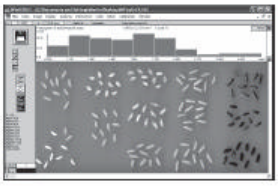
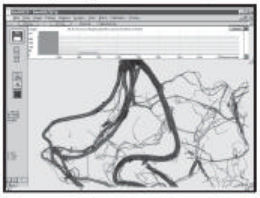


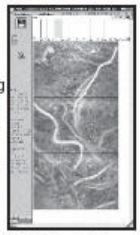
WinRHIZO™ - root morphology, projected & surface area, volume, root topology & architecture, color,...



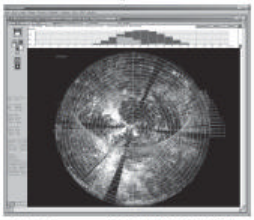
WinCAM™ - color area measurement of soil area covered with grass, color level and uniformity, plant disease and defect areas,...

(Photo left is a courtesy of Dr. P. Salon, USDA-NRCS Plant Material Program)

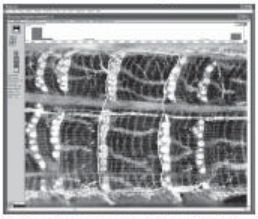
WinRHIZO Tron™ - Interactive analysis of roots in rhizotron & soil: distribution of root length, area, volume & number of tips as function of diameter • Analyse simultaneously neighbouring images of a tube in time & track root growth



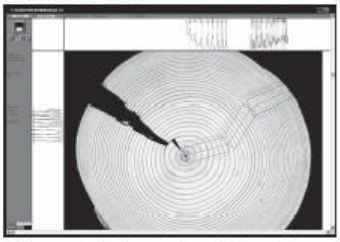
WinSCANOPY™ - Canopy & solar radiation analysis from hemispherical images captured by a standard digital or DSLR camera with calibrated fisheye lens (180°) on a self-leveling mount



WinCELL™ - Anatomical analysis & quantification of wood-cell structure parameters over annual rings, color analysis, automatic / interactive analysis modes, debris filtering,...



WinDENDRO™ - Tree-ring analysis & more: interactive quantification of compression & reaction wood, disk area, perimeter, form coefficient, average radius, rings/cm in function of distance to pit,...



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Overview: Wednesday, June 15

8:30 AM	Registration desk opens.	
8:30-10:00	CSPP Outgoing Executive Meeting	SUB4-20
9:00	Poster hall open for set-up and viewing	DINW
9:00-10:15	Open Meeting Canadian Journal of Botany: discussion of journal name change	HOR
10:15-12:00	**Plenary Session I** **Roots and the underground environment**	HOR
12:00- 2:00	Lunch. Free to all conference registrants.	
2:00-3:30	**Concurrent Sessions Block A**	
	A1. Nutrient use efficiency (CSPP symposium, part 1)	HOR
	A2. Contemporary issues in statistics, data management, plant biology and agricultural research (CSA/CSHS symposium part 1)	ET2001
	A3. Systematics and evolution I	ET1003
	A4. Development of the vascular system	ET1013
	A5. Pulse crops	ET1017
	A6. Pathology: molecular aspects I	ET1001
	A7. Fusarium I	ET1018
3:30-3:45	Break. No refreshments provided.	
3:45-5:15	**Concurrent Sessions Block B**	
	B1. Nutrient use efficiency (CSPP symposium , part 2)	HOR
	B2. Contemporary issues in statistics, data management, plant biology and agricultural research (CSA symposium, part 2)	ET2001
	B3. Systematics and evolution II	ET1003
	B4. Development and plant structure	ET1013
	B5. Molecular aspects of abiotic stress	ET1017
	B6. Gene expression in pathogenesis	ET1001
	B7. Fusarium II	ET1018
5:15-7:30	Dinner break. No food provided by conference organizers.	
7:30PM	Registration desk closes	
7:30-9:30	**Poster Reception.** Soft drinks and snacks provided.	DINW

Sessions Detail: Wednesday, June 15

8:30 – 10:00 CSPP Outgoing Executive Meeting

9:00 – 10:15 Open meeting
Canadian Journal of Botany journal name change discussion.

10:15 – 12:00 Plenary session

Plenary session Room: Horowitz

I. Roots and the underground environment

10:15 I.1 **A systems approach to understanding root development**
Philip Benfey, Duke University

11:00 I.2 **The role of arbuscular mycorrhizal fungi in terrestrial communities and ecosystems**
John Klironomos, University of Guelph

11:30 - 12:00 I.3 **Invisible ecosystems**
Scott Wilson, University of Regina

12:00 – 2:00 Lunch

2:00 – 5:15 Concurrent sessions A, B

Special Symposium Room: Horowitz

A1. Nutrient Use Efficiency (part 1)

Chair: Allen Good

2:00 A1.1 **Legume genomics and biotechnology: role in sustainable acquisition of nutrients**
Carroll Vance, University of Minnesota

2:45 A1.4 **Physiological role of alanine aminotransferase in *Arabidopsis thaliana***
Yo Miyashita, University of Alberta

3:00 A1.5 **What poplar trees choose to eat: 15N discrimination as an indicator of nitrogen dynamics in *Populus trichocarpa***
Hannah Buschhaus, University of British Columbia

3:15 - 3:30 A1.6 **Nitrogen use efficiency of field-grown bell green peppers**
Laura Van Eerd, University of Guelph

Special Symposium — Room: ET2001

A2. Contemporary issues in statistics, data management, plant biology and agricultural research (part 1)

Chair: Alan Moulin

2:00	A2.1	**Mixed model analyses (REML) for three different series of agronomic trials.** Ken McRae, Agriculture and Agri-Food Canada, Kentville, NS
2:30	A2.3	**Biplot analysis of genotype by environment data** Weikai Yan, ECORC-AAFC, Ottawa
3:00 -3:30	A2.5	**Information systems for crop performance data: motivation, status, and potential** Nicholas Tinker, Agriculture and Agri-Food Canada, Ottawa

A3. Systematics and Evolution I

Room: ET1003

2:00	A3.1	**Near the root of monocot phylogeny?** Sean Graham, University of British Columbia
2:15	A3.2	**The phylogeny of *Danthonia* DC. (Poaceae: Danthonioideae) in North America** Elizabeth Reimer, University of Saskatchewan
2:30	A3.3	**Anomalous periderm in Middle Eocene *Decodon allenbyensis*: Understanding aerenchyma development and the importance of a novel aquatic bark system.** Stefan Little, University of Alberta
2:45	A3.4	**Quantitative relationships between floral characters in the genus *Anthurium* (Araceae)** Mathieu Chouteau, Université de Montréal
3:00	A3.5	**Molecular phylogeny of the genus *Philodendron* (Araceae) : clarification of its taxonomic position and species level classification** Marie-Pierre Gauthier, Université de Montréal
3:15 -3:30	A3.6	**Inference of C4 photosynthesis evolution in *Flaveria* (Asteraceae) based on phylogeny** Athena McKown, University of Toronto

A4. Development of the vascular system

Room: ET1013

2:00 A4.1 **Vein pattern formation in Arabidopsis leaves**
Elizabeth Schultz, University of Lethbridge

2:15 A4.2 **Cloning and expression analysis of *FKD1* - a gene that directs vein formation in response to auxin**
Hong Wei Hou, University of Lethbridge

2:30 A4.3 **Arabidopsis eIF-5A1 is a positive regulator of xylem development**
Fengshen Ma, University of Waterloo

2:45 A4.4 **Promoter activities of deoxyhypusine synthase and eukaryotic translation initation factor 5A1 in *Arabidopsis thaliana* suggest a role in xylogenesis, leaf senescence and flower senescence**
Jeremy Duguay, University of Waterloo

3:00 A4.5 **Changes in cherry fruit tracheary element structure and function during development and ripening.**
Carol Peterson, University of Waterloo

3:15 -3:30 A4.6 **"Pre- and post- genome" approaches to investigating the genetic regulation of wood development**
Lee Johnson, University of British Columbia

A5. Pulse crops

Room: ET1017

2:00 A5.1 **Intercropping pulse species with barley: assessing agronomic feasibility and benefits**
Sheri Strydhorst,University of Alberta

2:15 A5.2 **Alternative tools for minimizing the damage of ascochyta blight in chickpea**
Yantai Gan, Agriculture and Agri-Food Canada, Swift Currrent, SK

2:30 A5.3 **Evaluation of disease management strategies for ascochyta blight of chickpea in Alberta**
Hafiz Ahmed, Alberta Research Council

2:45 A5.4 **Validation of assessments of partial resistance to mycosphaerella blight in pea**
Bruce Gossen, Agriculture and Agri-Food Canada, Saskatoon

3:00 A5.5 **Impact of lodging and foliar fungicide on severity of mycosphaerella blight and yield in field pea lines**
Bruce Gossen, Agriculture and Agri-Food Canada, Saskatoon

3:15 -3:30 A5.6 **Etiology of Stemphylium blight of lentil (*Lens culinaris*) in Saskatchewan.**
Edmore Mwakutuya, University of Saskatchewan

A6. Pathogenesis: molecular aspects I

Room: ET1001

2:00 A6.1 **Differential expression and localization patterns of type III peroxidase genes isolated from *Blumeria graminis*-infected wheat epidermal cDNA library**
Guosheng Liu, University of Saskatchewan

2:15 A6.2 **Towards the identification of the biological function of N gene**
Prasanna Bhomkar, University of Lethbridge

2:30 A6.3 **Spatial organization and detoxification roles of thiol methyltransferases in relation to the glucosinolate-myrosinase defense system in plants**
Priyum Koonjul, Institut de Recherche en Biologie Végétale, Montréal

2:45 A6.4 **The N-terminal trunk of plant cystatins determines their inhibitory specificity against cysteine proteinases**
Dominique Michaud, Université Laval

3:00 -3:15 A6.5 **IPTG-controlled gene expression during symbiosis of *Sinorhizobium meliloti* with alfalfa**
Sirin Adham (Yaeesh), University of Waterloo

A7. Fusarium I

Room: ET1018

2:00 A7.1 **Assessment of artificial inoculation methods and deoxynivalenol levels in barley lines representing various candidate sources of resistance to Fusarium Head Blight.**
Jennifer Geddes, Agriculture and Agri-Food Canada, Lethbridge

2:15 A7.2 **Efficiency of cycloheximide and deoxynivalenol for in vitro screening for Fusarium Head Blight resistance in wheat**
Hassan Soltanloo, Agriculture and Agri-Food Canada, Lethbridge

2:30 A7.3 **First report of association between soybean sudden death syndrome (SDS), caused by *Fusarium solani* f.sp. *Glycines*, and black walnut (*Juglans nigra*)**
Ameur Manceur, University of Guelph

2:45 A7.4 **Potential mechanisms of biological control by *Gliocladium catenulatum* against fusarium root and stem rot of greenhouse cucumber**
Syama Chatterton, Simon Fraser University

3:00 A7.5 **Predicting fusarium head blight and DON in spring wheat by quantifying spores per head following anthesis.**
Xiaowei Guo, University of Manitoba

3:15 -3:30 A7.6 **Resistance of *Brassica napus* Cultivars to *Fusarium oxysporum* f.sp. *conglutinans***
Jeremy Klassen,University of Manitoba

3:30 – 3:45 Break
3:45 – 5:15 Concurrent session B

Room: Horowitz

Special Symposium

B1. Nutrient Use Efficiency (part 2)

Chair: Allen Good

3:45	B1.1	**Nitrogen Use Efficiency and Crop Improvement** Steve Rothstein, University of Guelph
4:30	B1.4	**Ionomic profiling of alfalfa varieties to enhance nutrient quality of cattle feeds.** Neil Harris, University of Alberta
4:45 -5:00	B1.5	**Transcript profiling of *Brassica napus* exposed to long-term phosphate stress** Tara Narwani, University of Alberta

Special Symposium

Room: ET2001

B2. Contemporary issues in statistics, data management, plant biology and agricultural research (part 2)

Chair: Alan Moulin

3:45	B2.1	**Multivariate analysis of agronomic data** Norm Kenkel, University of Manitoba
4:15	B2.3	**Spatial variability of soil and the scaling problem** R. Gary Kachanoski, University of Alberta
4:45 -5:15	B2.5	Roundtable discussion

B3. Systematics and evolution II

Room: ET1003

3:45 B3.1 **The evolutionary origins of *Erigeron trifidus***
Jenny Burke, University of Lethbridge

4:00 B3.2 **Sequence heterogeneity of the envelope-like domain in the Egyptian cotton *Gossypium barbadense***
Essam Zaki, Mubarak City for Research

4:15 B3.3 **Phylogeny of *Populus* (Salicaceae) based on Sequence Characterised Inter Simple Sequence Repeat (SCISSR) data**
Mona Hamzeh, Concordia University

4:30 B3.4 **Fagaceae fruits from the Eocene of Vancouver Island, BC**
Randal Mindell, University of Alberta

4:45 B3.5 **Permineralized monocots from the Middle Eocene Princeton Chert**
Selena Smith, University of Alberta

5:00 -5:15 B3.6 **Comparative genomics to assess gene age and horizontal gene transfer**
Tom Hsiang, University of Guelph

B4. Development and structure

Room: ET1013

3:45 B4.1 **A role for ethylene in trichome branching**
Jonathan Plett, Queen's University

4:00 B4.2 **A structural analysis of peridial development in *Aporothielavia leptoderma* has ecological and taxonomic significance.**
Matthew Greif, University of Alberta

4:15 B4.3 **Arabidopsis RACK1 mediates multiple developmental processes and acts with the heterotrimeric G-protein complex to positively regulate lateral root initiation and development**
Jin-Gui Chen, University of British Columbia

4:30 B4.4 **Chemistry and Structure of Soybean Root Epidermis**
Xingxiao Fang, University of Waterloo

4:45 B4.5 **Unusual characteristics of early endosperm and embryo development in the dwarf mistletoe (*Arceuthobium americanum*): formation of an endosperm caecum and dislodgment of the zygote from the embryo sac wall**
Cynthia Ross, Thompson Rivers University

5:00 -5:15 B4.6 **Plant cell architecture - The structural role of actin filaments and microtubules in anisotropic cellular growth**
Anja Geitmann, Université de Montréal

B5. Molecular biology of abiotic stress

Room: ET1017

3:45 B5.1 **RNA secondary structure functions as a thermosensor**
Jessica Brown, University of Alberta

4:00 B5.2 **Multiple heat signaling pathways in tobacco cells**
Raj Dhindsa,McGill University

4:15 B5.3 **Characterization of a novel stress-induced member of the GRAS family in tobacco**
Beatrix Czikkel, University of Western Ontario

4:30 B5.4 **A novel stress-regulated protein interacts with the transcriptional activator GCN5 in *Arabidopsis thaliana***
Abdelali Hannoufa, Agriculture and Agri-Food Canada

4:45 B5.5 **Cellular responses to aluminum stress : Genomics and Proteomics approach**
Manjeet Kumari, University of Alberta

5:00 -5:15 B5.6 **Promoter analysis in transient assay using anthocyanin reporter gene constructs in wheat (*Triticum aestivum* L.)**
Ketan Doshi, Agriculture and Agri-Food Canada, Lethbridge

B6. Gene expression in pathogenesis

Room: ET1001

3:45 B6.1 **Analysis of the cell wall and secreted proteomes of *Sclerotinia sclerotiorum***
Bill Yajima, University of Alberta

4:00 B6.2 **Bacterial phytotoxin acts in systemic activation of the plant antimicrobial compound**
Babana Amadou, Sherbrooke University

4:15 B6.3 **Expression analysis of a lipase gene from *Fusarium graminearum***
Jie Feng, University of Saskatchewan

4:30 B6.4 **Gene expression analysis during mycoparasitism**
Danielle Morissette, McGill University

4:45 B6.5 **Transcriptome analysis of *Colletotrichum coccodes*/ *Abutilon theophrasti* pathogenic interaction**
Amelie Dauch, McGill University

5:00 -5:15 B6.6 **US-1 and US-8 genotypes of *Phytophthora infestans* differentially induced the expression of genes encoding pathogenesis-related proteins PR-2, PR-3 and PR-9 in potato**
Xiben Wang, University of Manitoba

B7. Fusarium II

Room: ET1018

3:45	B7.1	**Application of DNA methods to identify and detect *Pythium*, *Phytophthora* and *Fusarium* species associated with soybean root rot in eastern Ontario and Québec** Tharcisse Barasubiye, Agriculture and Agri-Food Canada, Ottawa
4:00	B7.2	**Comparison of inoculum sources on development of fusarium head blight and deoxynivalenol content in wheat in a disease nursery** Allen Xue, Agriculture and AgriFood Canada, Ottawa
4:15	B7.3	**Corn transformation for Fusarium resistance** Anju Gulati-Sakhuja, University of Guelph
4:30	B7.4	**Diversity of *Gibberella zeae* isolates from Manitoba** Mathieu Dusabenyagasani, University of Manitoba
4:45 -5:00	B7.5	**Introgression of Fusarium head blight (FHB) resistance in Canadian wheat germplasm by *in vitro* and marker-assisted selections.** Ana Badea, Agriculture and Agri-Food Canada, Lethbridge

5:15 – 7:30 Supper break

7:30 – 9:30 PM Poster reception

Overview: Thursday, June 16

8:00 AM	Registration desk opens.	
	Continental breakfast. Free to all conference participants.	HOR
8:30-10:30	**Plenary Session II** Global food security and biotechnology	HOR
10:30	Posters and exhibits open for viewing.	
10:30-11:00	Break. Coffee and other beverages provided.	
11:00-12:30	**Concurrent Sessions Block C**	
	C1. Biochemistry I	ET2001
	C2. Forest biodiversity	NR1001
	C3. Bioproducts and alternative crops	ET1013
	C4. Hormones in abiotic stress	ET1017
	C5. Diseases of horticultural species	ET1018
	C6. Pathology: molecular aspects II	ET1001
	C7. Fusarium III	ET1003
	GIS Workshop (advance payment required)	BioSci B-418
12:30- 2:00	Lunch. No food provided by conference organizers, except for meeting participants	
12:30- 2:00	CSPP Business Meeting. Box lunches provided.	ET1001
12:30- 2:00	CSA General Meeting. Box lunches provided.	ET1003
2:00-3:30	**Concurrent Sessions Block D**	
	D1. Shoot apical meristems (CBA symposium, part 1)	ET1001
	D2. Phytochemicals in human health research: bioactivity vs. physiological relevance. (CSHS symposium, part 1)	ET1003
	D3. Conservation and diversity I	ET1018
	D4. Salinity stress	ET2001
	D5. Biochemistry II	NR1003
	D6. Crop resistance to disease	ET1017
	D7. Brassica pathogens	ET1013
	D8. Teaching in the plant sciences	NR1001
3:30-3:45	Break. No refreshments provided by conference organizers.	
3:45-5:15	**Concurrent Sessions Block E**	
	E1. Shoot apical meristems (CBA symposium, part 2)	ET1001
	E2. Phytochemicals in human health research: bioactivity vs. physiological relevance. (CSHS symposium, part 2)	ET1003
	E3. Conservation and diversity II	ET1018
	E4. Rhizosphere	ET1013
	E5. Reactive oxygen	ET2001
	E6. Pathogen detection	ET1017
	E7. Teaching in the plant sciences	NR1001
7:30 PM	Posters and exhibits close. Registration desk closes.	
7:30-9:00PM	CSPP Gold Medal Symposium	ET2001
7:30-9:00PM	CBA Weresub Lecture	ET2002

Sessions Detail: Thursday, June 16

8:00 Continental breakfast
8:30 – 10:30 Plenary session

Plenary Session — Room: Horowitz

II. Global Food Security and Biotechnology

8:30	II.1	**Global Food Security and Biotechnology** MS Swaminathan, MS Swaminathan Foundation
9:20	II.2	**Biotechnology and world hunger: controversy and solutions** Herbert Kronzucker, University of Toronto
9:55	II.3	**Making plants medicinal: Transgenic tobacco, a biosafe platform for the production of biopharmaceuticals for oral administration** Jim Brandle, AAFC London

10:30 – 11:00 Coffee break

11:00 – 12:30 Concurrent sessions C

C1. Biochemistry

Room: ET2001

11:00	C1.1	**Differential biological availability of iron chelated with N,N'-di(2-hydroxyethyl)-ethylenediamine-N,N'-diacetic acid (HBED) to two Strategy I green algae** Harold Weger, University of Regina
11:15	C1.2	**LexA: A novel function in *Synechocystis* sp. strain PCC 6803: Redox responsive gene expression** George Owttrim, University of Alberta
11:30	C1.3	**The Expression of Quinol Terminal Oxidases in the Cyanobacterium *Anabaena variabilis* and the Marine Centric Diatom *Thalassiosira pseudonana*** Allison McDonald, University of Toronto at Scarborough
11:45	C1.4	**A quinone redox cycle active during powdery mildew infection of wheat** David Greenshields, Dept. of Biology University of Saskatchewan
12:00	C1.5	**Significance of P700+-reduction by electron donors generated through alternative pathways in *Hypogymnia physodes* L., and *Cucumis sativus* L.** Steve Boisvert, Université du Quèbec à Trois-Rivières
12:15 -12:30	C1.6	**Towards understanding the role of PR10 proteins.** Nat Kav, University of Alberta

C2. Forest biodiversity

Room:NR1001

11:00	C2.1	**Floods, fire and ice: disturbance ecology of riparian cottonwoods** Stewart Rood, University of Lethbridge
11:15	C2.2	**Un-deserted islands: Conserving forest plant biodiversity using aggregated variable retention harvesting.** Mark Pokorski, University of New Brunswick (Saint John)
11:30	C2.3	**Spatial pattern of forest floor bryophytes in mixedwood stands of northern Alberta** Richard Caners, University of Alberta
11:45	C2.4	**Enzyme activities in rhizospheres of Douglas-fir regenerating after wildfire of differing severities** Melanie Jones, Okanagan University College
12:00	C2.5	**Tracking bryophyte community reassembly in the Acadian forest 9 years after forest harvest** Krystal Mathieson, University of New Brunswick (Saint John)
12:15 -12:30	C2.6	**Responses of understory plants to selection harvest in an Acadian forest** Zheng Fang, University of New Brunswick (Saint John)

C3. Bioproducts and alternative crops

Room: ET1013

11:00	C3.1	**Microarray analysis of flax bast fibre development** Melissa Roach, University of Alberta
11:15	C3.2	**The effects of altered carbohydrate allocation and metabolism on plant growth and cellulose yield** Heather Coleman, University of British Columbia
11:30	C3.3	**Polyhydroxybutyrate production in transgenic plants** Pornpa Suriyamongkol, University of Alberta
11:45	C3.4	**Seed yield improvement in fenugreek (*Trigonella foenum graecum* L.) using mutation breeding.** Saikat Basu, Agriculture and Agri-Food Canada, Lethbridge
12:00	C3.5	**Jerusalem artichoke development as a multipurpose crop** Sharla Eldridge, Alberta Research Council
12:15 -12:30	C3.6	**Is improvement in non-bloat causing sainfoin possible?** Surya Achara, Agriculture and Agri-Food Canada, Lethbridge

C4. Hormones in abiotic stress

Room: ET1017

11:00 C4.1 **Interactions between abscisic acid and ethylene in salt-stressed tomato roots**
Aine Plant, Simon Fraser University

11:15 C4.2 **Analysis of the Arabidopsis cell suspension phosphoproteome in response to low temperature and abscisic acid treatment**
Rami El Khatib, University of Calgary

11:30 C4.3 **Brassinosteroid functions in several abiotic stress responses of Arabidopsis thaliana and *Brassica napus***
Sateesh Kagale, University of Western Ontario

11:45 C4.4 **Morphological and hormonal studies of transgenic canola (*Brassica napus*) plants expressing the gene 1-aminocyclopropane-1 –carboxylic acid (ACC) deaminase**
Anisul Islam, University of Calgary

12:00 C4.5 **The effect of ethylene and temperature on suberin lamella deposition in the exodermis and endodermis of *Zea mays* L. roots.**
Chris Meyer, University of Waterloo

12:15 -12:30 C4.6 **Rhythm in the expression of a PIP2 gene in roots of *Pisum sativum* and its response to HgCl2 and ABA treatment imply AQPs are critical for radial water entry.**
Philip Beaudette,Trent University

C5. Diseases of horticultural species

Room: ET1018

11:00 C5.1 **Effect of 1-Methylcyclopropene treatment on postharvest decay in apple cvs. Empire and McIntosh.**
Deena Errampalli, Agriculture and Agri-Food Canada, Vineland

11:15 C5.2 **Internal Fruit Rot of Greenhouse Sweet Peppers in Alberta**
Jian Yang, Alberta Research Council

11:30 C5.3 **The Effect of Different Rates of Nitrogen and Calcium Fertilizers on Incidence and Severity of Septoria Late Blight and Celery Yield**
Cheryl Trueman, University of Guelph

11:45 C5.4 **Important Pathological Problems of *Agave tequilana* in Mexico**
Leopold Fucikovsky, Colegio de Postgraduados, Carretera México-Texcoco

12:00 C5.5 **Blueberry anthracnose in BC – weather conditions required for infection, overwintering strategy, and potential biological control of disease.**
Nitin Verma, Simon Fraser University

12:15 -12:30 C5.6 **Identification of *Botrytis mali* Ruehle existing within the Botrytis population causing fruit decay on apple in the orchards of British Columbia.**
DanO'Gorman, AAFC, Summerland

C6. Molecular aspects of pathogenesis II

Room: ET1001

11:00	C6.1	**An importin-a homolog, MOS6, is required for *snc1*-dependent constitutive disease resistance signaling in Arabidopsis.** Kristoffer Palma, University of British Columbia
11:15	C6.2	**Ubiquitination is required for plant disease resistance signaling** Sandra Goritschnig, University of British Columbia
11:30	C6.3	**Reduced Expression of Eukaryotic Translation Initiation Factor 5A2 Imparts Strong Pathogen Resistance to *Arabidopsis thaliana*** Greg Perry, University of Waterloo
11:45 -12:00	C6.4	**Genetic transformation of interspecific *Exacum* hybrids with a thaumatin-like protein (tlp) construct for enhanced fungal resistance** Faride Unda, University of British Columbia

C7. Fusarium III

Room: ET1003

11:00	C7.1	**Characteristics of *Fusarium graminearum*-positive wheat fields in Alberta, 2002 and 2003** T. Kelly Turkington, AAFC. Lacombe
11:15	C7.2	***In vitro* evaluation of *Fusarium spp.* and fusarium head blight resistance in barley at two temperatures.** Krishan Kumar, Alberta Agriculture, Food and Rural Development, Lacombe
11:30	C7.3	**Influence of soil type on incidence and severity of fusarium wilt (*Fusarium oxysporum* f. sp. *conglutinans*) of canola (*Brassica napus*) in Alberta.** Ralph Lange, Alberta Research Council
11:45	C7.4	**Relative susceptibility of cereal grains to fusarium head blight** Andy Tekauz, Agriculture and Agri-Food Canada
12:00 -12:15	C7.5	**Role of *Fusarium* species in rusty root development on ginseng roots in British Columbia** Zamir Punja, Simon Fraser University

12:30 – 2:00 **Lunch**
CSPP Business Meeting
CSA General Meeting

2:00 – 5:15 **Concurrent sessions D, E**

Special Symposium Room: ET1001

D1. Shoot apical meristems (part 1)

Chair: Bill Remphrey

2:00	D1.1	**The shoot apical meristem: an historical perspective** Taylor Steeves, University of Saskatchewan
2:30	D1.3	**The shoot apical meristem and organization of primary vascular architecture** Nancy Dengler, University of Toronto
3:00 -3:30	D1.5	**In-vitro development of shoot meristems of conifers** Mohammed Tahir, University of Manitoba

Special Symposium Room: ET1003

D2. Phytochemicals in human health research: bioactivity vs. physiological relevance (part 1)

Chair: Shahrokh Khanizadeh

2:00	D2.1	**Characterization of anthocyanins in saskatoon fruit to determined the antioxidant potential of the fruit** Jocelyn Ozga, University of Alberta
2:45 -3:30	D2.4	**Human health and medicinal plants: quality and efficacy of plant based medicines** Praveen Saxena, University of Guelph

D3. Conservation and diversity I

Room: ET1018

2:00 D3.1 **Dammed rare plants! An exploration of factors on dam reservoirs limiting survival and establishment of two Atlantic Coastal Plain flora species at risk**
Jennifer Lusk, Acadia University

2:15 D3.2 **More than one way to grow a plant: establishing viable *ex situ* populations of 3 rare and endangered plants**
Joni Kemp, Memorial University of Newfoundland

2:30 D3.3 **Assessing levels of genetic diversity in threatened populations of Western Spiderwort (*Tradescantia occidentalis*)**
Kirsten Remarchuk, University of Saskatchewan

2:45 D3.4 **Interactions between plant competition, herbivory, and abiotic stress in an arctic-alpine community**
Matthew Mitchell, University of Alberta

3:00 D3.5 **Genetic diversity of *Arabidopsis thaliana* populations alters stand characteristics and feeding by *Trichoplusia ni***
James Cahill, University of Alberta

3:15 -3:30 D3.6 **Measuring plant competition: applying yield-density models to multi-species systems.**
Peter Jolliffe,University of British Columbia

D4. Salinity stress

Room: ET2001

2:00 D4.1 **Probing the mechanism underlying PR 10-mediated enhanced germination of *B. napus* under saline conditions**
Sanjeeva Srivastava, University of Alberta

2:15 D4.2 **Functional analysis of sodium transport genes expressed with both spatial and temporal control in rice.**
Darren Plett, Australian Centre for Plant Functional Genomics

2:30 D4.3 **Changes in the physiology and metabolome of *Thellungiella salsuginea* in response to osmotic stress**
David Guevara, McMaster University

2:45 D4.4 **Ribosomal Protein S15A: Heterogeneity within the small ribosomal subunit of *Arabidopsis thaliana***
Jacqueline Hulm, University of Saskatchewan

3:00 D4.5 **Salt-responsive expression of alpha dioxygenase genes in *Arabidopsis***
Theingi Aung, Simon Fraser University

3:15 -3:30 D4.6 **The influence of salt stress on genomic stability**
Alex Boyko, University of Lethbridge

D5. Biochemistry II

Room: NR1003

2:00 D5.1 **Genetic engineering of fatty acid desaturation in *Arabidopsis thaliana* and *Brassica napus***
Saleh Shah, Alberta Research Council

2:15 D5.2 **Enhancing growth and seed yield in canola by suppression of deoxyhypuysine synthase expression via vacuum-infiltration of Agrobacterium**
Tzann-Wei Wang, University of Waterloo

2:30 D5.3 **Cloning and biochemical characterization of a novel *Arabidopsis thaliana* methyltransferase involved in phosphocholine synthesis**
Michael BeGora, McMaster University

2:45 D5.4 **Calcium and pH sensitivity of phospholipase D in tomato fruit protoplasts and sub-cellular preparations**
Krishnaraj Singh Tiwari, University of Guelph

3:00 D5.5 **Expression analysis of select *PREPHENATE DEHYDRATASE-LIKE* (*PDL*) genes in *Arabidopsis thaliana***
Susanne Kohalmi, University of Western Ontario

3:15 -3:30 D5.6 **Modulating the papain inhibitory activity of a tomato cystatin by single mutations at a positively selected amino acid site**
Dominique Michaud, Universite Laval

D6. Crop resistance to disease

Room: ET1017

2:00 D6.1 **Evidence for a second seedling leaf rust resistance gene in the Thatcher-Lr1 near-isogenic wheat line RL6003**
Brent McCallum, Agriculture and Agri-Food Canada

2:15 D6.2 **Identification of an Amplified Fragment Length Polymorphism (AFLP) marker linked to a spot blotch resistance gene in barley using bulked segregant analysis.**
Habibollah Ghazvini, CRC, AAFC

2:30 D6.3 **Influence of Rootstock, Incubation Temperature and Duration of Incubation on Bacterial Canker Severity Caused by *Pseudomonas syringae* pv. *syringae* in Peach**
Tiesen Cao, University of Alberta

2:45 D6.4 **Potential impact of a stem rust race in Kenya with wide virulence on Canadian wheat production**
Thomas Fetch Jr., Agriculture and Agri-Food Canada

3:00 D6.5 **Screening for Resistance to Tan Spot, Septoria Nodorum Blotch and Septoria tritici Blotch in Wheat, Durum and Wild Relatives.**
Pawan Singh, North Dakota State University

3:15 -3:30 D6.6 **Screening maize for resistance to common rust, eyespot, and northern leaf blight**.
Xiaoyang Zhu, Agriculture and Agri-Food Canada

D7. Pathogens of Brassica species

Room: ET1013

2:00 D7.1 **Comparison of single spore isolation techniques for *Plasmodiophora brassicae***
Shiming Xue, University of Alberta

2:15 D7.2 **Induction of systemic acquired resistance by *Leptosphaeria biglobosa* in canola to blackleg disease caused by *Leptosphaeria maculans***
Yu Chen, University of Manitoba

2:30 D7.3 **Investigating the molecular basis of resistance to the necrotrophic fungus *Alternaria brassicae***
Nidhi Sharma, University of Alberta

2:45 D7.4 **Mapping genes for resistance to *Leptosphaeria maculans* in *Brassica juncea***
Jed Christianson, University of Alberta

3:00 D7.5 **Pseudomonas chlororaphis strain190 and biological control of Blackleg in Canola: Understanding the array of potential mechanisms involved.**
Rajesh Ramarathnam, University of Manitoba

3:15 -3:30 D7.6 **Susceptibility of *Brassica napus* at different growth stages to *Leptosphaeria maculans* and its relationship to weather conditions.**
Kaveh Ghanbarnia, University of Manitoba

D8. Teaching in the plant sciences

Room: NR1001

2:00 D8.1 **Plant biology for non-believers**
David Cass, University of Alberta

2:30 D8.3 TBA

3:00 -3:30 D8.5 TBA

3:30 – 3:45 Coffee break
3:45 – 5:15 Concurrent sessions

Special Symposium Room: ET1001

E1. Shoot apical meristems (part 2)

Chair: Bill Remphrey

3:45 E1.1 **Visual modelling of shoot apical meristem development**
Przemek Prusinkiewicz, University of Calgary

4:15 E1.2 **An investigation into the mechanism of shoot bending in a clone of *Populus tremuloides* exhibiting 'crooked' architecture.**
Ashley Linden, University of Manitoba

4:30 E1.3 **Chasing the golden angle of needle trace-divergence in lodgepole pine**
Art Fredeen, University of Northern British Columbia

4:45 E1.4 **Wound-associated de novo meristem generation in Arabidopsis foliar explants.**
Steven Chatfield, University of Guelph

5:00 E1.5 **Determination of the shoot apical meristem in microspore derived**
5:15 **embryos of *Brassica napus* cv Topaz**
Mark Belmonte, University of Manitoba

Special Symposium Room: ET1003

E2. Phytochemicals in human health research: bioactivity vs. physiological relevance (part 2)

Chair: Shahrokh Khanizadeh

3:45 E2.1 **Designer Fruits with Enriched Phytochemicals for Human Health**
Rong Tsao Agriculture and Agri-Food Canada, University of Guelph

4:30 E2.3 **Plants and Neurochemistry**
Susan Murch, University of Guelph and Institute for Ethnomedicine, Provo, UT

E3. Conservation and diversity II

Room: ET1018

3:45 E3.1 **Parasitic-host plant interaction as a mechanism facilitating horizontal transfer of mitochondrial genes in land plants**
Sasa Stefanovic, University of Toronto at Mississauga

4:00 E3.2 **Phylogeographic genetic analysis of Pitcher's thistle (*Cirsium pitcheri*)**
Martha Coleman, Trent University

4:15 E3.3 **Characterizing plant biodiversity in naturalized pastures of the maritimes**
Yousef Papadopoulos, Agriculture and Agri-Food Canada, Truro

4:30 E3.4 **Horizontal and vertical seed banks from Ritchie's prisere on the Churchill River Estuary, MB**
Richard Staniforth, University of Winnipeg

4:45 E3.5 **Geographic distribution of chloroplast DNA haplotypes of *Oxyria digyna* (Polygonaceae) and implications for the postglacial history of western Canada.**
Geraldine Allen,University of Victoria

5:00 E3.6 **Co-occurrence of rare vascular plants in the Northern Rocky Mountains of Alberta**
Joyce Gould, University of Alberta

E4. Rhizosphere

Room: ET1013

3:45 E4.1 **Impacts of fertilization on fine roots and ectomycorrhizas under young lodgepole pine and interior spruce stands in the interior of British Columbia**
Shannon Berch, British Columbia Ministry of Forests

4:00 E4.2 **A comparison of fungal communities associated with nodules and roots of gray alder (*Alnus incana*) and the roots of paper birch (*Betula papyrifera*)**
Sam Skinner, University of Alberta

4:15 E4.3 **Interaction between wheat cultivar and bacterial isolate for rhizosphere colonization and root accumulation of Pseudomonas-derived 2,4-diacetylphloroglucinol**
Patricia Okubara, USDA ARS

4:30 E4.4 **Combined effects of elevated atmospheric CO2 and rhizobial strains on nitrogen fixation and cold acclimation in alfalfa**
Annick Bertrand,AAFC, Ste.Foy

4:45 E4.5 **Multiple isolates of species of Monodictys from the roots of *Saxifraga oppositifolia* from the Canadian High Arctic**
Melissa Day, University of Alberta

5:00 E4.6 **Microorganisms stimulating growth and development of greenhouse tomato plants**
Valerie Gravel, Université Laval

E5. Reactive oxygen

Room: ET2001

3:45	E5.1	**Visualizing reactive oxygen species formation in plant cells treated with stressors** Daryl Enstone, University of Waterloo
4:00	E5.2	**Potential role of thiol-disufide oxidoreductase-based antioxidant system in aluminum (Al) and cadmium (Cd) resistance** Diana Lopez-Santiago, University of Alberta
4:15	E5.3	**Class-1 hemoglobin and antioxidant metabolism in alfalfa roots** Abir Igamberdiev, University of Manitoba
4:30 -4:45	E5.4	**Characterization of the alternative oxidase of *Chlamydomonas reinhardtii* in response to nitrogen source and oxidative stress** Wenze Li, University of Western Ontario

E6. Pathogen detection

Room: ET1017

3:45	E6.1	**A PCR approach to detecting bacterial blight organisms in an epidemiological study in dry bean fields in southern Alberta** Michael Harding, Alberta Agriculture, Food & Rural Development
4:00	E6.2	**Refinement of PCR-based methods for detecting and quantifying the bacterial spot pathogen on seeds and transplant seedlings** Diane Cuppels, AAFC, London
4:15	E6.3	**Discrimination of carrot diseases in storage using headspace volatile metabolite profiles** Appanna Vikram, McGill University
4:30	E6.4	**Characterization of *Plasmodiophora brassicae* populations from Alberta, Canada** Stephen Strelkov, University of Alberta
4:45	E6.5	**The development of multiplex real-time PCR to monitor biological control agents in the orchard** Won-Sik Kim, AAFC, Vineland
5:00 -5:15	E6.6	**Double antibody sandwich enzyme-linked immunosorbent assay testing for detection plum pox potyvirus using polyclonal antibodies.** Deena Errampalli, AAFC, Vineland

E7. Teaching in the plant sciences (cont'd)

Room: NR1001

3:45 -5:15 **Roundtable discussion**

Title: "What do biology students need to know about botany?"

Panelists: Hugues Massicotte, UNBC
Marie Davey, University of Alberta
Cindy Graham, University of Calgary
Frederique Guinel, Wilfrid Laurier University

Moderators: Heather Addy, University of Calgary
Randy Currah, University of Alberta

5:15 – 7:30 Supper break

7:30 – 9:00PM CSPP Gold Medal Symposium ET 2001

My long journey in plant research: contributions to flavonoid biochemistry
Ragai Ibrahim, Concordia University

7:30 – 9:00PM CBA Luella K. Weresub Lecture ET 2002

Rust fungi and a rusty mycologist: The morphology, life cycle and taxonomy of rust fungi
Yasu Hiratsuka, Canadian Forest Service, Edmonton

Overview: Friday, June 17

8:00 AM	Registration desk opens.	
	Continental breakfast. Free to all conference participants.	HOR
8:30-10:30	**Plenary Session III** Plants, Canada and Climate Change	HOR
10:30	Posters and exhibits open for viewing.	
10:30-11:00	Break. Coffee and other beverages provided.	
11:00-12:15	**Special Plenary Lecture: David Suzuki** The challenge of the 21st century: setting the real bottom line	HOR
12:15	Book signing: David Suzuki, University of Alberta Bookstore	
12:15 - 2:00	Lunch. No food provided by conference organizers, except for meeting participants	
12:15 – 1:45	Open session with NSERC Program Officers. Box lunches provided.	ET1003
12:15 – 1:45	Plant Canada Board Meeting. Lunch provided.	SUB4-20
2:00	Registration desk closes permanently	
2:00	Posters and exhibits close permanently All posters must be removed before 3:00PM	
2:00-6:00	Excursions. Meet in SUB lobby (SUB Stage) for transportation.	
7:00-9:00PM	Open session: Opening the 'model tree' tool-box: A discussion of new opportunities and challenges for poplar research	NR2090
7:30-9:00PM	Canadian Journal of Botany executive meeting	

Sessions detail: Friday, June 17

8:00 Continental breakfast
8:30 – 10:30 Plenary session

Plenary Session — Room: Horowitz

III. Plants, Canada and Climate Change

8:30 III.1 **Biosphere solutions to climate change and clean energy: New challenges and opportunities for plant science research**
David Layzell, Queen's University / BIOCAP

9:10 III.2 **Effects of climate on yield of five crops in family *Brassicacae* in southern Ontario**
Alan McKeown, University of Guelph

9:30 III.3 **Climate change and northern hardwood forests: key messages from the Aspen Free Air Carbon Dioxide Enrichment (FACE) Experiment**
Kevin Percy, CFS Fredericton

10:00 III.4 **Plant genetic and evolutionary responses to ecological crisis in Canada**
Quentin Cronk, University of British Columbia

10:30 – 11:00 Coffee break

11:00 – 12:15 Special lecture

The challenge of the 21st century: setting the real bottom line — Room: Horowitz
David Suzuki, David Suzuki Foundation

12:15 – 2:00 Lunch break
12:15 – 1:45 Plant Canada Board Meeting
12:15 – 1:45 Open session with NSERC Program Officers

2:00 – 6:00 Excursions

7:00 – 9:00PM Open discussion

Opening the 'model tree' tool-box: A discussion of new opportunities and challenges for poplar research
Brian Ellis, University of British Columbia

Overview: Saturday, June 18

8:00AM	Continental breakfast. Free to all conference participants.	ETLC
8:30-10:00	**Concurrent Sessions Block F**	
	F1. Resistance to diseases: Unraveling the plant responses (CPS symposium part 1)	ET1001
	F2. Arctic & Alpine Plants (CBA symposium, part 1)	ET1003
	F3. Molecular biology of hormone responses	ET1013
	F4. Natural products I	ET1017
	F5. Soil and forage management	ET1018
	F6. Carbon flows	ET2001
	F7. Tree physiology	ET2002
10:00-10:15	Break. Coffee and other beverages provided.	ETLC
10:15-11:45	**Concurrent Sessions Block G**	
	G1. Resistance to diseases: Unraveling the plant responses (CPS symposium part 2)	ET1001
	G2. Arctic & Alpine Plants (CBA symposium, part 2)	ET1003
	G3. Photosynthesis and light responses	ET1013
	G4. Natural products II	ET1017
	G5. Breeding and crop improvement	ET1018
	G6. Phytoremediation and xenobiotics	ET2001
	G7. Cell biology	ET2002
12:00 - 2:00	Lunch. No food provided by conference organizers, except for meeting participants	
12:00 - 2:00	CBA Annual General Meeting. Box lunches provided.	ET1003
12:00 - 2:00	CSHS Annual General Meeting. Box lunches provided.	ET1001
12:00 - 2:00	CPS Awards Luncheon	Lister Dining
12:00 - 2:00	CSPP Incoming Executive Meeting. Lunch provided.	SUB420
2:00-3:30	**Concurrent Sessions Block H**	
	H1. Horticulture and greenhouse crops	ET1001
	H2. Cold stress	ET1003
	H3. Mycology	ET1018
	H4. Tree genomics and herbivory	ET1013
	H5. Pathogen resistance mechanisms	ET1017
	H6. Crop management and disease	ET2001
3:30-3:45	Break. No food provided by conference organizers	
3:45-5:15	CPS Business Meeting	ET1018
3:45-5:15	**Concurrent Sessions Block J**	
	J1. Agronomy	ET1001
	J2. Physiology and environment	ET1003
	J3. Tissue culture	ET1013
	J4. Seeds	ET1017
6:00PM -11:00PM	**Closing Banquet.** Meet in SUB lobby for transportation.	Ft. Edm

Session details: Saturday, June 18

8:00 Continental breakfast
8:30 – 11:45 Concurrent sessions F,G

Special Symposium Room: ET1001

F1. Resistance to diseases: Unraveling the plant responses (part 1)

Chair: André Laroche

8:30 F1.1 **Systemic Acquired Resistance in Tobacco and Arabidopsis**
John Draper, University of Wales

9:15 -10:00 F1.4 **Whirly: a new family of plant transcription factors involved in defence responses**
Normand Brisson, Université de Montréal

Special Symposium Room: ET1003

F2. Arctic and alpine plants

Chair: Mark Dale

8:30 F2.1 **Pattern and process at the alpine treeline**
David M. Cairns, Texas A&M University, College Station TX

9:00 F2.3 **Top-down vs. Bottom-up regulation of vegetation in a Boreal Forest understorey**
Roy Turkington, University of British Columbia

9:30 -10:00 F2.5 **Climate warming and the (in)stability of plant-herbivore interactions in subarctic alpine meadows.**
David Hik, University of Alberta

F3. Hormone signals

Room: ET1013

8:30	F3.1	**Control of Arabidopsis transition to flowering: A novel route involving ABA binding to FCA** Fawzi A. Razem, University of Manitoba
8:45	F3.2	**Characterizing cytokinin oxidase (CKX) throughout the development of R50 (sym 16), a pea mutant accumulating cytokinins** Mark Held, Wilfrid Laurier University
9:00	F3.3	**A gain-of-function mutant of *AtMKK9* affects multiple hormone signals in Arabidopsis.** Corinne Cluis, University of British Columbia
9:15	F3.4	**Cytokinin-induced changes in gene expression and epigenetic inheritance in Arabidopsis** Donna Lindsay, University of Saskatchewan
9:30	F3.5	**Developmental regulation of the GA biosynthesis genes, GA20ox, GA3ox and GA2ox during germination and young seedling growth of pea (*Pisum sativum* L.)** Belay Ayele, University of Alberta
9:45 -10:00	F3.6	**Functional analysis of GAMYB gene in rice** S. M. Shahinul Islam, Niigata University

F4. Natural products I

Room: ET1017

8:30	F4.1	**Functional analysis of *EgMyb1*, a *Eucalyptus* R2R3-Myb gene involved in the regulation of phenylpropanoïd metabolism** Sylvain Legay, Université Laval
8:45	F4.2	**Use of an array to assay phenylpropanoid pathway expression in developing bean (*Phaseolus vulgaris* L.) seeds** K. Peter Pauls, University of Guelph
9:00	F4.3	**Selective cytotoxicity of red grape wine polyphenols against MCF-7 breast cancer cells** Gopinadhan Paliyath, University of Guelph
9:15	F4.4	**Evaluation of antioxidant and anti-proliferative properties of polyphenols from novel grape lines** Jissy Jacob, University of Guelph
9:30 -9:45	F4.5	**Plant ?-glutamyl hydrolases and folate polyglutamates: characterization, compartmentation, and co-occurrence in vacuoles** Andrew Hanson, University of Florida

F5. Soil and forage management

Room: ET1018

8:30 F5.1 **Spatial and temporal variability of crop yield and soil fertility in relation to landscape**
Alan Moulin, AAFC

8:45 F5.2 **The effect of nitrogen source, rate and season of application on forage yield, protein content and nitrogen uptake**
Rigas Karamanos,Western Cooperative Fertilizers Limited

9:00 F5.3 **Influence of forage management and species on soil mineral nitrogen supply rates and seasonal dynamics**
Vern Baron, Agriculture and Agri-Food Canada, Lacombe

9:15 F5.4 **Forage yield and persistence of three short-lived perennial grasses in monoculture and mixture with alfalfa at a semiarid location in southern Saskatchewan.**
Paul Jefferson, Agriculture and Agri-Food Canada, Swift Current

9:30 -9:45 F5.5 **Evaluation of the Illinois Soil Nitrogen Test for Estimating Potentially Mineralizable Soil Nitrogen**
Karl Slawinski,University of Manitoba

F6. Carbon flow

Room: ET2001

8:30 F6.1 **For peat's sake: The role of fungi in peatland carbon dynamics**
Markus N. Thormann,Canadian Forest Service, Edmonton

8:45 F6.2 **The Sensitivity of Douglas-fir (*Pseudotsuga menziesii* Mirb.) Root Respiration to Seasonal Temperature Change in Three Age Classes of Coastal Forest**
Tyler Abbey,University of British Columbia

9:00 F6.3 **The role of common mycorrhizal networks in belowground carbon flow between plant neighbours**
Leanne Philip, University of British Columbia

9:15 F6.4 **Uptake of inorganic carbon by the acidophilic green alga *Chlamydomonas acidophila***
Brian Colman, York University

9:30 F6.5 **Non-photosynthetic carbon metabolic flux in *Chlamydomonas reinhardtii*: 13C-isotopomer distribution ratios of proteinogenic amino acids**
Hank Bestman, King's University College

9:45 -10:00 F6.6 **Grazing and topographic effect on short-term litter decomposition in a natural rangeland**.
Johnny Montenegro-Ballestero, University of Alberta

F7. Tree physiology

Room: ET2002

8:30 F7.1 **Are hybrid poplar clones commonly grown in the Canadian Prairies adaptable to soil moisture deficit?**
Bobbi Nash, University of Saskatchewan

8:45 F7.2 **Phytohormonal regulation of transpiration(E) and stomatal conductance (Gs) among canopy layers of a mature Sugar maple (*Acer saccharum*) stand.**
Ian Reeves, Trent University

9:00 F7.3 **Growth and water transport in mycorrhizal seedlings of *Pinus banksiana* and *Picea glauca* treated with NaCl**
Virginia Jacob-Cervantes, University of Alberta

9:15 F7.4 **Carbon Isotope Discrimination provides a powerful option for the identification of high Water Use Efficient Cashew (*Anacardium occidentale* L.) clones for crop improvement**
S Raju, University of British Columbia

9:30 -9:45 F7.5 **Functional analyses exploring the impact of high nitrogen availability on wood formation in *Populus***
Frederic Pitre, Université Laval

10:00 – 10:15 Coffee break
10:15 – 11:45 Concurrent sessions G

Special Symposium Room: ET1001

G1. Resistance to diseases: Unraveling the plant responses (part 2)

Chair: André Laroche

10:15 G1.1 **An alternative agriculture approach opens a new door for enabling sustainable disease tolerance in crops**
Autar K. Mattoo, USDA-ARS, Beltsville MD

11:00 G1.4 **High frequency of rearrangements in R-gene loci of the progeny of tobacco plants exposed to compatible virus**
Igor Kovalchuk, University of Lethbridge

11:15 G1.5 **Methyl-jasmonate and salacylic acid regulate expression of defence-related genes and reduce infection by common bunt in wheat (*Triticum aestivum* L.)**
Denis Gaudet, AAFC, Lethbridge

11:30 -11:45 G1.6 **Bacterial Cyclic b-(1, 2) Glucan acts in systemic suppression of Plant Defense Responses**
Kamal Bouarab, Université de Sherbrooke

Special Symposium Room: ET1003

G2. Arctic and alpine plants (part 2)

Chair: Geraldine Allen

10:15 G2.1 **Evolution of arctic species of bluegrass (*Poa*)**
Lynn Gillespie, Canadian Museum of Nature

10:45 G2.3 **Migration and Speciation in Moonwort Ferns (*Botrychium* subgenus *Botrychium*) in North America.**
Donald Farrar, Iowa State University, Ames, IA

11:15 -11:45 G2.5 **Haplotype diversity patterns among arctic and alpine *Packera* species: species level promiscuity and the heartbreak of hybridization.**
John Bain, University of Lethbridge

G3. Photosynthesis and light responses

Room: ET1013

10:15 G3.1 **A comparison of methods to measure photosynthesis in field-grown soybean**
Malcolm Morrison, Agriculture and AgriFood Canada, Ottawa

10:30 G3.2 **Deactivation of excess excitation energy in plants exposed to low light and chilling stress**
David Joly, Universite du Quèbec à Trois-Rivières

10:45 G3.3 **Reversal in foliar orientation from normal by reverse application of phototropin (s)/ resultants in *Epipremnum aureum* or Sakina Akhter effect in *E.aureum***
Sheikh Babar,

11:00 G3.4 **The role of ethylene in phytochrome – cryptochrome cross-talk in Arabidopsis seedlings grown under low and high R/FR ratios with equal blue light irradiance**
Linda Walton, University of Calgary

11:15 -11:30 G3.5 **Uncoupling the effect of light quality from light irradiance on development of *Helianthus annuus* hypocotyls: Changes in gibberellin (GA) and indole-3-acetic acid (IAA) levels and in ethylene evolution.**
Leonid Kurepin, University of Calgary

G4. Natural products II

Room: ET1017

10:15 G4.1 **Bioprospecting the wild blueberry: impact of nutrient management practices and biproduct utilization**
David Percival, Nova Scotia Agricultural College

10:30 G4.2 **Genetic transformation of Bee Balm (*Monarda didyma*)**
Soheil Mahmoud, OUC

10:45 G4.3 **Modification of the carotenoid pathway in carrot roots to produce novel ketocarotenoids**
Jayaraj Jayaraman, Simon Fraser University

11:00 G4.4 **The relationship among morphological and anatomical features and technical ethnobotanical uses of the common cattail (*Typha latifolia* L.).**
Laura Hoskin, University of Northern BC

11:15 G4.5 **Phytochemical and agronomic variations in *Echinacea pallida* var. *angustifolia* plants from cultivated populations in British Columbia and Washington grown under common greenhouse conditions.**
Alain Boucher, University of British Columbia

11:30 -11:45 G4.6 **Incorporation of molecular and chemical methodologies as a reliable approach for Echinacea species identification**
Hameed Khadair, University of Alberta

G5. Breeding and crop improvement

Room: ET1018

10:15 G5.1 **Characterization in Hard Spring wheat and phenotypic analysis in DH population RL4452/AC Domain for seed dormancy and pre-harvest sprouting resistance**
Golam Rasul, AAFC, Winnipeg

10:30 G5.2 **Introgression between *Sinapis alba* (Yellow mustard) and *Brassica napus* (Oil-seed rape) as a potential source for genetic enhancement and crop improvement**
Wajahatullah Khan, AAFC, Saskatoon

10:45 G5.3 **Optimum Growth Conditions for the Selection of dual-purpose flax in the CDC Breeding Program**
Alison Burton, University of Saskatchewan

11:00 -11:15 G5.4 **QTL mapping of coleoptile length, root length, and seed vigor index in wheat under normal and osmotic stress conditions**
Mohsen Mohammadi, University of Alberta

G6. Phytoremediation and xenobiotics

Room: ET2001

10:15 G6.1 **Identification of plant species suitable for phytoremediation of hydrocarbon/metal contaminated soils.**
Jan J. Slaski, Alberta Research Council

10:30 G6.2 **Effects of paper mill sludge on growth of plants in mine tailings**
Scott Green, University of Manitoba

10:45 G6.3 **Does addition of crude oil to sub-boreal forest mini-ecosystems alter established ectomycorrhizal communities?**
Susan Robertson, University of Northern British Coulmbia

11:00 G6.4 **Phytoremediation of nickel contaminated soils using sunflower (*Helianthus annuus* L.) colonized by arbuscular mycorrhizal fungi**
Keomany Ker, University of Ottawa

11:15 G6.5 **Phytoremediation: Effects of AM colonization in 'wild' Tobacco plants grown in Zn-contaminated soils**
Patrick Audet, University of Ottawa

11:30 -11:45 G6.6 **The role of sclerids in the Cyperaceae: Relation to plant habit**
Sarah Bogart, Nipissing University

G7. Cell biology

Room: ET2002

10:15	G7.1	**Molecular control of nuclear and subnuclear targeting of the plant CDK inhibitor ICK1** Larry Fowke, University of Saskatchewan
10:30	G7.2	**Looking beyond the cytosol: Housekeeping enzymes targeted to multiple subcellular compartments in *Arabidopsis thaliana*** SM Schoor, University of Waterloo
10:45	G7.3	**Determination of cis-acting regulatory elements of Arabidopsis ribosomal protein genes ATRPL23A-1 and -2** Rory Degenhardt, University of Saskatchwan
11:00	G7.4	**Compartmentation of de novo NADP(H) biosynthesis: Subcellular and Tissue Distribution of NAD(H) Kinases in *Arabidopsis thaliana*** Jeffrey Waller, Queen's University Biology Department
11:15	G7.5	**Characterization of peroxisomal malate dehydrogenase; a microtubule binding protein.** Michelle Freeman, University of Calgary
11:30 -11:45	G7.6	**An efficient, new selectable marker for the selection of transgenic plants** Sima Babayeva, Institut de recherche en biologie végétale, Université de Montréal

12:00 – 2:00 **Lunch**
CBA Annual General Meeting
CSHS Annual General Meeting
CPS Awards Luncheon

12:00 – 2:00 **CSPP Incoming Executive Meeting**

2:00 – 5:15 **Concurrent sessions H, J**

H1. Horticulture and greenhouse crops

Room: ET1001

2:00	H1.1	**An overview of Northern Vigour® research in horticultural crops** Tanino, K.K.,University of Saskatchewan
2:15	H1.2	**Aquaponics: A novel greenhouse production system and its challenges** Vipan Bansal, Crop Diversification Centre South, AAFRD, Brooks
2:30	H1.3	**Modeling CO_2 emission scenarios in greenhouse tomato production** Diane Edwards,University of British Columbia
2:45	H1.4	**N fertilization and rib discoloration incidence in crisphead lettuce** Sylvie Jenni, Agriculture Canada, St-Jean-sur-Richelieu
3:00	H1.5	**Breeding Strawberries Rich in Antioxidant Phenolic Compounds** Shahrokh Khanizadeh, Agriculture and Agri-Food Canada
3:15 -3:30	H1.6	**Automated monitoring of greenhouse plants** David Ehret, Pacific Agri-Food Research Centre, Agassiz BC

H2. Cold stress

Room: ET1003

2:00 H2.1 **Transcript Profiling in Arabidopsis thaliana using Serial Analysis of Gene Expression (SAGE)**
Steve Robinson, Agriculture Canada

2:15 H2.2 **Differential enzyme activities in *Brassica napus* cell suspension cultures exposed to low temprature and osmotic stresses.**
Yongzhong Shi, University of Alberta

2:30 H2.3 **Changes in gene expression in microspore-derived cell suspension cultures of *Brassica napus* exposed to low temperature or osmotic stress**
Chris Kazala, University of Alberta

2:45 H2.4 **Characterization of low temperature regulated genes using microarray analysis and real-time PCR in winter wheat (*Triticum aestivum*)**
Denise Nilsson, Agriculture and Agrifood Canada

3:00 H2.5 **Relationship between genetic polymorphism and cold tolerance in alfalfa**
Yves Castonguay, Agriculture and Agri-Food Canada, Ste-Foy

3:15 H2.6 **Winter Rye Glucanases Play a Dual Role in the Cold**
-3:30 Mahmoud Yaish, University of Waterloo

H3. Mycology

Room: ET1018

2:00 H3.1 **Characterization of fungal root endophytes allied to the ericoid mycobiont *Rhizoscyphus ericae* (= *Hymenoscyphus ericae*) and to the *Piceirhiza bicolorata* ectomycorrhizal morphotype**
Lynne Sigler, University of Alberta

2:15 H3.2 **Arthropod dispersal of cycloheximide resistant fungi in central Alberta.**
Matthew Greif, University of Alberta

2:30 H3.3 **First report of the teleomorph of *Colletotrichum truncatum***
-2:45 Cheryl Armstrong-Cho, University of Saskatchewan

H4. Tree genomics and herbivory

Room: ET1013

2:00 H4.1 **The Arborea Project: A functional genomics approach to identifying regulators of wood formation in conifers**
Janice Cooke, Arborea Project

2:15 H4.2 **Investigation of the role of genes encoding LIM-only proteins in poplar wood formation**
Florian Lafarguette, Université Laval

2:30 H4.3 **Genomics discovery of lodgepole pine defense mechanisms against the mountain pine beetle-vectored fungal pathogen (*Ophiostoma clavigerum*) and evaluation of a new conifer microarray chip for gene expression profiling across the pine family.**
Natalia Kolosova, University of British Columbia

2:45 H4.4 **Exploring large-scale transcriptional responses in hybrid poplar to herbivory by forest tent caterpillars: Genome organization, cDNA cloning and gene expression of a Kunitz trypsin inhibitor family**
Steven Ralph, University of British Columbia

3:00 -3:15 H4.5 **Traumatic resinosis in Sitka spruce – a resistance mechanism against attack by the white pine weevil?**
Farhad Ghavami, Simon Fraser University

H5. Pathogen resistance mechanisms

Room: ET1017

2:00	H5.1	**A proteomic analysis of resistance to *Erysiphe pisi* in mutants of *Pisum sativum* × *P. fulvum*** Roger Zhang, Alberta Research Council
2:15	H5.2	**Altered expression of proteins caused by Fusarium head blight infection in spikes of wheat cultivars with various levels of resistance** Wenchun Zhou, AAFC
2:30	H5.3	**Characterization of phenolic compounds in two pine species resistant to the European race of *Gremmeniella abietina*, the causal agent of scleroderris canker** Danny Rioux, Natural Resources Canada, Ste-Foy
2:45 -3:00	H5.4	**High throughput analysis of gene expression in snow mold resistant winter wheat using microarray analysis.** Carolyn Peniket, AAFC, Lethbridge

H6. Crop management and disease

Room: ET2001

2:00	H6.1	**Control of Sclerotinia Head Rot in Sunflower** Khalid Rashid, AAFC, Morden
2:15	H6.2	**Impact of four types of lime on potato scab incidence in eight potato soils** Kenneth Conn, AAFC, London
2:30	H6.3	**Effect of crop rotation on severity of red root rot and grain yield in corn** Stéphan Pouleur, Agriculture et Agroalimentaire Canada, Ste-Foy
2:45 -3:00	H6.4	**Effect of fish emulsion pre-plant soil amendment on common scab of potato and tuber yield in eight commercial potato field soils** Pervaiz Abbasi, Agriculture and Agri-Food Canada, London

3:30 – 3:45 **Coffee break**
3:45 – 5:15 **Concurrent sessions J**
CPS business meeting

J1. Agronomy

Room: ET1001

3:45 J1.1 **Forage yield and quality of traditional forage legumes under acid soil conditions**
Surya Acharya, AAFC, Lethbridge

4:00 J1.2 **Measuring phyllochrons in barley to use for seeding date recommendations**
Patricia Juskiw, Field Crop Development Centre,Lacombe AB

4:15 J1.3 **Role of crop residue in mitigating frost damage in early sown canola**
Karl Volkmar, AAFC, Brandon

4:30 J1.4 **Economic Impacts of Tillage, Phosphorus, and Preceding Crops on Flax**
Mohammad Khakbazan, AAFC, Brandon

4:45 -5:00 J1.5 **Spatial Variations of Wild Blueberry Leaf Chlorophyll and Nitrogen Levels Using Hyperspectral Techniques**
Camille Bourguignon, Nova Scotia Agricultural College, Truro

J2. Physiology and environment

Room:ET1003

3:45 J2.1 **Detection of source-sink imbalances by UV-induced fluorescence in sunflower plants**
Guy Samson, Université du Québec à Trois-Rivières

4:00 J2.2 **Combined effects of ultraviolet-B radiation and CO2 on Brassica napus**
Mirwais Qaderi, University of Calgary

4:15 J2.3 **Carbon isotope discrimination as a selection criterion for improved water use efficiency and productivity of barley on the prairies**
Anthony Anyia, Alberta Research Council

4:30 J2.4 **Physiological responses and dry mass partitioning in three ecotypes of Stellaria longipes grown at elevated CO2.**
Gillian Donald, University of Calgary

4:45 J2.5 **Study of cold acclimation and freezing tolerance in different species of Brassicaceae.**
Nirmala Sharma, University of Saskatchewan

5:00 -5:15 J2.6 **Comparative gene expression analysis during cold acclimation in winter and spring wheat.**
Patrick Gulick, Concordia University

J3. Tissue culture

Room: ET1013

3:45	J3.1	**Isolated microspore culture of Canadian 6x triticale cultivars.** Eric Amundsen, Agriculture and Agri-Food Canada, Lethbridge
4:00	J3.2	**Isolation, culture and genetic engineering of triticale protoplasts** Archana Chugh, Agriculture and Agri-Food Canada, Lethbridge
4:15	J3.3	**Overcoming TDZ-induced inhibition of adventitious shoot elongation and developing a one-step in vitro cloning procedure using zeatin in strawberry.** Samir Debnath,Agriculture and Agri-Food Canada, St.John's
4:30	J3.4	**Genes involved in microspore embryogenesis in *Brassica napus* microspore culture identified by differential display using an Arabidopsis microarray.** John Chan, University of Guelph
4:45	J3.5	**Expression of the plant CDK inhibitor ICK1 leads to cell death in Arabidopsis microspores and pollen** Hong Wang,University of Saskatchewan
5:00 -5:15	J3.6	**Alternation of morphology and cation contents in bamboo xylem mutant, *vse*, derived from somaclonal variation** Choun-Sea Lin, China Institute of Technology

J4. Seeds

Room: ET1017

3:45	J4.1	**An ABA receptor and its docking protein: Potential involvement in ABA signal transduction related to seed dormancy.** Ashraf El-Kereamy, University of Manitoba
4:00	J4.2	**A model for seed dormancy regulation mediated by the ethylene receptors in Arabidopsis thaliana** Graham Thurston, Carleton University
4:15	J4.3	**Abscisic acid signaling networks in seed storage product accumulation in microspore-derived embryos of *Brassica napus* using a targeted cDNA microarray.** Musrur Rahman, National Research Council
4:30	J4.4	**Proteomics of embryo and endosperm during seed germination in tomato** Vipen Sawhney, University of Saskatchewan
4:45	J4.5	**A soybean seed protein with carboxylate-binding activity** Sangeeta Dhaubhadel, Agriculture and Agri-Food Canada, London
5:00 -5:15	J4.6	**Isolation and characterization of the arginase promoters in loblolly pine (*Pinus taeda* L.) and Arabidopsis** Disa Brownfield, University of Alberta

ABSTRACTS

Plenary session I: Roots and the underground evironment

I.1 A systems approach to understanding root development

PHILIP N. BENFEY, JI-YOUNG LEE, JULIETTE COLINAS, HONGCHANG CUI, TEVA VERNOUX, MITCH LEVESQUE
Biology Dept, Duke Univ, Durham, NC

Most eukaryotic development begins with a fertilized egg, which undergoes a series of cell divisions to generate a multicellular organism in which the diverse cell types function in harmony. Central processes in development include creating distinctions between cells and producing coordination among different cells so that they function as units. In plants, both processes have been shown to rely heavily on cell-to-cell communication and activation and/or repression of subsets of genes. Signals that pass between cells frequently result in the activation of signaling pathways within cells. These signals can result in modifications of transcription factors, which bind to specific sites on the DNA causing the activation or repression of genes. The products of the activated genes are then synthesized. Among them are transcription factors that bind other DNA sequences, activating or repressing a new set of genes. The term transcriptional networks is used for the sequential activity of transcription factors that activate or repress the expression of other transcription factors responsible for developmental regulation.

While signaling and transcription are equally important for development, high through-put techniques for identifying the nodes and links in transcriptional networks have matured more rapidly. Specifically, the critical datasets include global expression profiles at cell-type specific resolution and direct binding data for identification of targets of transcription factors expressed in the cells of interest. For plants the simplifying aspects of development in an organ such as the root, make it highly tractable for the application of these approaches.

The Arabidopsis root develops continuously from four sets of stem cells in its tip. These stem cells (or initials) divide asymmetrically to regenerate themselves and produce a daughter cell, which in turn divides asymmetrically to generate the first cells of each of the root lineages. Because plant cells don't move, these cell lineages are constrained in cell files. Thus, in the root, each stage of development is found in a specific set of cells along the longitudinal axis, with the youngest cells in each file being closest to the initials. The other simplifying aspect of root development is that, at least for the four outer layers of cells, the root can be viewed as a radially symmetric cylinder.

With the goal of identifying the transcriptional networks that regulate plant development we have developed a method to determine global expression patterns at cell-type specific regulation. We have generated a map of gene expression for many of the cell types including the stem cells in the root of Arabidopsis. From this dataset we have identified transcription factors with tissue-specific expression in the root. We have made transcriptional and translational fusions of these transcription factors to GFP and introduced them into plants. The resulting expression patterns are being analyzed to improve the expression map, identify regulatory regions responsible for tissue-specific expression, and determine when transcription factors move intercellularly. In a complementary project we are using convergent genomics approaches to identify the targets of transcription factors that control stem cell identity and radial patterning.

I.2 The role of arbuscular mycorrhizal fungi in terrestrial communities and ecosystems

JOHN KLIRONOMOS
Univ of Guelph, Dept of Integrative Biology, Guelph, ON N1G 2W1.

Arbuscular mycorrhizal fungi are common in terrestrial ecosystems around the world. They form symbiotic associations with the vast majority of terrestrial plants, and it has long been recognized that they can stimulate the growth of individual plants. They do this by providing their host plants with greater access to limiting nutrients and protection from pathogens. However, the role of these fungi on the structuring of plant communities and the functioning of ecosystems has received much less attention. Here, I will present a number of studies of arbuscular mycorrhizal fungi and their influence on plant co-existence, plant diversity, plant community succession, nutrient cycling, and ecosystem productivity. Overall, these fungi can have immense influence on the structure and functioning of terrestrial communities and ecosystems. As a result they

should be considered in ecosystem management and in plans for rehabilitating and restoring degraded ecosystems.

I.3 Invisible ecosystems

SCOTT WILSON
Univ of Regina

Roots are the dominant component of cold and arid ecosystems such as tundra and grasslands. Recent studies at the northern edge of the Great Plains show that temperate forests are also dominated by belowground interactions. As a result, belowground processes are key to understanding patterns of diversity and responses to global change in many Canadian ecosystems. For example, differences between native and invasive grasses can significantly decrease ecosystem carbon sequestration as invasion proceeds. Knowledge about below ground interactions can, in turn, be used to manage invasions.

Plenary session II
Global Food Security and Biotechnology

II.1 Global Food Security and Biotechnology

PROF M S SWAMINATHAN
Chairman, National Commission on Farmers, Govt. of India
President, National Academy of Agricultural Sciences, India

1. Agriculture in the developing world is generally characterised by small holdings operated by resource poor farmers. The smaller the farm, the greater is the need for marketable surplus in order to get some cash income. Also land is a shrinking resource for agriculture and small farmers will have to produce more food and other products from diminishing per capita arable land and irrigation water availability. However productivity improvement will have to be achieved without unfavourable ecological or social consequences, as explained by M S Swaminathan in his Presidential Address to the Agriculture Section of the Indian Science Congress at Varanasi on 4 January 1968. The relevant quotation is given below.

> "Exploitative agriculture offers great dangers if carried out with only an immediate profit or production motive. The emerging exploitative farming community in India should become aware of this. Intensive cultivation of land without conservation of soil fertility and soil structure would lead, ultimately, to the springing up of deserts. Irrigation without arrangements for drainage would result in soils getting alkaline or saline. Indiscriminate use of pesticides, fungicides and herbicides could cause adverse changes in biological balance as well as lead to an increase in the incidence of cancer and other diseases, through the toxic residues present in the grains or other edible parts. Unscientific tapping of underground water will lead to the rapid exhaustion of this wonderful capital resource left to us through ages of natural farming. The rapid replacement of numerous locally adapted varieties with one or two high-yielding strains in large contiguous areas would result in the spread of serious diseases capable of wiping out entire crops, as happened prior to the Irish potato famine of 1854 and the Bengal rice famine in 1942. Therefore the initiation of exploitative agriculture without a proper understanding of the various consequences of every one of the changes introduced into traditional agriculture, and without first building up a proper scientific and training base to sustain it, may only lead us, in the long run, into an era of agricultural disaster rather than one of agricultural prosperity."

2. Based on the above analysis, the concept of an Ever-green Revolution which is defined as improving productivity in perpetuity without ecological harm was developed. The ever-green revolution methodology is based on an integrated natural resources management strategy and is not just commodity centered as the green revolution technologies. In particular soil health care, water conservation and management and integrated pest management receive attention on a systems basis. Also ever-green revolution technologies involve participatory breeding with farm families and participatory knowledge management systems.

3. Biotechnology involves a basket of technological approaches to promoting the productivity, profitability, sustainability and stability of major farming systems. The

biological software essential for sustainable agriculture like biopesticides, biofertilizers, vermiculture and similar techniques receive intensive attention.

4. The aspect of Biotechnology of concern to organic farmers is recombinant DNA technology. Such transgenics have been the subject of considerable debate and discussion. Biosafety protocols are still being evolved in different countries, although the Cartagena Protocol based on the Global Biodiversity Convention is now being implemented. What is now needed is the desegregation of the various concerns relating to genetically modified crops and find an answer to each of these concerns which is satisfactory from the point of view of science and society. Some of these concerns are:

- Who owns the technology?
- Will it be covered by IPR thereby resulting in social exclusion with reference to access to technology?
- Will the technologies address the real problems of small farmers?
- What are the environmental implications of GMOs?
- What are the food safety implications?

Thus, issues of equity, ethics, environment, economics and health safety are involved.

5. It is generally said that the Green Revolution was a product of public good research, while the Gene Revolution is largely in the hands of private sector. What steps should be taken to ensure that the new biological technologies are pro-poor, pro-nature and pro-women? These issues will be discussed in the lecture.

II.2 Biotechnology and world hunger: controversy and solutions

HERBERT J. KRONZUCKER
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The First and Second Green Revolutions are at the foundation of modern agriculture, and in particular the Second Green Revolution has staved off the starvation of millions. Largely as a result of the concerted research efforts of the Consultative Group on International Agricultural Research (CGIAR), impressive yield increases have been achieved in all major crop species. The yield of the world's foremost crop species, rice, has tripled since the 1960s. However, despite intense continuing research, recent yield gains have failed to keep pace with human population growth. Global food demand is expected to more than double by 2050. Moreover, high requirements of modern crop varieties for fertilizer, pesticides, and water pose at times grave environmental problems. The challenge to scientists is formidable if responsible agriculture is the goal, and if future famines on account of insufficient food production are to be avoided. This plenary talk discusses the necessity and promise of a Third Green Revolution that, in addition to improved agricultural management practices, draws upon a combination of techniques of classical breeding and, more controversially, biotechnology. Concrete biotechnological research initiatives currently underway in the area of nutrient absorption by plants are highlighted. In addition, key political and ethical issues are addressed.

II.3 Making plants medicinal: Transgenic tobacco, a biosafe platform for the production of biopharmaceuticals for oral administration.

JIM BRANDLE
Agriculture and AgriFood Canada, 1391 Sandford St., London, Ontario, CANADA N5V 4T3.

While the synergy created by collaborations between agricultural biotechnology and experimental medicine have brought molecular farming this far, it is becoming increasingly evident that the lack of biosafe production systems is a barrier to the success of transgenic plants for the production of biopharmaceuticals. In anticipation of that roadblock we started our efforts with a non-food crop, tobacco, as our platform. Using human interleukin-10, a contra-inflammatory cytokine, we showed that tobacco can produce biologically active cytokines and, using a mouse model of inflammatory bowel disease, demonstrated that it can be fed directly to test animals and have a strong positive clinical effect. However, no system is "made to measure" in its' native state and enhancements are necessary to ensure that the platform is both safe and economical. In response to that need we developed low-nicotine tobacco plants for recombinant protein

production with no selectable marker, with reduced non-host DNA, that are visually distinguishable (variegated) and male sterile, and with significantly enhanced protein accumulation capability. Now we are combining the many elements in one genetic background using both conventional and molecular breeding. The net result will be a platform that is both biosafe and practical.

Plenary session III

Plants, Canada and Climate Change

III.1 Biosphere solutions to climate change and clean energy: New challenges and opportunities for plant science research.

DAVID LAYZELL
CEO and Research Director, BIOCAP Canada Foundation and Queen's Research Chair in Biology, Queen's Univ Kingston, ON

Traditionally, the major driver for government and industrial investment in plant science research has been linked to the production of food, feed and fibre by our agricultural and forestry sectors. More recently, concerns about climate change and the sustainable supply of clean energy have created new challenges and opportunities for researchers in the plant sciences and related disciplines. My presentation will provide a background on the scale and nature of the climate change / energy challenge in Canada and identify some of the emerging areas for research in the plant sciences. I will also provide a brief overview on the work of BIOCAP Canada (www.biocap.ca), an national research foundation that promotes research to find biological solutions to the challenges of climate change and clean energy.

III.2 Effects of climate on yield of five crops in family *Brassicacae* in southern Ontario.

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The response of many vegetable crops to climate has not been well quantifed. In a previous study we demonstrated that the interactions between growing season climate were more complex than could be described by mean temperature and precipitation. Number of hot days (>30^{o}C) and days with precipiation were better corelated with several cool season vegetable crops. We also showed a return to more hot days per year in the mid 1908's onward after a cooling period from the early 1950s to early 1980s. To further investigate this relationship five crops of the *Brassicacae* family, broccoli (*Brassica olereacea* var *italica* L), cabbage (*Brassica olereacea* var *capitata* L), cauliflower (*Brassica olereacea* var *botrytis* L), rutabaga (*Brassica napus* L. var *napobrassica* (L.) Reichb) and radish (*Raphanus sativus* L.) were examined for relationships between yield and seasonal weather patterns for all yearly yield data on record. The itertive chi-squared technique was used to find correlations between daily temperature and precipitation records and annual marketable yield. The analysis showed that all five crops had some correlation between yield and mid to end-of-season temperature. Broccoli, cabbage and radish showed a reduced marketable yield with increasing occurance of days with maximum temperatures exceeding about 30^{o}C, while years with high yields of cauliflower and rutabaga had a deficit of warm days later in the season. For cauliflower high yield years were associated with an excess of days with *ppc* > 15 mm; in other words, cauliflower benefits from September rains. High yield years for cauliflower also showed a lack of days with T_{max} > 21^{o}C in October, which is when the processing crop is harvested. Rutabaga was similar to cauliflower except the Tmax was 17^{o}C. This work also suggests that the brassica vegetables cannot be treated as a uniform responding group in study of climate effects on yield. The gains in yield in the mid portion of the last Century may have been more to do with cooling than management. High temperatures decreased yields for cabbage, cauliflower and rutabaga by 0.27, 0.10 and 0.31 (t ha^{-1})/(day > 30^{o}C), respectively. For each crop this translates to about a 10% yield loss for every ten days in the growing season that the temperature reaches 30^{o}C or above espcially in late summer onwards.

III.3 Rising levels of greenhouse gases and forests in a changing climate: key messages from the Aspen FACE Project

KEVIN PERCY

Natural Resources Canada, Canadian Forest Service-Atlantic Forestry Centre, 1350 Regent Street, Fredericton, N.B., Canada E3B 5P7

There is a large degree of scientific consensus around the view that the world's climate is changing. While much attention is given to changes in global temperature and precipitation patterns, many other aspects of physical climate having direct and immediate feedback to plant productivity are also varying. Considerably less attention, however, is given aspects of the changing chemical climate, the main drivers of physical climate change. The first and third most important greenhouse gases contributing to radiative forcing (global warming) are historically increasing levels of atmospheric CO_2 and tropospheric O_3. Elevated levels of CO_2 have generally been shown to stimulate growth and productivity in carbon-limited plants such as forest trees. On the other hand, O_3 is phytotoxic above certain threshold concentrations and is known to reduce growth/productivity, alter patterns of C allocation, and predispose trees to increased incidence of insect and disease. Co-occurence of these two greenhouse gases over extensive areas of Canadian and world forest makes model predictions of potential "forest fertilization" from CO_2 problematic. Data from the world's largest, multi-year, ecosystem-scale Free Air Carbon Dioxide Enrichment (Aspen FACE) Experiment in northern Wisconsin, USA, have yielded consistent, and often counterintuitive signals on community-level response to CO_2 and O_3 in some of the most widely distributed forest tree species in North America. Experimental species include 5 genotypes of the fast-growing, climate-adaptable, early-success ional species trembling aspen *(Populus tremuloides).* Key above-ground findings will be summarized and placed into context of bottom-up and top-down ecosystem feedback and O_3 offsets of potential benefits from increasing atmospheric CO_2.

III. 4 Plant genetic and evolutionary responses to ecological crisis in Canada

QUENTIN CRONK
Univ of British Columbia

The rediscovery of Mendel's work independently by de Vries, Correns and Tschermak in 1900 unleashed a torrent of research on plant genetics which has continued undiminished to the present day. It has now been hugely augmented by genomics. Despite this. it is somewhat surprising that insufficient knowledge exists to make definitive predictions about icroevolutionary change in response to new ecological crises such as human-driven climate change. The recent whole genome sequencing of Black cottonwood (*Populus trichocarpa*) provides a promising model organism for such studies. Black cottonwood occurs from California to Alaska, and from sea level to nearly 1000 metres, so encompassing numerous climatic environments. There are now prospects for whole genome variation scans across the entire range of the species, using the new genomic tools available. Such "total range ecogenomic characterization" (TREC) approaches could form the baseline against which the genetic effects of environmental change could be monitored. Black cottonwood is enormously variable. The individual sequenced (Nisqually-1) is heterozygous approximately every 100 base pairs over a genome of c. 500 million base pairs. The challenge facing modern geneticists and ecologists is to understand which of these variations are neutral and which have ecological consequences and thus provide the raw materials for adaptive responses to environmental change. Achieving this unification of genomics and ecology (the so-called ecomolecular synthesis) is made urgent by the expected rates of change to natural ecosystems and populations over the next century.

Special Lectures

CSPP Gold Medal Lecture
My long journey in plant research: contributions to flavonoid biochemistry

RAGAI IBRAHIM
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This review is a retospective of a part of the author's contributions to the field of flavonoid biochemistry over the past four decades. It emphasizes novel aspects of some enzymes that catalyze later steps in flavonoid biosynthesis with reference to their biological significance. These include the methyltransferases, glucosyltransferases and an oxoglutarate-dependent dioxygenase that are involved in polymethylated flavonol glucoside synthesis and its compartmentalization in the semi-aquatic

saxifragaceous weed, *Chrysosplenium americanum.* Time permitting, the use of homology-based modeling of methyltransferases will be discussed in relation to gene function prediction of this important gene superfamily. The complete review will appear in the May issue of the Canadian Journal of Botany.

CBA Luella K. Weresub Lecture
Rust fungi and a rusty mycologist: The morphology, life cycle and taxonomy of rust fungi
YASU HIRATSUKA
Northern Forestry Centre, Canadian Forest Service, Edmonton, AB T6H 3S5
The following aspects of my investigations of the rust fungi (Uredinales), conducted during my tenure at the Canadian Forest Service, will be reviewed and discussed. 1) Morphological types of spermogonia and their significance to taxonomy of rust fungi; 2) Life cycle, cytology and nomenclature of western gall rust; 3) Definitions and nomenclature of spore states of rust fungi; 4) Publication of Illustrated Genera of Rust Fungi (2003) with G. B. Cummins.

Concurrent Session Abstracts

A1. Nutrient use efficiency (part 1)

A1.1 Legume Genomics and Biotechnology: Role in Sustainable Acquisition of Nutrients

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Since 1960 N fertilizer use has risen some 13-fold, accompanied by a 6.5-fold increase in P fertilizer use. Expanded use of these inputs in the developed world has contributed to compromising air and water quality while in developing countries the lack of N and P fertilizer availability limits crop production and quality. Concurrently, Earth's population has reached 6 billion people and this trend is on target towards 10 billion by 2040. Today some 0.8-1 billion people are undernourished due to lack of protein or nutrient insufficiency. Legume production and use are critical to sustainable acquisition of nutrients and to providing dietary protein to a growing world population. Legumes are grown on 25% of the Earth's arable land. They provide 33-60% of humankind's dietary N, depending upon the country in question. Moreover, 35% of the world's edible oil is derived from legumes. Because they can fix atmospheric N2, legumes can reduce fertilizer N inputs thereby aiding to reduce excessive N inputs in developed countries while supplying beneficial N to crops in developing countries. Legumes also have some unique adaptations to acquire P from low P soils. However, legumes are frequently grown under conditions that limit their growth and development. To keep up with the food requirements of a rapidly growing population, maintain adequate N2 fixation, and enhance nutritional quality, genomic and biotechnology research on legumes needs to be focused on improving traits related to these constraints. Using advances in *Medicago*, *Lupinus*, and *Phaseolus* as examples, this presentation will identify research thrusts that have potential to enhance either P or N acquisition and use. Findings with legume species may be applied to other plant families.

Note: In honor of Professor G. H. N. Towers

A1.4 Physiological role of alanine aminotransferase in *Arabidopsis thaliana*

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Many species of plants have been shown to accumulate alanine in addition to other end products of the fermentation pathways under a low oxygen stress. Alanine aminotransferase (AlaAT) catalyzes the amino group transfer from glutamate to pyruvate to produce alanine and 2-oxoglutarate. Up-regulation of AlaAT in response to a hypoxic stress suggests the involvement of AlaAT in low oxygen stress tolerance and nitrogen metabolism under such a condition.

We used a functional genetics approach to fully characterize the physiological role of AlaAT using Arabidopsis as our model plant. Screening of T-DNA tag lines that have insertions in the *AlaAT* genes yielded one knock-out mutant in *AlaAT1* (At1g17290) and 3 lines that have insertions in the promoter region of *AlaAT2* (At1g72330).

The *AlaAT1* knock-out mutant (*alaat1*) showed undetectable level of the enzyme activity, and the absence of the normal transcript and the protein in *alaat1* was confirmed by RT-PCR and Western blot analysis, respectively. This result indicated that the *AlaAT1* gives rise to the most of the AlaAT activity in Arabidopsis at least under a normal condition. However, the involvement of only *AlaAT1* in the hypoxic stress response is not likely since both *AlaAT1* and *AlaAT2* were shown to be induced at transcriptional level when plants were hypoxically treated.

In addition to the T-DNA tag lines, we have generated transgenic lines that overexpress barley *AlaAT*. Increased levels of AlaAT activity were detected in the overexpressing lines.

We are currently conducting several experiments to find novel phenotypes of the mutants and the overexpressors. When plants were grown on the medium supplemented with alanine, *alaat1* showed reduced growth, suggesting that AlaAT1 may convert alanine to pyruvate to use it as a nitrogen source. The growth and the survival rate of the plants when subjected to low-oxygen stress are also being studied.

A1.5 What poplar trees choose to eat: 15N discrimination as an indicator of nitrogen dynamics in *Populus trichocarpa*

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Nitrogen's isotopic composition ($d^{15}N$) varies in natural systems. This variation carries the potential for providing information on plant nutrient dynamics and its underlying physiology, but mechanisms causing isotope discrimination are poorly characterized. As trees, poplars provide a unique model for unraveling ^{15}N discrimination during uptake and assimilation in terrestrial plants. Isotope discrimination factors (*D*) should vary as a function of supply relative to demand. We are testing this hypothesis on *P. trichocarpa* stecklings maintained on NH_4^+ in hydroponics. Pruning treatments are being used to alter either the tree's uptake capacity (reduce roots) or nutrient demand (reduce shoot). Previous studies with herbaceous species have shown that altering the shoot:root ratio significantly impacts nitrogen assimilation rates. Following treatment, *D* is determined using a substrate depletion method whereby the relative ^{15}N enrichment occurring during uptake and assimilation provides a measure of net discrimination. NH_4^+ is measured using the Phenol-Hypochlorite method, and N is collected for isotope ratio mass spectrometry by ammonium diffusion and deposition onto acidified glass fiber disks. Initial results show increasing *D* values (0 to 12 ‰) for increasing NH_4^+ concentrations. Additionally, data indicate a shoot pruning effect on *D* within 30 minutes, which continued for the following three days of sampling. The decrease in *D* with pruning found in these initial results may indicate a decrease in NH_4^+ ion efflux relative to influx across root cell plasmamembranes. The understanding emanating from this research will lead to an improved basic understanding of nitrogen isotope discrimination in higher plants and will contribute to models of $d^{15}N$ variation in plant physiology and ecology.

A1.6 Nitrogen use efficiency of field-grown bell green peppers

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Due to increasing crop input costs and increasing pressure from society on the agricultural sector to protect consumers and the environment, there is potential to improve nitrogen (N) use efficiency (NUE) allowing for optimal crop yields while minimizing environmental N losses. Three field experiments were conducted to determine the effects of different N rates, timing of application, and technology (i.e. a nitrification inhibitor combined with an urease inhibitor) on bell green pepper yield and NUE. Preplant ammonium nitrate was broadcast applied at 0, 35, 70, 140, and 210 kg N ha^{-1}, as well as two split application of 35 plus 35 and 70 plus 35 kg N ha^{-1} applied preplant and at first fruit set, respectively. A preplant application of UMAXX®, a urea-based fertilizer that contains a nitrification inhibitor and an urease inhibitor, was broadcasted at 70 kg N ha^{-1}. At harvest, the following parameters were evaluated: total and marketable yield; crop quality including fruit weight, length, diameter, and wall thickness; shoot and fruit percent total N and phosphorus; and soil mineral N. Results presented here are based on the first year of a four year study. Although plants with 0 or 35 kg N ha^{-1} applied displayed symptoms of N deficiency, the quantity of N applied (0-210 kg N/ha), the method of application (preplant vs split) and N source (ammonium nitrate vs nitrification and urease inhibitor) did not influence total and marketable yield nor crop quality. Total percent N in the fruit and shoot was lower in non-fertilized plants compared to plants receiving 70 or 210 kg N ha^{-1}. But there were no differences between N treatments in plant total percent phosphorus. There were considerable differences between trial field locations in soil mineral N, yield, NUE, and plant N uptake and removal. For example, NUE (expressed as the difference in plant N from fertilized vs. non-fertilized plots divided by quantity of fertilizer applied) ranged from 49% to 105% at 70 kg N ha^{-1} depending on the field. Moreover, NUE decreased as N rates increased from 70 to 210 kg N ha^{-1}. Based on an N budget at harvest, the 70 and 210 kg N ha^{-1} fertilized plots had on average 35 and 190 kg N ha^{-1}, respectively, more residual N (i.e. crop N residue plus soil mineral N at 0-75 cm depth) than non-fertilized plots. Overall, preliminary results indicate opportunities to optimize NUE and reduce potential

A2. Contemporary issues in statistics, data management, plant biology and agricultural research

(part 1)

A2.1 Mixed model analyses (REML) for three different series of agronomic trials.

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The scope of mixed models exploded after the statistical procedure of Restricted Maximum Likelihood (REML) became established. Agronomic researchers use many different mixed models with REML to, for example, model spatial variation within fields, investigate cultivar by environment interactions, and combine cultivar information from series of similar trials. This study considers the performance of cultivars when they are assessed over one or more target regions. We present mixed models for three different types of trials: 1) Cereals - annual crops with trials as random effects in a single target region; 2) Corn hybrids - an annual crop over a range of target regions with the measured corn heat units at each trial as a covariate; 3) Forages - perennial crops in a single target region. These studies focus more on describing the performance of cultivars rather than on formal significance tests for statistical differences between cultivars.

A2.3 Biplot analysis of genotype by environment data

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ECORC-AAFC, Ottawa

Due to the ever-presence of genotype by environment interactions (GE), crop variety trials are usually conducted at multiple locations or environments, resulting in genotype by environment two-way data. Although the original incentive of variety trials is for cultivar evaluation, genotype-by-environment data are also valuable for understanding the target environment and the test locations. Complete analysis of a genotype by environment dataset consists of 1) mega-environment investigation, 2) test environment evaluation, and 3) GE interpretation, in addition to 4) genotype evaluation. Genotype evaluation is meaningful only when the first three issues are adequately addressed, explicitly or implicitly. A biplot is a scatter plot that approximates and displays a two-way table by both its row factors and column factors so that relationships among column factors, relationships among row factors, and interactions between row and column factors can be visualized. A GGE biplot is a biplot that approximates and displays the genotype main effect (G) and GE of a genotype by environment two-way table, which are the two sources of variation that are relevant to, and must be considered simultaneously, in genotype and test environment evaluation. GGE biplot analysis has evolved into an important technique for genotype by environment data analysis, whereby all four issues aforementioned can be graphically and effectively addressed. In this paper I will present discussions on the following topics: 1) objectives of genotype by environment data analysis; 2) principles of biplot analysis; and 3) biplot analysis of genotype by environment data. The discussions should be applicable to biplot analysis of other types of two-way data.

A2.5 Information systems for crop performance data: motivation, status, and potential.

N.A. TINKER
AAFC, Ottawa

There is increasing motivation for efficient storage and retrieval of crop performance data. This is driven by numerous related factors: a desire to increase the value of crop performance tests; an awareness of the need to integrate data from multiple sources; new opportunities to link phenotypes with genomic data; an increased number of stakeholders who need access to data, and an increased need for convenient access to data that is current, complete, and accurate. There is also a growing sophistication and awareness of the role and capabilities of modern informatics techniques in biological research – an area that has become known as “bioinformatics”. Bioinformatics, in partnership with statistics, can play a vital role in increasing the value of crop performance data. However, much of this role remains to be developed and adopted by the communities who gather and use this data. Part of the problem is that phenotypic data is highly complex: it requires extensive information about the context and conditions under which it was collected, standardization of plant growth stages, descriptions of varieties, classification of traits, and explanations of measurement techniques. If context is neglected, the data is useless, but if context is overly detailed, it may not be recorded or it may be used improperly. Several solutions

have been developed to address these needs. These include commercial software packages, open-source collaborations, and a new application co-developed by the author. Each solution has strengths and weaknesses, and each addresses different types of needs. This presentation will discuss the current status and availability of crop performance databases, and will elaborate on the challenges that must be met to achieve future potential.

A3. Systematics and evolution I

A3.1 Near the root of monocot phylogeny?
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A series of recent phylogenetic studies have shed light on relationships among the deepest branches of the monocot portion of the tree of life, culminating in the recognition that a small family of "melanthioid" monocots, Petrosaviaceae, is likely the sister group of all monocots except *Acorus* and Alismatales. Most studies also agree that *Acorus* defines the deepest split in monocot phylogeny, but several have reported a placement of this taxon within Alismatales. We examine current hypotheses of basal monocot relationship using a large plastid data set and published mitochondrial data, and explore the possibility that *Acorus* may become misplaced during phylogenetic analysis due to an elevated rate of molecular evolution in at least part of the mitochondrial genome. Strong conflicts have also been reported within Alismatales concerning the relative arrangements of its three major component clades: Araceae (incorporating the duckweeds), Tofieldiaceae (another "melanthioid" splinter) and the core alismatid families (Alismataceae, Zosteraceae and others). We estimate error rates on the inference of these discordant relationships, and summarize additional findings on relationships among the core alismatid families.

A3.2 The Phylogeny of *Danthonia* DC. (Poaceae: Danthonioideae) in North America.
ELIZABETH REIMER [1], J. HUGO COTA-SÁNCHEZ [1]

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The genus *Danthonia* (oat-grass) makes up a minor component of the Canadian prairies. Eight species occur in North America (NA) but the degree of phylogenetic relationships is unknown. In order to elucidate relationships within the genus, and to determine associations with South American (SA) and European (EUR) species from which they are presumed to have evolved, a molecular phylogeny was constructed from the sequence of the *trn*L intron and *trn*L-F intergenic spacer of the chloroplast genome. Micro- and macromorphological characters were examined using a scanning electron microscope and stereomicroscope to determine whether morphological characters support the phylogenetic hypothesis constructed from the chloroplast marker. Molecular analysis supports a monophyletic genus *Danthonia*, distinct from other genera within the subfamily Danthonioideae; however, preliminary analyses indicate that within the genus, NA *Danthonia* species cannot be separated from those of SA or EUR based on either morphological or molecular data. No morphological characters examined to date have been successful in resolving terminal taxa, or in differentiating NA species from their SA and EUR relatives. The morphological data set shows high incidence of homoplasious characters suggesting, quite possibly, complex patterns of evolution, including convergence of morphological characters, putative hybridization, and reproductive strategies.

A3.3 Anomalous periderm in Middle Eocene *Decodon allenbyensis*: Understanding aerenchyma development and the importance of a novel aquatic bark system.
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Aerenchyma, typical of aquatic plants, is known in both primary and secondary tissues in the submerged axes of *Decodon allenbyensis* Cevallos-Ferriz et Stockey, from the Middle Eocene Princeton Chert. Recent work has allowed us to describe the morphogenesis of these arenchymatous

tissues in detail, showing the transition from young roots, with delicate primary tissues to larger woody axes with lacunate phellem and wood. Patterns of development of the primary cortex aerenchyma, in the fossil, contrast with that seen in species of Onagraceae (e.g. *Ludwigia* L.). Aerenchymatous tissues of secondary origin in the fossil, arising from a phellogen (lacunate phellem), are also known in several aquatic to semi-aquatic taxa in extant Lythraceae, Melastomataceae, Onagraceae, Euphorbiaceae, Myrtaceae and Fabaceae. Our recent investigations have shown the woody fossil roots, with phellem, connected to exceptionally large woody axes, some with up to 18 growth increments. The largest axes possess a distinct and unique complex of secondary tissues arising from a phellogen system. Bands of phelloids alternate with bands of non-active phloem (with lacunae); such "lacunate phloem" as well as the aquatic rhytidome are currently unknown in any living or fossil taxa. These aquatic adapations for the fossil roots are now known to a degree that rivals that for most extant taxa, and contributes to the large number of detailed characters known for the whole-plant concept of *D. allenbyensis* at the Princeton Chert.

A3.4 Quantitative relationships between floral characters in the genus *Anthurium* (Araceae)

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Angiosperm flowers are regarded as complex and integrated systems where floral traits coevolve together to ensure and maximize pollen transfer. In regard to this problem, pollen size and number may be involved in different associations with other floral traits. To determine such relationships, which may provide insight to understand floral evolution, we studied 20 species of *Anthurium*, which are tropical long living epiphyte plants.

The characters analysed were pollen number, volume, stigma area, style length and flower number per inflorescence. Contrary to what has been reported in the literature, no correlation was found between pistil length and pollen volume nor pollen size and number at the interspecific level. Instead, pollen number was positively correlated with flower peduncle diameter. Finally, the total stigmatic area of the inflorescence was positively correlated with both pollen grain number and pollen grain size per flower. These results suggest that adaptative constraints driving pollen size and number could be different in the genus *Anthurium* than what is found in temperate Angiosperm. Also, the relationship between pollen (number and size) and stigma area could clarify whether or not those 2 floral traits have a functional link. The possible evolutionary advantages of those interactions are discussed in relation to the pollination cycle, life cycle and habitat of *Anthurium.*

A3.5 Molecular phylogeny of the genus *Philodendron* (Araceae) : clarification of its taxonomic position and species level classification

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The genus *Philodendron* (Araceae) is a large neotropical group and its classification remains unclear. Previous classifications have been based on morphological characters, mainly on characters of inflorescences and flowers. Yet, the original classification by Engler in 1913, with few modifications, is still the one used today. Here we present a molecular approach to verify the traditional classification. Two nuclear markers, ITS and ETS, and the chloroplast DNA region *rpl*16, were sequenced and analysed for almost 50 species of *Philodendron* and its close relative *Homalomena.* Our samples are mainly from Mexico, central America and northeastern South America. These analyses allow us to resolve the relationship between *Homalomena* and *Philodendron* and among species of *Philodendron* sampled. According to the data, the genus *Homalomena* is monophyletic and is nested within *Philodendron*, which is therefore paraphyletic. The sister clade to *Homalomena* is *Philodendron* subgenus *Meconostigma*, which is also supported as monophyletic by the molecular data. Below the subgeneric level, the group obtained in our phylogeny do not correspnd to previous classifications based on morphology. However, the three DNA regions

yield similar groupings. Mapping morphological characters of the inflorescences onto the molecular phylogeny improves our understanding of the evolution and classification of *Philodendron* and helps with the choice of morphological characters for futures classifications.

A3.6 Inference of C_4 photosynthesis evolution in *Flaveria* (Asteraceae) based on phylogeny.

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A well-resolved phylogeny of *Flaveria* (Asteraceae) is presented using morphological, life history, and DNA sequence data (chloroplastic *trn*L-F, nuclear ITS and ETS). Data were collected from 21 of 23 known *Flaveria* species and two Flaveriinae outgroups. Bayesian and parsimony analyses of each data set yielded congruent results, and the phylogeny is based on the analysis of all data combined. This phylogeny is used to infer evolutionary relationships among *Flaveria* species, center of origin and dispersal patterns, and C_4 photosynthesis evolution. Our phylogeny demonstrates that all C_3 *Flaveria* are ancestral to intermediate (C_3-C_4 and C_4-like) and fully expressed C_4 *Flaveria* species. There are two strongly-supported, derived clades (A and B) present. C_3-C_4 intermediates are found in both clades, and are evolutionary intermediates in clade A, but not necessarily in clade B. C_4-like photosynthesis is also derived separately in each clade and is the pre-existing condition to C_4 photosynthesis in clade A. Fully expressed C_4 photosynthesis is only present in clade A and may have evolved up to three times within this clade. We infer the center of origin of *Flaveria* is the Puebla-Oaxaca region of Mexico. The evolution of intermediate and C_4-like photosynthesis in *Flaveria* is hypothesized to have occurred following genetic "preconditioning" and intense environmental selection pressures. C_4 photosynthesis follows the evolution of both annualism and self-compatibility in *Flaveria*, supporting the hypothesis that generation time may play an important role in evolving fully expressed C_4 photosynthesis.

A4. Development of the vascular system

A4.1 Vein pattern formation in *Arabidopsis* leaves

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The patterning of veins provides both mechanical stability and efficient transport routes to the leaf and must be closely coordinated with leaf shape. The primary model to explain the formation of vein pattern is auxin canalization, whereby exposure of some cells to elevated auxin causes those cells to adopt a vascular fate and to become more efficient auxin transporters. Directional auxin transport in these cells results in two classes of neighbouring cells: those to which auxin is transported adopt a vascular fate, those from which auxin is drained adopt a non-vascular fate. The auxin response pattern in developing leaves has led to the proposal that sequential auxin sources are responsible for the formation of vascular pattern: 1) the midvein is established through auxin entering the leaf from an external source, 2) secondary and higher order veins are established through auxin produced along the leaf margin. We have identified a number of mutations that result in defective vein patterning and alter either auxin transport or response. Mutants in *FORKED1 (FKD1)* show a leaf-specific reduced auxin response that results in a lack of distal vein junctions and hence an open venation pattern. We have cloned *FKD1* and will present an analysis of its expression pattern during leaf development. Mutants in *AUTOBAHN* (*ABN*) have narrow, elongated leaves with a proliferation of midveins and an altered pattern of auxin response which our analysis suggests results from defective auxin influx. The asymmetry of auxin response pattern seen early in leaves mutant for *ASYMMETRIC LEAF1* or *2* (*AS1, AS2)* is correlated with later asymmetries in both vascular pattern and leaf shape. Based on our analysis of these mutants, we propose a model by which the correct spatial and temporal distribution of auxin within leaves is interpreted

to establish not only vein pattern, but also leaf shape.

A4.2 Cloning and expression analysis of *FKD1* - a gene that directs vein formation in response to auxin.

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We are establishing the mechanism by which the novel *Arabidopsis thaliana* gene *FKD1* directs leaf vein pattern formation in response to auxin . We have previously described plants mutant for *FORKED1 (FKD1),* which have reduced leaf response to auxin and show lack of distal vein meeting in leaves. Our analysis of *fkd1*-1 has allowed us to propose a model whereby *FKD1* is necessary for the auxin response that directs vein patterning, and responds to different auxin concentration thresholds. We have now identified *FKD1* through map-based cloning. We first defined the *FKD1* locus to a region of about 30 genes on chromosome 3. We then screened SALK lines identified as having T-DNA insertions within these genes. Several SALK lines, all corresponding to insertions in a single gene had *fkd1-1*-like phenotypes. These lines do not complement *fkd1*-1, suggesting that they represent alleles of *FKD.* Sequencing of *fkd1-1* reveals a base substitution of "G" for "A" within the putative *FKD1* gene in the 6th intron adjacent to the 7th extron, suggesting a possible splicing defect. Transformation of *fkd1-1* plants with genomic DNA including the putative wild type *FKD1* gene resulted in a reversion to the wild type phenotype, indicating that we have indeed identified the *FKD1* gene. The *FKD1* gene encodes two mRNAs as the result of incomplete splicing of the transcript. These are predicted to encode two proteins, one is possibly targeted to the chloroplast, and the other associated with the plasma membrane. We are currently assessing the relative abundance of the two splicing products at different points in development by RT-PCR. As well, we are establishing if either or both splice products are necessary to revert the *fkd1-1* phenotype to wild type. Finally, we have constructed *FKD1* promoter reporter gene fusions (*FKD1*::*GUS*, *FKD1*::*LUC*, *FKD1:*:GFP) in order to assess *FKD1* expression pattern in wild type plants.

A4.3 *Arabidopsis* eIF-5A1 is a positive regulator of xylem development

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Eucaryotic translation initiation factor 5A (eIF-5A) is thought to facilitate protein synthesis by participating in the nuclear export of specific mRNAs. In *Arabidopsis,* there are three isoforms of eIF-5A, and one of them, *At*eIF-5A1, is expressed in senescing tissues. More recently, we found that *At*eIF-5A1 is also expressed in developing xylem. In order to determine whether *At*eIF-5A1 plays a role in xylem formation, its full-length cDNA was constitutively over-expressed in transgenic *Arabidopsis* plants. These plants had larger rosette leaves as well as taller and thicker inflorescence stems in comparison to control plants, reflecting more rapid growth. In addition, their main inflorescence stems exhibited a significant enhancement in xylem development. In both vascular and interfascicular regions, cell layers and tissue thicknesses were increased by up to 80% and 60%, respectively, depending on the line. Increased xylem growth was more pronounced in secondary xylem than in primary xylem. Moreover, constitutive antisense suppression of *At*eIF-5A1 resulted in stunted statures and delayed leaf senescence as well as ~30% reduction in xylem tissue in comparison with control plants. eIF5A is post-translationally modified by conversion of a conserved lysine to hypusine. To confirm that it is the hypusinated form of eIF5A1 that regulates xylogenesis, we also generated transgenic *Arabidopsis* plants with constitutive over-expression of a mutant form of *At*eIF-5A1 that cannot be hypusinated. The mutation was made by changing the codon, AAG, for the conserved lysine to GCG (for alanine), thereby disabling hypusination of *At*eIF-5A1. In the mutant population, there was no change in xylem development relative to control plants. Thus, *At*eIF-5A1 appears to positively regulate xylogenesis, possibly by facilitating the programmed death of treachery elements, and such a function requires activation of the protein through hypusination.

A4.4 Promoter activities of deoxyhypusine synthase and eukaryotic translation initation factor 5A1 in *Arabidopsis thaliana*

suggest a role in xylogenesis, leaf senescence and flower senescence

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Numerous reports have documented the importance of deoxyhypusine synthase (DHS) and eukaryotic translation initiation factor 5A (eIF-5A) in human cell proliferation and apoptosis. However, relatively little attention has been given to their plant kingdom counterparts, despite their ubiquitous presence and highly conserved sequences. DHS activates eIF-5A by modifying a conserved lysine to the unusual amino acid hypusine, in one of the most specific post-translational modifications discovered to date. Recent evidence shows there are multiple isoforms of eIF-5A and it is believed that each functions as a shuttle protein, responsible for transporting a specific subset of mRNAs (either proliferation-related or senescence-related) from the nucleus to the cytoplasm for translation. To further elucidate the roles of DHS and eIF-5A1, one of three eIF-5A isoforms in *Arabidopsis*, the promoter activities of both were characterized during growth and development of transgenic *Arabidopsis* plants using GUS as a reporter gene. The levels of DHS::GUS and eIF-5A1::GUS expression were strongly up-regulated at the onset of leaf senescence, and this high expression was maintained until leaf senescence terminated. Furthermore, and unexpectedly, DHS and eIF-5A1 were transiently and co-ordinately expressed in the vascular tissues of developing cotyledons, rosette leaves and stems. The timing and localization of the GUS expression imply a role for eIF-5A1 in xylogenesis. In addition, DHS and eIF-5A1 were co-ordinately expressed in anther tissues during the degenerative stages of pollen production. Taken together, these results suggest that DHS and eIF-5A1 regulate programmed cell death associated with xylogenesis, leaf senescence and flower senescence in *Arabidopsis*.

A4.5 Changes in cherry fruit tracheary element structure and function during development and ripening.

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Developmental changes during fruit ripening have been studied extensively in grape, where there are three stages of growth; the last one is associated with a hydraulic isolation of the fruit xylem from that of the rest of the plant. This hydraulic isolation correlates with stretching and breakage of the tracheids within the fruit. Cherry fruit also has three stages of growth and becomes somewhat hydraulically isolated during the latest stage. Details of this process in cherry were observed by making measurements of hydraulic conductivity, dissections, clearings, and macerations of the fruit and pedicel during fruit development. The vasculature of the fruit consists of a peripheral system of bundles that form a network in the outer mesocarp, and send branches inward as far as the mid-mesocarp. There is also a more internal set of bundles that run along the ridges of the stone. Two of these bundles enter a channel in the stone and connect with a branching system in the seed coat. The conducting cells of the xylem system of the fruit are all tracheids with spiral thickenings, in contrast to the xylem of the pedicel where the conducting elements are mainly vessels with simple perforation plates. During the first phase of fruit development, the tracheids in the growing fruit are immature and still contain protoplasts. During the second phase of development, characterized by no appreciable increase in volume but a maturation of some tissues, the tracheids lose their contents and become mature. During the third phase (in which the fruit volume increases markedly) tracheid groups stretch and break, and tyloses form within their lumens. The hydraulic isolation thus achieved is believed to be important for fruits that are very sweet, as the fruit continues to be fed by the phloem while backflow through the xylem is prevented when tree water potentials are low.

A4.6 "Pre- and post- genome" approaches to investigating the genetic regulation of wood development

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Our interest is in the genetic regulation of wood formation. We have utilized a variety of approaches, reflecting the rapidly expanding realms of possibility in the "pre- and post-genomics era" to gain some understanding of

this complex process. Currently, analysis of transcriptional changes during stem development in poplar is being carried out, with the use of15.5k gene cDNA microarrays. "Pre-genome," our work focused on questions regarding the specific relationship of auxin to wood formation. To tackle these questions, a poplar ortholog of the Arabidopsis auxin response transcription factor *Monopteros* was cloned from cDNA and geomic libraries. Functional analysis of this key regulator of auxin perception includes attempted rescue of the mutant phenotype in Arabidopsis, preparation and analysis of transgenic poplars overexpressing the *MP* ORF, and conditional silencing of the Arabidopsis *MP* to gain insight into a role for ARFs in interfascicular fibre formation.

A5. Pulse cropping and disease

A5.1 Intercropping Pulse Species with Barley: Assessing Agronomic Feasibility and Benefits

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Research is being conducted to develop more sustainable cropping systems for the black soil zones of Alberta by intercropping pulses with barley to increase spatial species diversity. To assess intercrop feasibility and benefits, field experiments were conducted at three sites in north-central Alberta in 2004. Faba bean (*Vicia faba*), lupin (*Lupinus angustifolius)* and pea (*Pisum sativum*) were grown at four planting densities (50, 100, 150, 200% of the recommended monoculture planting density (PD)) as monocultures and intercropped with barley (at 25% normal PD). All tests were grown without added nitrogen (N) fertilizer.

Preliminary results indicate that pea and faba bean can successfully compete and grow with barley. Faba bean comprised 46-78% of the intercrop seed yield (depending on pulse PD) while pea comprised 50-76% of the intercrop seed yield. Over-yielding (land equivalent ratio >1) occurred in 63% of the faba bean-barley and pea-barley intercrops. Barley from faba-barley intercrops had a higher protein content (119.6 g protein kg^{-1}) than monoculture barley (104.3 g protein kg^{-1}), while barley from pea-barley intercrops contained 115.6 g protein kg^{-1}. This suggests that high protein barley could be grown without additional N fertilizer thereby reducing input costs.

Lupin did not compete well with barley and may be unsuitable for intercropping as it comprised only 7-22% of the intercrop seed yield. Lupin, however, showed promise as a new monocrop. In high rainfall environments, lupin monocultures yielded 2.2-3.5 t seed ha^{-1} with protein yields of 1.0 t ha^{-1}. This pulse crop may be suitable for increasing temporal and economic diversity in Alberta cropping systems.

Using pulses to increase species diversity in Alberta cropping systems appears feasible. To further understand intercropping ecology this project is also investigating plant competitive interactions, soil microbial diversity, plant residue decomposition and nutrient cycling.

A5.2 Alternative tools for minimizing the damage of Ascochyta blight in chickpea

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Seeds of chickpea (*Cicer arietinum* L.) are an important source of plant dietary protein. This annual legume also provides an alternative to conventional crops in diversifying cereal-based monoculture in northern Great Plains. However, the production of chickpea has been devastating in the past number of years due to ascochyta blight (Ab), a disease caused by *Ascochyta rabiei.* Producers rely heavily on fungicides to control the disease, with up to six applications being used during a single growing season. Heavy use of fungicides increases production costs and may lead to serious environmental concerns. This study was conducted to develop alternative tools to minimize the damage caused by Ab. The study was focused on i) determining optimal plant canopy structure with improved plant architecture to manipulate

microenvironments under which the disease infection occurs, and ii) using alternative seeding patterns to increase the coverage of fungicides on the canopy, and iii) improving the efficiency of fungicide application. The study also determined the susceptibility of cultivars currently available in northern Great Plains as well it compared the intensity of infections on leaves, stems, and pods among cultivars, to develop spray strategies. The ultimate goals are to reduce disease pressure, minimize fungicide usage, and improve crop yield and environmental benefits.

A5.3 Evaluation of disease management strategies for ascochyta blight of chickpea in Alberta

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Blight caused by *Ascochyta rabiei* (Pass.) Labr. [teleomorph: *Didymella rabiei* (Kovacheski) v. Ayx (= *Mycospharella rabiei* Kovacheski] is a devastating disease in many chickpea (*Cicer arietinum* L.) growing regions of the world. In southern Alberta, the disease is persistent in farm fields, causing severe yield loss depending on the location, chickpea cultivar grown, cultivation practices, and amount of precipitation. Experiments were conducted at Brooks, Lacombe and Vegreville, AB in 2004 to evaluate 25 chickpea cultivars/lines for resistance, and to determine the effect of host plant resistance, plant population density, and application of foliar fungicides on the rate of ascochyta blight development. Ascochyta blight was consistently more severe on unifoliate cultivars of chickpea, such as the kabuli cultivar 'Dwelley' compared to those with a pinnate leaf structure, such as the desi cultivar 'Myles'. A positive linear relationship between plant population density and disease severity, and a negative linear relationship between disease severity and yield, were observed. Foliar application of the fungicides Headline and Quadris reduced disease severity and disease progress significantly compared to the non-treated control. Four to five spray applications were required to keep the disease severity level at 50% up to 75 days after inoculation. These results suggest that integration of cultivar resistance, reduced plant density, crop rotation and fungicide treatments could be useful as tools for managing ascochyta blight of chickpea.

A5.4 Impact of lodging and foliar fungicide on severity of mycosphaerella blight and yield in field pea lines.

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The objective of this study was to assess the impact of lodging and foliar fungicide (several applications of chlorothalonil) on the severity of mycosphaerella blight caused by *Mycosphaerella pinodes*, and on seed yield and 1000-seed weight of 10 pea genotypes. The test was conducted under irrigation at Saskatoon and Outlook SK for two years. Lodging was reduced by letting the crop grow up through a wire mesh 30 cm above the ground. Blight severity on stems, foliage, and pods was rated 3 - 4 times per year. The effects of reduced-lodging and fungicide were additive; both treatments reduced blight severity and increased seed yield and weight. Early lodging was associated with the largest reductions in seed yield and seed weight. Application of foliar fungicide had a smaller impact and was likely not cost-effective in most years. Small but significant differences in disease severity were present among the field pea lines, verifying the presence of partial resistance. We conclude that losses caused by mycosphaerella blight may be reduced by breeding for improved resistance to lodging, and that breeding for partial resistance may also hold promise.

A5.5 Impact of seeding and disease initiation dates on seed yield and severity of mycosphaerella blight in field pea

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Mycosphaerella blight (*Mycosphaerella pinodes*) reduces field pea (*Pisum sativum*) production throughout Canada. Although disease severity depends to a great extent on environmental conditions, especially moisture, the plant growth stage at which the pathogen attacks also affects disease outcome. Field trials were conducted at Edmonton, AB and Morden, MB in 2004 to assess the impact of seeding date on blight severity in eight field pea cultivars. Foliar and stem disease severity was significantly greater where pea plots were seeded 3-4 wk earlier than in later-seeded plots. However, yield potential was reduced at later seeding dates. Field plots were established at Vermilion, Vegreville and Edmonton, AB in 2004 to determine the effects of disease initiation date on the severity of mycosphaerella blight on field pea and the potential to cause yield loss. Disease initiation in early July caused the greatest level of disease severity. Disease severity was significantly reduced by a one-week delay in disease initiation, and was further reduced by an additional week of delay in disease initiation. However, delay of disease initiation beyond the third week of July did not result in further reduction in the disease level. Relatively dry weather may have mitigated disease pressure during late July and early August. Disease pressure caused a 13% reduction in yield in plots with the earliest disease initiation date at the Vegreville site, compared to plots where disease was initiated in mid-July. However, differences in yield were not statistically significant at the Vermilion and Edmonton sites.

A5.6 Etiology of Stemphylium blight of lentil (*Lens culinaris*) in Saskatchewan.

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Stemphylium botryosum results in large-scale defoliation on lentil and the prevalence of the disease has been increasing in Saskatchewan. Although not much is known about the biology of the fungus, yield losses of up 62% have been reported in Bangladesh. Preliminary meteorological studies which have been carried out in India and Bangladesh have shown that the disease requires high temperatures (23 to 30°C) and relative humidity of 94% for optimal disease development. The fungus is a poorly researched and no information is available on the biology of the pathogen in a semi-arid climate. Experiments were conducted to determine the optimal environmental factors for *S. botryosum* spore germination and development, such as age of fungal cultures, light regimes, temperature and wetness periods. The light regimes and the age of the fungal cultures did not have an effect of spore germination. High temperatures favored the germination of *S. botryosum* and the optimum temperature was between 25 and 30°C. However, germination of 30 to 40% was reached at temperatures as low as 5°C after 20h of incubation. The interrupted wetness periods significantly reduced the spore germination after exposure of 2h of wet conditions. The spores were sensitive to dry periods longer than 12h. The fungus required optimum leaf wetness of above 12 h and temperatures of between 25 and 30°C for disease development. However, considerable disease severity was prevalent at leaf wetness periods of 6h and also at lower temperatures as well. Our results imply that the fungus may be prominent late in the season, when warm and moist conditions are prevalent. Low severity can be experienced under unfavourable conditions as it seems to be well adaptable to the environmental conditions.

A6. Molecular aspects of pathogenesis I

A6.1 Differential expression and localization patterns of type III peroxidase genes isolated from *Blumeria graminis*-infected wheat epidermal cDNA library

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Plant peroxidases (class-III PRXs) are members of a large gene family that catalyze oxidoreduction between H_2O_2 and various reductants. Unlike most other enzymes, PRXs may have roles in both the production and scavenging of reactive oxygen species (ROS) and are involved in a broad spectrum of physiological processes. Previous studies have indicated the involvement of PRXs in defense against pathogen infection in various plant species and the pathogen-inducible *PRXs* have hence been categorized into the *PR*-9 family of pathogenesis-related *(PR*) genes. However, information regarding systemic investigation of *PRXs* in a specific pathosystem is still limited. In order to identify all potential pathogen-induced *PRX* genes and their functions in wheat-powdery mildew pathosystem, we established a cDNA library from leaf epidermis of diploid wheat (*Triticum monococcum*) after infection with fungus *Blumeria graminis* f. sp. *tritici* (*Bgt)* and identified 36 cDNAs representing 10 peroxidase genes (designated *TmPRX1* to *TmPRX10*) from 2500 expressed sequence tags (ESTs). Alignment of the deduced amino acid sequences and phylogenetic clustering with PRXs from other plant species demonstrated that these PRXs fall into four distinct groups. Differential expression and tissue-specific localization among the members were observed during the *Bgt* attack using northern blots and RT-PCR analyses. Consistent with its abundance in the EST collection, *TmPRX1* expression showed the highest induction during pathogen attack and fluctuated in response to the fungal parasitic stages. *TmPRX1-TmPRX6* were predominantly expressed in mesophyll cells, whereas *TmPRX7-TmPRX10,* which feature a putative C-terminal propeptide, were mainly detectable in epidermal cells. Using TmPRX8 as a representative, we demonstrated that its C-terminal propeptide was sufficient to target a GFP fusion protein to the vacuoles in onion cells. Finally, differential expression profiles of the *TmPRXs* after abiotic stresses and signal molecule treatments were used to dissect the potential role of these peroxidases in multiple stress and defense pathways.

A6.2 Towards the identification of the biological function of N gene.

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The induction of localized plant cell death or hypersensitive response (HR) is one of the commonly employed defense strategies in plants to limit the spread of pathogenic microorganisms. The molecular interactions in the tobacco mosaic virus (TMV) induced HR in tobacco has been the subject of intense study over several years. The N gene and the corresponding avirulence (avr) protein of this interaction have been extensively characterized. However many important questions still remain unanswered. It has been demonstrated that two alternatively spliced transcripts, N_s (full-length) and N_l (truncated), are generated from the single N gene and the ratio of these transcripts before and after TMV infection is critical to achieve complete resistance to TMV. However the biological role of these alternative transcripts in the function of the N gene, the nature of the corresponding N protein(s) and the interaction of the N protein and its corresponding viral elicitor has not yet been elucidated. In an attempt to follow-up these questions, we have cloned the N gene (tagged with an HA epitope) under the control of the N promoter in a binary vector and independent transgenics have been generated in tobacco and *Arabidopsis*. Western blots will be performed on plants (under conditions promoting the expression of the epitope-tagged version of the N protein) to monitor the generation and possible purification of full-length and truncated versions of the N protein. Efforts would be made to purify other proteins possibly important for the interaction of the N protein with the avirulence protein using co-immunoprecipitation. Immuonolocalization studies using HA-tagged antibodies would allow us to directly visualize and monitor the interaction of the N protein with other cellular components critical to the N gene-mediated signaling pathway. Experiments are underway to realize these objectives.

A6.3 Spatial organization and detoxification roles of thiol methyltransferases in relation to the glucosinolate-myrosinase defense system in plants

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Our group has described a novel class of plant enzymes — thiol methyltransferases

(TMTs) — that constitute a putative third component of the glucosinolate-myrosinase defense system in Brassicaceae. In the present work, the spatial organization of TMTs in relation to myrosinase in *Brassica* was determined using *in situ* hybridization, immunolocalization and myrosinase activity assay on tissue imprints. The TMT transcript and/or protein were localized primarily in the phloem and to a lesser extent in the epidermis, paralleling the localization of myrosinase. Two roles for TMTs have been postulated: detoxification of reactive thiols produced upon glucosinolate hydrolysis and production of volatiles that further defend the plant against pests. We found that transgenic plants, roots, mammalian cells and bacteria containing TMTs had greater tolerance to exogenously applied thiocyanate — a major glucosinolate hydrolysis product in *Brassica*. As TMTs can also methylate and volatilize halides, we tested the impact of TMT on halide detoxification and found that TMT-overexpression enhanced the tolerance of organisms to iodide ions. Results from ongoing experiments are consistent with the view that TMTs are part of the glucosinolate-based defense against pests and are involved in metabolic detoxification.

A6.4 The N-terminal trunk of plant cystatins determines their inhibitory specificity against cysteine proteinases

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Cystatins regulate cysteine proteinases in several physiological processes in plants, including organogenesis, storage protein turnover, programmed cell death, tolerance to abiotic stresses, and defense against herbivorous predators and pathogens. Here we engineered N-terminal truncated variants of cystatins from different plant families to assess the impact of the N-terminal trunk on the inhibition of cysteine proteinases. Current structural models suggest that amino acids surrounding a conserved glycine residue in the N-terminal trunk of animal cystatins have a strong impact on the inhibitor's specificity, but the role of the N-terminal region in plant cystatins still remains equivoqual. In agreement with earlier studies reporting that the N-terminal trunk is not essential for cereal cystatins to inhibit cysteine proteinases, truncated forms of different plant cystatins lacking 15 to 20 amino acids at the N terminus exhibited easily detectable, but moderate [and comparable] activity against the model cysteine proteinase, papain. In contrast, complete versions of the same cystatins strongly inhibited papain, with estimated $K_{i(app)}$ values ranging from 10^{-8} to 10^{-11} M. These observations, while confirming the ability of plant cystatins with no N-terminal trunk to inhibit cysteine proteinases, also point out the significant impact of this structural element on the inhibitor's specificity and efficiency against target enzymes. This conclusion is in line with the recently demonstrated occurrence of positively selected amino acid sites in the N-terminal trunk of *Le*CYS8, a cystatin unit from the 88-kDa tomato inhibitor, multicystatin (see accompanying display).

A6.5 IPTG-Controlled Gene Expression during Symbiosis of *Sinorhizobium meliloti* with Alfalfa

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The interaction of the bacterium *Sinorhizobium meliloti* with alfalfa is a model system for the study of root nodule symbioses. The availability of the *S. meliloti* genome sequence has led to ongoing functional genomics studies. In this context, we are particularly interested in determining the functions of each of the 78 members of the Short Chain Dehydrogenase / Reductase (SDR) protein family encoded in the *S. meliloti* genome. We aim to carry out phenotypic investigations under both free-living and symbiotic conditions. As part of this work, we require the ability to regulate the expression of individual genes. We have constructed a broad host range expression vector, pSA005, which generates N-terminal StrepII tag fusions. The tag is important for confirmation of production of full length protein product. Regulated gene expression is mediated by the *tac* promoter and *lacI*^{q}. We report here the control of gene expression from this vector during symbiosis, through the incorporation of IPTG directly in the plant nutrient solution.

A7. Fusarium I

A7.1 Assessment of artificial inoculation methods and deoxynivalenol levels in

barley lines representing various candidate sources of resistance to *Fusarium* Head Blight.

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Fusarium Head Blight (FHB) is one of the most devastating diseases affecting the production of barley and other cereal grains throughout Canada and the world. FHB infection results in drastic decreases in crop yield and severe reduction in grain quality. The most common species causing FHB in North America is *Fusarium graminearum* (Schwab). The fungus produces mycelical extensions that rapidly spread among the florets, leaving shrivelled kernels and floral pieces covered with a pink or white film of mycelium displaying elevated deoxynivalenol (DON) levels. 19 barley lines representing FHB candidate resistance and susceptible sources were point inoculated or spray inoculated in the greenhouse with 40,000M/ml of *F. graminearum* macroconidia. Following inoculation barley plants were kept at 24°C and 95% humidity for 3 days to favour disease establishment and were then returned to normal growing conditions at 21°C without humidity control (45%). 18 days post-inoculation heads were collected and spikelets displaying symptoms of FHB infection were rated for progression of the disease. These lines were also evaluated in the Brandon, MB nursery from 2000-2004. The number of discoloured spikelets produced by point inoculation, indoor spray inoculation, and disease and DON assessment were compared for each of the 19 lines. Barley lines representing candidate sources of FHB resistance or susceptible FHB sources showed varying degrees of symptoms due to fungal infection in the three tests. Barley lines representing more or intermediate FHB resistant sources showed consistent levels of resistance in the three tests. Artificial inoculation methods and DON quantification enable us to rate the level of resistance of the barley lines, with confidence. A higher level of FHB resistance will guarantee lower risks for the farmer associated with crop losses due to reduced grain yield, low quality grain, and DON contamination.

A7.2 Efficiency of cycloheximide and deoxynivalenol for *in vitro* screening for Fusarium Head Blight resistance in wheat

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Selection for resistance to Fusarium head blight (FHB) among wheat lines is difficult because of the environmental effects on the expression of resistance. Previous reports indicate that toxin accumulation is correlated with the level of FHB susceptibility, and that trichothecenes are associated with aggressiveness of *Fusarium graminearum*. Thus, *in vitro* selection techniques that employ purified pathogen toxins or culture filtrates for screening lines may provide many practical advantages. A study was conducted to evaluate cycloheximide and deoxynivalenol (DON) as potential screening tools for FHB resistance in wheat. Following field and greenhouse screening of 317 Iranian wheat varieties for FHB resistance, 7 lines selected for a seedling germination test in presence of DON at 10, 15 or 20 ppm, or cycloheximide at 3, 6, 9 or 12 ppm. A factorial design with 3 replications was employed and the percent germination at 5ºC and percentage of seeds with a coleoptile length greater than 2 mm, were recorded. Differences due to varieties and levels of DON and cycloheximide, were observed ($P<0.01$). DON, at 10 ppm, permitted detection of the greatest variability among the varieties and the correlation coefficient between field ratings for FHB resistance and percent germination in the presence of DON was 0.72 ($P. <0.01$). For cycloheximide at 3, 6, 9 ppm, the correlation between means of FHB resistance and tolerance to DON was 0.637 ($P <0.01$) and 0.474 ($P <0.05$) for percent germination and coleoptile extension, respectively. With further refinements of this technique, it may be possible to develop an efficient, in *vitro* screening technique for FHB resistance.

A7.3 First report of association between soybean sudden death syndrome (SDS), caused by *Fusarium solani* f.sp. *glycines*, and black walnut (*Juglans nigra*)

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Tree-based intercropping is an agroforestry technology for growing crops between trees in wide alleys. The University of Guelph Agroforestry Research Site (est. 1986) consists of 26 ha of crops interplanted with 2 ha of trees (12 species, including black walnut [*Juglans nigra* L.]) in 15 m wide alleys. Soybean sudden death syndrome (SDS) is caused by *Fusarium solani* f.sp. *glycines* (*Fsg*), and symptoms include severe root rot and interveinal leaf chlorosis/necrosis. SDS typically occurs as discrete patches of disease. The objective of this study was to compare the presence and distribution of SDS in a soybean-tree intercrop with a soybean monoculture. On 20-21 July (R2-R3 stage) and 9-10 August 2004 (R4-R5), the intercrop and monoculture were surveyed and assessed for the presence of patches of SDS. No patches were found in the monoculture. In the intercrop, 34 patches (total area of 128 m^2) and 66 patches (220 m^2) were found on each sampling date, respectively. Patches occurred exclusively within 2-5 m of black walnut trees. Seven isolates of *F. solani* and six of *F. oxysporum* were recovered from diseased roots and tested for pathogenicity. Isolates of *F. solani* produced disease severity scores of 2.5/5 for root rot and 6/100 for leaf damage, and *F. oxysporum* caused scores of 1/5 and 1/100. Koch's postulates were completed for the *F. solani* isolates, confirming the presence and association of *Fsg* with the SDS symptoms. A greenhouse assay for disease potential was completed with soil collected from SDS patches, in front of other tree species (e.g. non-walnut) in the intercrop, and in the monoculture. After 7 weeks (early R1) roots were assayed for the presence of *F. solani.* In soil collected from SDS patches 30% of the roots were infected, while less than 1% of plants were infected in the other treatments. The results suggest that allelopathic compounds produced by black-walnut, such as juglone, may pre-dispose soybean plants to *Fsg* infection.

A7.4 Potential mechanisms of biological control by *Gliocladium catenulatum* against fusarium root and stem rot of greenhouse cucumber.

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Fusarium oxysporum f. sp. *radicis-cucumerinum* D.J. Vakalounakis (*Forc*) causes fusarium root and stem rot of greenhouse cucumbers (*Cucumis sativus* L.). *Gliocladium catenulatum* Gilman and E. Abbot strain J1446, formulated as Prestop WP and Prestop Mix by Verdera Oy, Finland, has been shown to be an effective biocontrol agent against *Forc*. An understanding of the mechanisms of action of *G. catenulatum* is essential to maximize disease control efficacy. The ability of *G. catenulatum* to induce systemic resistance in cucumber plants was examined using a split-root assay in growth room trials. There was no significant difference between mortality or root fresh weight in plants treated with *Forc* only on one-half of the root system compared to plants treated with *Forc* on one-half + *G. catenulatum* applied to the other half of the root system. Compared to untreated controls, cucumber plants treated with *G. catenulatum* exhibited higher levels of ?-1,3-glucanase and chitinase in the roots, but not in the leaves, and only at 7 days post-application. Neither polyphenoloxidase nor peroxidase enzyme activity was increased in the roots or leaves at 3 or 7 days post-application. These results indicate that *G. catenulatum* does not likely induce defense responses in cucumber plants. When grown on medium containing chitin or *Forc* cell walls as the sole carbon source, *G. catenulatum* produced chitinase and ?-1,3-glucanase, but levels were significantly lower than *Trichoderma harzianum* T-22. When grown on laminarin, ?-1,3-glucanase production by *G. catenulatum* peaked at 3 days at levels significantly higher than *T. harzianum*, but enzyme levels rapidly declined after 7 days in culture. The production of ?-1,3-glucanase *in situ* may contribute to biological control efficacy, and the colonization ability of *G. catenulatum* on cucumber roots is likely also an important factor, which will be studied using a GUS-transformed strain.

A7.5 Predicting fusarium head blight and DON in spring wheat by quantifying spores per head following anthesis.

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In North America, *Gibberella zeae* (anamorph = *Fusarium graminearum*) is the principal cause of fusarium head blight (FHB) on small grain cereals. Although disease-forecasting models are available, a more accurate and reliable prediction model based on both weather conditions and levels of *Fusarium* inoculum is needed. This study was designed to quantify relationships among levels of *Fusarium* inoculum from wheat stubble, spores/head, deoxynivalenol (DON) levels in grains, and weather effects. The experiment was conducted in 17 producer fields sown with spring wheat cultivars, either AC-Barrie or AC-Superb, in Manitoba in 2004. Four sampling sites (0.5 × 0.5 m^2 each) were positioned in each field for measurement of colonies on stubble and heads, FHB index, percent fusarium-damaged kernels (%FDK) and DON levels in grains. Ten prototype spore traps were placed in each field and spores were collected between anthesis and two weeks following. Levels of *Fusarium* found in heads depended on both inoculum levels on stubble and weather conditions. There were strong relationships between spores/head and colonies/head (R^2=0.95), FHB index (R^2=0.88), %FDK (R^2=0.79), and DON (R^2=0.70). R^2 values of rainfall ~ FHB index, %FDK, and DON levels were 0.89, 0.86 and 0.81, respectively. Counting spores/head soon after anthesis appears to be a promising strategy for predicting FHB and DON levels in harvested wheat.

A7.6 Resistance of *Brassica napus* Cultivars to *Fusarium oxysporum* f. sp *conglutinans*.

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Fusarium wilt of canola (*Brassica napus* L.), caused by *Fusarium oxysporum* f. sp *conglutinans* (Wollenweber) Snyder & Hansen, is a disease that has recently been discovered in Western Canada. As such very little is known about the host-pathogen interaction for this disease. Since it appears that varieties differ in their susceptibility to this disease, one of the first priorities has been to determine the amount of genetic resistance currently available in commercial *B. napus* varieties and advanced breeding lines. To assess the reaction of *B. napus* to this pathogen, lines were artificially inoculated with pathogen spores at a concentration of 1 x 10^6 to 1 x 10^7 conidia/ml at the seedling stage using a root-dip method in a greenhouse setting. A water bath was used to regulate soil temperature at 26°C to provide optimal conditions for pathogen development. Initial results of 76 lines tested indicate that the majority of lines (59), to be resistant. Only relatively few (12), were rated as susceptible. The remaining five lines showed an intermediate value, which appears to be due to genetic segregation rather than partial resistance, as individual plants were in most cases either healthy or severely infected. Since most lines show strong resistance, control of this disease should be possible by the simple measure of avoiding susceptible cultivars.

B1. Nutrient use efficiency (part 2)

B1.1 Nitrogen Use Efficiency and Crop Improvement

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The desire to improve nitrogen use efficiency in crop plants is driven by the increasing cost of nitrogen fertilizer and by pressure to decrease environmental degradation caused by current farming practices. There are two ways to improve nitrogen use efficiency. One can either increase the yield of cultivars grown under current nitrogen fertilization protocols or one can maintain yield under decreased nitrogen regimes. The types of changes one might make in plant development and primary metabolism to develop crop genetics for the realization of these scenarios will be discussed as well as the issues around their commercialization. Experiments will be described where corn lines were grown under defined nitrogen conditions with the use of whole genome transcript profiling as a way to start defining the problem and possibilities. Finally, experiments using Arabidopsis as a model system for this trait in an attempt to decipher some of the regulatory genes involved will be described.

B1.4 Ionomic profiling of alfalfa varieties to enhance nutrient quality of cattle feeds.

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Availability of high quality, nutrient-balanced feeds is essential to reducing the cost of cattle production and to maintaining healthy animals. We have used ionomic profiling to quantify genotypic variation in shoot elemental concentrations for 11 alfalfa (*Medicago sativa*) varieties. Alfalfa shoot samples were collected in June 2004 from the Lacombe 2001 Western Forage Testing System trial and were analysed by ICP-MS for 16 elements essential for plant and animal growth. Most plant macronutrients (Ca, K, Mg, P, S) showed low varietal variation (6–15% RSD), whereas several trace elements exhibited greater variation (e.g. Cr 28%, Na 54%). Multivariate analysis of the ionomic profile dataset identified varieties with altered potential for accumulation of several elements (Co, Cr, Fe, Na). These preliminary results demonstrate the potential of this approach to identify nutrient-dense varieties. A systematic and wide ranging assessment of inorganic nutrient quality in forage crops is warranted.

B1.5 Transcript profiling of *Brassica napus* exposed to long-term phosphate stress

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Global plant productivity is restricted by a limited supply of phosphorus, an essential plant macronutrient. By identifying genes that exhibit differential expression under phosphate stress, plants with enhanced phosphate-use efficiency can be developed. Hydroponically grown *Brassica napus* was exposed to a long-term phosphate depletion, deficiency, and recovery regime. Root and shoot tissue was harvested throughout the experiment for physiological and genomic analysis. *Arabidopsis thaliana* microarray slides probed with isolated *B. napus* RNA revealed approximately 1000 genes in the roots and 2600 genes in the shoots with statistically significant up- or down-regulation over the entire stress period. Interestingly, only a small subset of these genes was found in both roots and shoots, indicating tissue specific expression patterns. One known response of phosphate-limited *B. napus* is enhanced root exudation of organic acids to increase solubilization of soil inorganic phosphate. In support of this, the transcript profiles of the shoot and root tissue show up- and down-regulation of key enzymes in glycolysis and the TCA cycle. This suggests allocation of carbon through these pathways is increased to generate additional citrate and/or malate for excretion by the roots.

B2. Contemporary issues in statistics, data management, plant biology and agricultural research (part 2)

B2.1 Multivariate analysis of agronomic data

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Multivariate data sets arise whenever many (more than two) variables are measured on each individual or object. While such data are commonly encountered in both agronomic surveys and experiments, the need to analyze such data using multivariate analysis is not always recognized nor appreciated. Indeed, many scientists view multivariate analysis with suspicion, and/or dismiss the approach as unnecessarily complicated. These attitudes reflect in part a lack of understanding of the statistical foundations of multivariate analysis – while virtually all scientists are exposed to univariate statistics, courses in multivariate analysis remain rare. In addition, potential users are often frustrated by the confusing and seemingly contradictory recommendations in the literature regarding multivariate methodologies, strategies and approaches. This presentation is based on my experiences, over two decades, as a multivariate analysis consultant to graduate students and colleagues, and as an assistant editor on numerous ecological journals. These two decades are characterized by a precipitous increase in the use of multivariate analysis in the agricultural and biological disciplines. However, my experiences indicate that analytical methods are often misapplied, and that users often employ sub-optimal multivariate approaches. The objective of my presentation is to dispel some of the

"mysteries" of multivariate analysis, and to discuss problems and challenges related to the selection of ordination, classification and canonical methods, variable standardization, data transformation, and resemblance measure selection. In particular, I dispel the notion of a single "best approach" to multivariate analysis. Because each study – and the resultant data – is unique, great care must be taken in selecting a methodology, strategy and approach best suited to the data and study objectives. Only by doing so will a correct and optimal multivariate analysis be obtained.

B2.3 Spatial variability of soil and the scaling problem

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Most fields are characterised by the presence of significant spatial variability of soil properties. The spatial variance is often dependent on the spatial scale of sampling. At a local (point) scale, soil properties can influence crop yield and many soil processes of interest, but the relationships are often highly non-linear. Thus, the prediction of average values or relationships (crop yield or soil processes versus soil properties) at large scales from local scale values and relationships is not trivial. Average values at larger scales will depend not only on average soil conditions, but also on the amount of variability (and spatial covariance) of major soil properties. The paper reviews methods of characterising the scale dependence of soil variability, and a stochastic framework for examining and predicting the influence of spatial variability and covariance of soil properties on the spatial scale dependence of soil processes and/or crop yield response is presented. Examples are given using crop yield response to fertilizer and greenhouse gas (nitrous oxide) flux from soil.

B3. Systematics and evolution II

B3.1 The evolutionary origins of *Erigeron trifidus*

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Systematic relationships within the genus *Erigeron* (Asteraceae: Astereae) remain obscure due to complex geographical distribution patterns, polyploidy, hybridization and agamospermy. *Erigeron trifidus* Hook. is restricted to three disjunct alpine regions of the Alberta Rocky Mountains and is a designated rare plant in the province. *Erigeron trifidus* was originally treated as a variety of E. compositus until Packer (1981), based on morphological, cytological and habitat differences, proposed that it be treated as a distinct species and that it had arisen via hybridization between *E. compositus* and *E. lanatus*. A restriction site analysis of cpDNA from the three species throughout the range of *E. trifidus* revealed 4 haplotypes. In most cases cpDNA haplotypes in *E. trifidus* were the same as in *E. lanatus*, suggesting *E. lanatus* as the maternal parent in the majority of crosses. An analysis of the nuclear ETS region further supports the hybrid origin hypothesis in that *E. trifidus* contains only repeat types present in one or the other of the putative parents. In addition, *E. trifidus* displays a higher percentage of within individual repeat type polymorphism than either putative parent. ETS and morphological data also reveal a north/south geographic pattern that separates *E. trifidus* into two groups, possibly indicating a separate origin for southern *E. trifidus*. Finally, both chloroplast and ETS data sets show a homogenous pattern of genetic variation for *E. trifidus* across the northern region despite spatial variation for these markers in the parental species. This suggests that northern *E. trifidus* originated through a single hybridization event and that colonization of the various areas within the northern region occurred post origin.

B3.2 Sequence heterogeneity of the envelope-like domain in the Egyptian cotton *Gossypium barbadense*

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The current study aimed to investigate the evolution of *envelope*-like sequences in the Egyptian cotton *G. barbadense*. DNA sequence determination and analysis of *envelope*-like sequences revealed that these sequences are heterogeneous in *G. barbadense*. The observed sequence diversity, however, seems to preserve the coding information. Phylogenetic analysis

demonstrated that plant *envelope*-like sequences group together, suggesting their monophyletic origin. *Gossypium envelope*-like sequences are, however, more closely related to elements present in other plant species. These results suggest that *envelope*-like sequences in cotton have evolved under functional constraint and likely to play a role in the life cycle of these elements.

B3.3 Phylogeny of *Populus* (Salicaceae) based on Sequence Characterised Inter Simple Sequence Repeat (SCISSR) data.

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The genus *Populus* comprises over 29 species classified into six sections and represents some of the most commercially exploited forest trees distributed throughout the Northern-hemisphere. Recent phylogenetic analyses of *Populus* based on molecular (chloroplast DNA and nuclear rDNA sequence) and morphological data showed limited resolution suggesting close evolutionary relationships among species. Since the variability of most "traditional" gene sequences used in phylogeny reconstruction were insufficient to resolve phylogenetic relationships among closely related species, we chose highly variable DNA sequences in the genomic regions flanked by microsatellites or inter simple sequence repeat regions to reconstruct the phylogeny of *Populus*. The resulting phylogenetic trees were congruent with existing phylogenetic trees of poplars but the resolution was higher. The species of section *Populus* were monophyletic, while polyphyletic relationships among species in the sections *Tacamahaca* and *Aigeiros* were observed. In contrast to previous phylogenetic studies of *Populus*, SCISSR based phylogeny revealed a close relationship between *P. nigra* and species of section *Tacamahca*. This relationship is further supported by various phenotypic data including disease susceptibility, interfertility and bud exudates suggesting introgressions between *P. nigra* and species of section *Tacamahaca*. Due to high variability and wider genome coverage, SCISSRs are valuable as a tool for resolving phylogenetic relationships among closely related taxa as well as for the detection of introgression and reticulate evolutionary events.

B3.4 Fagaceae Fruits from the Eocene of Vancouver Island, BC

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The Beech family (Fagaceae) has a rich Tertiary fossil record. Compressed leaves are common, but fruits are much rarer. Because fruits have taxonomically important characters that often delimit genera within the family, fossil forms are very informative. For this reason, the recent discovery of three distinct fruit types from the Eocene Appian Way fossil locality on Vancouver Island are important in our interpretation of reproductive morphology and distribution within the family through time. The fruits occur in calcareous nodules that preserve anatomy and morphology in three dimensions. Fruits are studied anatomically using the cellulose acetate peel technique, and AMIRA 3.1 visualization software to reconstruct external forms. Three types of Fagaceae fruits have been identified. The first is enclosed in a spiny, indehiscent cupule that bears a single nut at maturity. The second is also cupulate, but has fewer spines and an elongate nut. Both of these taxa show strong affinities to the genus *Castanopsis*, now endemic to Southeast Asia and are contemporaneous with the earlist known fossils of the genus. The third fruit is a triangular nut with axile placentation and six ovules and shows strongest affinities to extant *Fagus*, and could represent the earliest anatomically preserved occurrence of the genus. Taken together, the Fagaceae fruits from Appian Way have a combination of features that evidence a structural stasis among some characters going back to the Eocene. Other features suggest ancestral states that differ from those known in extant taxa.

B3.5 Permineralized monocots from the Middle Eocene Princeton Chert

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Monocots are rarely preserved in the fossil record, due partly to their occurrence in ephemeral habitats, and partly to their often

herbaceous habit. Given this, when any fossil monocots are found they have the potential to greatly contribute to our understanding of this group. Among the dicots, ferns, fungi and conifers that are found at the Middle Eocene Princeton Chert locality (Allenby Fm), numerous monocot remains have been identified. Specimens from the Princeton Chert have exceptional cellular detail preserved, allowing for detailed anatomical descriptions of taxa, and in some cases whole-plant reconstructions. Many of these fossil monocots represent vegetative organs of plants. To date, five monocots have been formally described from this locality: stems with attached leaves and roots of *Soleredera rhizomorpha* (Liliales) and *Ethela sargantiana* (Cyperaceae/Juncaceae); a coryphoid palm, *Uhlia allenbyensis* (Arecaceae), known from stems with attached leaves and roots; petioles of *Heleophyton helobiaeoides* (Alismataceae); and seeds of *Keratosperma allenbyense* (Araceae). In addition, there are several undescribed taxa: flowers and fruits with affinities to Aponogetonaceae and Potamogetonaceae (Alismatales), and two types of rhizomes that have yet to be allied with any living group. Many of the extant taxa in these groups have a preference for aquatic habitats, and the anatomical characters of the fossils support the idea that the Princeton Chert represents an in situ aquatic depositional environment. The diversity preserved by the Princeton Chert shows an aspect rarely seen in palaeoecosystems, the preservation of herbaceous monocots, and helps to provide a clear picture of this Middle Eocene wetland. As more work is done on comparative anatomy of monocots, these fossils can be better placed in a phylogenetic context, allowing us to better determine patterns of character evolution within the group.

B3.6 Comparative genomics to assess gene age and horizontal gene transfer

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With the advent of high throughput genomic sequencing and the availability of complete genomes, we can examine relationships between organisms using thousands of sequences. Among eukaryotes, fungi have some of the smallest genome sizes and hence represent the highest number of eukaryotic genomes sequenced, almost all within the last five years. Comparisons between fungal genomes have revealed a common set of fungal sequences, many of which are also conserved in plants, animals or bacteria. By using evolutionary timelines based on geological evidence or molecular clocks, the age of different gene sets can be estimated. In this study, the origins of particular gene sets were characterized as dating from the split between eukaryotes and prokaryotes, the radiation of the major eukaryotic taxa (plants, animals and fungi), or the divergence of fungal divisions (ascomycota and basidiomycota). This method of gene similarity comparisons does not exclude the possibility that genes may have been transferred between species since their divergence, and there is evidence for horizontal gene transfer between bacteria and their animal or plant hosts. Hence an examination of horizontal transfer was done by comparing a genome of a fungal plant pathogen with its plant host, as well as with other fungal genomes to see whether particular sequences are shared only between the pathogen and host, and not found in close relatives of the pathogen or other organisms. The results of this study involving gene age and horizontal gene transfer are discussed in this presentation.

B4. Development and plant structure

B4.1 A role for ethylene in trichome branching

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The single celled trichomes of *Arabidopsis thaliana* are used to model several aspects of cell development. We have been studying the effect of both Gain Of Function (GOF) and Loss Of Function (LOF) mutations to the 5 *Arabidopsis* ethylene receptors on the branching of trichomes. Wild type (Col.) *Arabidopsis* trichomes have three or four branches, as did most of the ethylene GOF and LOF receptor mutants. The two exceptions are *etr2-3*, a LOF mutant which had unbranched trichomes, and *ers1-1,* a GOF mutant which had two and three branched

trichomes. These results indicate different roles for the ethylene receptors in control of cell development. The effect of differential ethylene signal transduction on cytoskeletal architecture, hormone treatment and overall trichome morphology will be presented.

B4.2 A structural analysis of peridial development in *Aporothielavia leptoderma* has ecological and taxonomic significance.

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The closed ascocarps of cleistothecial ascomycetes display a variety of structural modifications that lead to ascospore release. One of these variants is represented by the cephalothecoid forms which have a cleistothecial wall made up of small disarticulating plates. As part of a larger study of ascocarp spore release mechanisms of cephalothecoid fungi we examined the ultrastructural features of the peridium of *Aporothielavia leptoderma* (Booth) Malloch and Cain. The peridium of *A. leptoderma* develops through branching and tip growth of peridial hyphae, and the outer peridial layer is constructed of cells with uniformly thickened cell walls. Peridial plates are not visible during the development of the cleistothecium and are only observable when the mature peridium dries and begins to rupture. Unlike other cephalothecoid species, *A. leptoderma* does not possess a mechanism that allows for the complete rupture of the peridium when dried. This indicates that alternate external inputs of energy are required for spore liberation. In addition to the potential ecological significance, these observations indicate that this species would be more satisfactorily disposed in the genus *Chaetomidium*, a genus that is similar in having a cephalothecoid peridium, evanescent asci, and single celled ascospores bearing a germ pore.

B4.3 Arabidopsis RACK1 mediates multiple developmental processes and acts with the heterotrimeric G-protein complex to positively regulate lateral root initiation and development

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The mammalian scaffold protein RACK1 (receptor for activated C kinase 1) belongs to a subgroup of the large family of WD40 repeat proteins including the heterotrimeric G-protein β subunit. RACK1 serves as an integrative point for many signals, playing a critical role in diverse signal transduction pathways. The Arabidopsis genome contains three RACK1 orthologs, however, little is known about their functions. I report here that one member of this gene family, *RACK1A*, mediates multiple developmental processes such as hypocotyl elongation and cotyledon formation in young seedlings, and leaf and root production in mature Arabidopsis plants. *RACK1A* preferentially expresses in the meristems of the shoot, and primary and lateral roots. A transcript-null mutation of *rack1a* results in decreases in lateral root formation. *RACK1A* is auxin inducible. Loss of *RACK1A* confers reduced sensitivity to auxin in adventitious and lateral root formation, suggesting that RACK1A positively regulates auxin-induced cell division in roots. The altered auxin sensitivity in *rack1a* mutant is similar to that in the null mutant of Arabidopsis heterotrimeric G-protein α subunit, *gpa1*, but opposite to that in the null mutant of Arabidopsis heterotrimeric G-protein β subunit, *agb1*. Genetic analyses revealed that RACK1a acts together with the heterotrimeric G-protein complex to regulate lateral root initiation and development. In comparison with *gpa1* and *rack1a* single mutants, lateral root formation in the *rack1a gpa1* double mutants was synergistically reduced. On the other hand, RACK1a acts as a suppressor for AGB1, because loss of *RACK1A* suppressed the excessive lateral root production in the *agb1* mutants. These results suggest that RACK1a and the heterotrimeric G-proteins may constitute a signaling complex to regulate lateral root formation.

B4.4 Chemistry and Structure of Soybean Root Epidermis

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The root epidermis is the outermost cell layer and, as such, it interacts with the soil environment. This layer is important as the first site of defence against harmful microorganisms. For soybean cultivars, it remains unknown whether there are any structural and chemical differences in the

epidermal cell walls, and how they may relate to resistance against soybean root rot pathogen (*Phytophthora sojae*). Results of histochemical and electron microscopical studies indicated that suberin may be present in the walls. Epidermal cell walls were isolated enzymatically and subjected to two different degradation methods, i.e. BF_3/MeOH transesterification and nitrobenzene oxidation. The compositions of depolymerisates of the cell walls determined by GC/MS indicated four dominant suberin monomers varying in chain length from C_{16} to C_{24}. In all epidermal cell walls, ?-hydroxycarboxylic acids were more abundant than diacids, carboxylic acids and alcohols. Two of the monomers detected (hydroxycarboxylic acid and a, ?-dicarboxylic acid) are known to be characteristic suberin markers. The quantitative chemical compositions significantly differed in epidermal cell walls of two soybean varieties. Walls of a resistant cultivar (Conrad) had almost twice as much aliphatic suberin as walls of a susceptible cultivar (OX760-6). This evidence supports the hypothesis that suberin in the epidermal walls of soybean roots could play a role in their resistance to soybean root rot.

B4.5 Intercropping barley, triticale and fababeans at Breton, Alberta.

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Intercrops of cereals and pulses have the potential to increase silage yield and protein content, and to decrease N fertilizer requirements compared to the same crops grown in monocultures. Barley, triticale and fababeans were grown alone and in mixtures on a Gray Luvisolic loam at Breton, Alberta in 2002, 2003 and 2004. Experiments assessed biomass yield and protein concentration at silage-stage of barley-triticale-fababeans intercrops compared to monocrops. The effects of seeding fababeans prior to cereals, changing the within-row and between-row seeding arrangement, and adding nitrogen fertilizer were also assessed. Drought conditions in 2002 and 2003 greatly reduced fababean biomass, producing dry matter yields of 2-3 Mg ha^{-1} for fababeans grown as monocrops. Dry matter yields of intercrops ranged from 3-8 Mg ha^{-1} and usually did not provide a yield advantage compared to monocrops of triticale or barley. With normal rainfall in 2004, one barley-triticale-fababeans intercrop treatment produced a greater biomass yield than a barley monocrop. Protein concentrations of whole plants at silage-stage were 14-19% for fababean and 6-9% for barley and triticale. The addition of nitrogen fertilizer at a rate of 100 kg N ha^{-1} decreased the fababean component of intercrops. Seeding the fababeans prior to cereals increased the fababean component of intercrops. In 2003, intercrops with fababeans seeded in separate rows had greater yields than intercrops with fababeans and cereals seeded in the same row. In 2004, intercrop yields did not differ with fababeans seeded in separate rows versus in same rows with cereals. Further research under conditions of normal rainfall is needed to adequately assess the potential of intercropping fababeans with barley and triticale for forage in west-central Alberta.

B4.6 Plant cell architecture - The structural role of actin filaments and microtubules in anisotropic cellular growth

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From a mechanical point of view plant cell growth is dominated by the equilibrium between the internal turgor pressure and the deformability of the cell wall. In plant cells, forces created by the polymerization of cytoskeletal elements have therefore frequently been assumed to be negligible compared to those established by cell wall and turgor. However, the equilibrium between these two dominant features is very delicate in tip growing cells such as pollen tubes, since growth is extremely fast and the continuous addition of new cell wall material plays into the equation. Furthermore, the tip growth process has an important mechanical aspect as tip growing cells are able to exert considerable forces thus allowing them to penetrate the surrounding matrix.

The delicate equilibrium governing the growth process indicates that forces created by the cytoskeleton are likely to be more evident

in tip growth than in other types of plant cell growth. It is well established that the actin cytoskeleton is responsible for the continuous stream of secretory vesicles to the growing pollen tube apex and thus plays a pivotal role for the supply of cell wall precursors necessary for apical expansion. However, low concentrations of drugs inhibiting actin polymerization such as cytochalasin D are able to arrest pollen tube growth without preventing cytoplasmic streaming. This finding together with the presence of a conspicuous subapical mesh of fine actin filaments indicates that actin might have a more direct, mechanical function in the pollen tube growth process. The role of microtubules in pollen tube growth is even more elusive, since their depolymerization does not inhibit pollen tube growth. We therefore assessed the role of both cytoskeletal elements in the mechanics of cellular architecture and growth processes in pollen tubes. To do so we developed an assay that allowed us to compare penetration capacity and thus determine the pollen tubes' growth force. Our results indicate that both microtubules and actin filaments play a role in the mechanics of pollen tube growth thus making this cell an excellent model system for the investigation of the structural role of the cytoskeleton in the mechanics of plant cell growth.

B5. Molecular responses to abiotic stress

B5.1 RNA secondary structure functions as a thermosensor

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Cyanobacteria survive at a diverse range of temperatures due to the induction of a specific set of cold shock proteins. Our lab has identified a DEAD-box RNA helicase, CrhC, from the cyanobacterium, *Anabaena* sp. strain PCC 7120, whose expression is regulated by temperature. CrhC expression is upregulated at both the transcript and protein levels in response to cold shock (20^{o}C), suggesting a role in adaptation to reduced growth temperature. The mechanism(s), by which the temperature-dependent expression of *crhC* is regulated, are not known.

At the transcriptional level, EMSA and DNA affinity chromatography analysis identified a 60 kDa putative repressor that bound to a 52 bp region within the *crhC* promoter. The binding region encompasses an AT-rich regulatory element that was shown to be important for transcriptional regulation. Phosphorylation studies showed that DNA binding is phosphorylation dependent, which suggests regulation by a cold-induced signal transduction pathway.

At the post-transcriptional level, temperature regulated expression is conveyed solely by the 5' UTR when *crhC* is expressed from a constitutive promoter in a heterologous system. Analysis of the 5' UTR secondary structure using MFOLD and *lux* reporter fusion constructs identified two stem-loop structures required for temperature-dependant *crhC* expression. Following transfer from 30^{o}C to 24^{o}C, MFOLD predicted a thermodynamic increase in the secondary structure of the 5' loop. Preliminary site-direct mutagenesis of the 5' UTR suggests that both loops are required for temperature-dependent expression. The secondary structure of the 5' UTR therefore appears to function as a thermosensor.

B5.2 Multiple heat signaling pathways in tobacco cells

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The nature of early events occurring during heat shock response in tobacco cells was examined. According to a well-accepted model for heat signaling, heat shock denatures cellular proteins and the denatured proteins are then sensed by an unknown mechanism to activate heat shock transcription factor(s) (HSF). The latter then activates *hsp* genes resulting in the synthesis of heat shock proteins. Previous work in our laboratory has shown that heat shock rapidly activates HAMK (Heat-shock Activated MAP Kinase) and that heat activation of HAMK is mediated by membrane fluidization (Sangwan et al., 2002, Plant Journal 31: 629-638). In this presentation, we will show that heat shock activation of HAMK and HSP70 expression are regulated by membrane fluidity through two separate pathways. In pathway-1, membrane fluidization leads to the activation of the mechanosensitive enzyme phospholipase C

which triggers lipid signaling through diacylglycerol (DAG) and inositol triphosphate (IP_3), phosphatidic acid (PA) and release of internal Ca^{2+}. In pathway-2, membrane fluidization leads to cytoskeleton remodeling, opening of Ca^{2+} channels, influx of cell wall Ca^{2+} and activation of Ca^{2+}-dependent protein kinases. Both pathways converge and activate HAMK and heat shock protein expression.

In pathway-1, heat activation of HAMK and HSP70 expression are inhibited by the inhibitors of PLC, as well as the inhibitors of DAG kinase which convert DAG to phosphatidic acid. However, the pathway-1 is mimicked at 25°C by treatment of cells with either PA, IP_3 or cADPR (cADP-ribose, releases calcium from internal stores).

In pathway-2, the heat activation of HAMK and HSP70 accumulation is prevented by membrane rigidifiers, cytoskeleton stabilizers and Ca^{2+} chelators and Ca^{2+} channel blockers. Pathway-2 is mimicked at 25°C by membrane fluidizers, cytoskeleton destabilizers and Ca^{2+} channel agonists and Ca^{2+} ionophore. It appears that heat shock response requires both pathways. The possible relationships of these findings to the published models will be discussed.

B5.3 Characterization of a novel stress-induced member of the GRAS family in tobacco.

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Development and survival of plants depends on reliable signaling mechanisms providing information on the actual state of the surrounding environment. The multilevel interaction of these mechanisms forms a regulatory network that allows plants to respond effectively to the changes in environment often through changes in gene expression. We have recently isolated a gene from tobacco that may help in our understanding of the link between environmental stress and altered gene expression. Based upon homology, this gene is a member of the plant specific GRAS family of transcription regulators. Denoted *NtGras1* the tentative protein product possesses all the characteristics of GRAS members including: conserved leucine heptad repeats (LHR I and LHR II), VHIID, PFYRE and SAW motifs. Changes in *NtGras1* expression were followed in *Nicotiana tabacum* Bright Yellow-2 cultured cells. Using RNA blot analysis we found that expression of *NtGras1* is barely detectable in control cells but strongly induced in response to treating the cells with antimycin A, hydrogen peroxide, salicylic acid, and cysteine. To determine the intracellular localization of the protein we fused the coding region of *NtGras1* with green fluorescent protein (*NtGras1-GFP*) and used particle bombardment with onion epidermal cells. Using fluorescence microscopy it is clear that NtGRAS1 is localized within the nucleus of the plant cell. Taken together these results suggest that *NtGras1* may be a key regulator of transcription under a diverse array of environmental stress conditions.

B5.4 A novel stress-regulated protein interacts with the transcriptional activator GCN5 in *Arabidopsis thaliana*

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An *Arabidopsis* protein, AtEMBL, was isolated based on its interaction with histone acetyltransferase (HAT) GCN5 in a yeast two-hybrid screen. RNA blot and quantitative RT-PCR analysis show that *AtEMBL* is expressed in flowers, leaves, stems and siliques, and the expression of the *Brassica napus* orthologs of *AtEMBL*, *BnEMBLs*, is developmentally regulated in siliques. Both *AtEMBL* and *BnEMBL3* are regulated by biotic stresses (flea beetle feeding and fungal infection) and abiotic stresses (cold, salt, salicylic acid and jasmonic acid). *In vitro* and *in vivo* protein-protein interaction assays indicate that AtEMBL interacts with GCN5 through the N-terminal region. Furthermore, AtEMBL activates expression of the *lac*Z reporter gene in yeast through recruitment of GCN5. Such recruitment is accompanied by an increase in histone H3 acetylation in the promoter driving *lac*Z expression, as determined by chromatin immunoprecipitation (ChIP). A higher level of *AtEMBL* gene expression is detected in the *gcn5* konckout mutant as compared to wild type *Arabidopsis*, indicating a functional association between GCN5 and AtEMBL. We conclude that AtEMBL is a stress response-regulated transcription factor that functions by recruiting GCN5 to target promoters to regulate the expression of associated genes.

B5.5 Cellular responses to aluminum stress: Genomics and Proteomics approach

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Aluminum (Al) is the most abundant metal in the earth's crust. When soil pH drops below 5.0, Al is solubilized to $Al3^{+}$. This toxic form of Al is the major limiting factor for plants growing in acidic soils. In this study we report both genomics and proteomics based approaches to identify Al- responsive genes. Arabidopsis plants were grown hyroponically and exposed to 25 µM $AlCl_3$ in an exposure medium containing 200 µM $CaCl_2$ at pH 4.3. The two physiological parameters used to determine the appropriate Al exposure concentration were the Relative Root Growth Increment (RRGI) and leaf anthocyanin content. Two time points, 6hrs and 48 hrs, were selected to find early and late Al responsive genes. Root tissue was harvested and distributed equally between total RNA and protein extraction for subsequent genomics and proteomic studies. 3DNA *Array 50* (Genisphere) methodology was used to label and hybridize RNA for microarray studies and the Ettan™ DIGE system (Amersham) was used for protein abundance analysis based on 2-D Fluorescence Difference Gel Electrophoresis (2-D DIGE).

Statistical analysis of microarray data indicated differential expression of nearly hundreds of genes for both time points. Statistically significant genes that also showed more than two fold change in their expression levels, were selected for further analysis. Our microarray data indicated that the responses at plant genome level occur rapidly during the initial phase of stress. Various transcription factors, splicing factors, and other DNA binding proteins were identified as Al responsive in this study. Many Al responsive genes identified in this study were found to be common to other abiotic stresses, supporting the existence of common pathways for abiotic stress tolerance.

B5.6 Promoter analysis in transient assay using anthocyanin reporter gene constructs in wheat (*Triticum aestivum* L.)

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Three barley tissue-specific promoters originating from genes encoding dehydrin (*Dhn12*), trypsin inhibitor BTI-CMe (*Itr1*) and lipid transfer protein (*Ltp1*) were transcriptionally fused to the *C1* and/or the *Bperu* gene(s) involved in the anthocyanin biosynthesis. Tissue-specific transient expression of these constructs was examined in different wheat tissues including stem, leaf, glumes, lemma, palea, anther, carpel, embryo and endosperm. The intensity of anthocyanin pigmentation was quantified using the Vision Lite 3.5 image analysis program. All constructs employing these three tissue-specific promoters only expressed anthocyanin in the scutellum and germ portions of the embryo. The three promoters were more effective than the *CaMV35S* promoter in directing anthocyanin production in the wheat embryo. *Ltp1* was stronger than *Itr1* and *Dhn12* in combination with *C1* and/or *B-peru* gene(s). Overall, the color intensity of anthocyanin was greater when the two genes where combined in a single construct, while individually, *C1* was stronger than *B-peru*. Analysis of nucleotide sequences of all three tissue-specific promoters revealed the presence of *ACGT* core sequence of *G*-box (*CACGTG*), *E*-box (*CANNTG*) and *RY* (*CCATGNN*) elements, that may be responsible for directing the embryo specific expression of anthocyanin in wheat.

B6. Gene expression in pathogenesis

B6.1 Analysis of the cell wall and secreted proteomes of *Sclerotinia sclerotiorum.*

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Sclerotinia sclerotiorum is a non-host-specific necrotroph capable of producing significant yield losses in a number of crops including canola, and a more thorough understanding of the proteome-level pathogenicity or virulence factors may offer novel targets for the development of disease resistance. The secreted and cell wall proteomes of the phytopathogenic fungus *S. sclerotiorum* were analyzed using 2-dimensional electrophoresis (2DE) and mass spectrometry (MS) in order to identify proteins that are necessary for disease induction and/or progression. We have identified a number of cell wall and secreted proteins, some of which are known to be

involved in producing disease symptoms in canola (e.g. endopolygalacturonase and acid protease). We are currently in the process of validating the potential roles of some of those identified proteins that have not been previously implicated in disease induction.

B6.2 Bacterial phytotoxin acts in systemic activation of the plant antimicrobial compound

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Plants attract a variety of intruders, from viruses, bacteria and fungi to insects. To protect themselves, plants have an armoury of weapons expressed constitutively or in response to a pathogen's presence, including the release of antimicrobial compounds. The phytoalexins are inducible chemical defences produced de novo by plants in response to diverse forms of stress, including microbial attack and elicitors. We show here a systemic activation of plant antimicrobial compounds by the bacterial phytotoxin, thaxtomin A, of the phytopathogen *Streptomyces scabei* that is associated with the transport of the toxin throughout the plant. This is, to our knowledge, the first report showing that a phytotoxin is systemically transported and activates the accumulation of the antimicrobial phytoalexin coumpound.

B6.3 Expression Analysis of a Lipase Gene from *Fusarium graminearum*

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A lipase gene was amplified from *Fusarium graminearum* strain PH-1 by polymerase chain reaction (PCR) and temporally named *FgLip1*. It codes for a predicted 591-amino-acid extracellular protein with a highly conserved *GXSXG* lipase-specific catalytic motifs and a predicted molecular weight of 63kDa. *FgLip1* exhibited 53% and 40-44% identity to *Botryotinia fukeliana* lipase and five *Candida rugosa* lipase precursors, respectively. Expression of *FgLip1* is specifically induced *in planta* and by starvation, wheat germ oil, and saturated fatty acid. The induction of expression by saturated fatty acid can be suppressed by the presence of unsaturated fatty acid. To test the potential function of *FgLip1* on the pathogenicity of *F. graminearum,* gene replacement transformation was conducted on PH-1. Strains with positive gene replacement were confirmed by Southern and Northern analyses. Pathogenicity test on a susceptible spring wheat cultivar showed no difference between gene replacement strains and the wild type, suggesting *FgLip1* is not essential for fungal pathogenicity.

B6.4 Gene expression analysis during mycoparasitism.

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In order to gain an in-depth understanding of the mycoparsitic process between the mycoparasite and its host at the gene level, suppressive subtracted hybridization (SSH) experiments were conducted coupled with microarray analysis to identify differentially expressed genes. Briefly, total RNA isolated from the interacting zone of the mycoparsite, *Stachybotrys elegans*, and its host *Rhizoctonia solani* were used as tester, and RNA from the two fungi growing alone were used as driver. A differential screening experiment was performed by printing the tester cDNA library on a microchip (2166 cDNA fragments in triplicate). To ensure the quality of the subtracted library, the microchip was hybridized with different ribosomal DNA from the two organisms. Interestingly, only 2.4% (52/2166) gave a signal. Considering that total RNA was the starting material, this result confirms the power of the SSH technique. . Hybridization experiments of the microchips with subtracted cDNA library of fungi when they are not interacting (subtracted driver)

showed that 174 expressed sequence tags (ESTs) were up-regulated during mycoparasitism, out of which 86% (149 ESTs) belonged to *S. elegans*, 10% (18 ESTs) belonged to *R. solani*, and 4% (7 ESTs) were difficult to identify. The possible function of these genes during mycoparasitism will be discussed. The expression of some of the up-regulated ESTs will be analyzed by quantitative Real-time RT-PCR in order to confirm their differential and temporal expression during mycoparasitism.

B6.5 Transcriptome analysis of *Colletotrichum coccodes*/*Abutilon theophrasti* pathogenic interaction.

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A Suppression Subtractive Hybridization (SSH) cDNA library was generated to gain an in-depth knowledge of the molecular determinants underlying host-specificity in the pathogenic interaction between the biological control agent *Colletotrichum coccodes* (DAOM 183088) known as Velgo® and its hostweed velvetleaf (*Abutilon theophrasti*). By using cDNA from the *C. coccodes*/velvetleaf pathogenic interaction as a tester and several driver cDNA populations (velvetleaf, *C. coccodes*, and *C. coccodes*/okra non-pathogenic interaction), the final number of expressed sequenced tags (ESTs) in the forward subtracted library was narrowed down to 975. Micro-array analysis is currently underway in order to screen the library with targets generated during the SSH process, identify ESTs corresponding to genes that are exclusive to the *C. coccodes*/*velvetleaf* interaction, as well as identify the source organism for each one of the ESTs. Preliminary results identified up-regulated genes, the majority (63%) of which originates from vlevetleaf. Only 2.8% of the ESTs gave positive signals when hybridized with plant and fungal ribosomal fragments, an indicative of good quality of the SSH library. Sequencing will be performed to identify the putative function of the isolated sequences. In addition, quantitative real-time PCR will be conducted to evaluate their temporal expression during the biocontrol-weed interaction under greenhouse conditions. The outcome of this study will lead to significant knowledge that could be applied to plant/biocontrol genetic engineering.

B6.6 US-1 and US-8 genotypes of *Phytophthora infestans* differentially induced the expression of genes encoding pathogenesis-related proteins PR-2, PR-3 and PR-9 in potato

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Late blight (LB), caused by *Phytophthora infestans* (Mont.) de Bary, is one of the most devastating plant diseases on potato, the fourth largest grown crop worldwide. The re-emergence of LB is considered to be related to shifts in the pathogen populations. To effectively control LB, it is important to have a better understanding of the responses that potato has developed with the current pathogen populations. In this study, two *P. infestans* genotypes, US-1 (old, mildly aggressive) and US-8 (new, highly aggressive), were used to inoculate two potato cultivars, Russet Burbank (RB, susceptible) and Kennebec (KB, moderately tolerant). Defense responses to infection were studied at two levels: (i) microscopic observations of potato leaf discs inoculated with *P. infestans*; and (ii) induction patterns of genes encoding three PR-proteins (*pr*-2, *pr-3* and *pr-9*) using northern blots analysis overtime and in three leaf strata of the whole plant. The accumulation of brown lignin-like materials and increases in the affinity of cell walls to trypan blue surrounding infection sites were observed in leaf discs collected from KB×US-1 24 hours after inoculation, and not in KB×US-8, RB×US-1, or RB×US-8. Northern blot analysis showed that the three *pr* genes were activated earlier in both cultivars in response to US-1 as compared to US-8. In addition, the induction of these genes occurred earlier in KB than in RB. Furthermore, *pr-2* and *pr-3* were found to be induced both locally and systemically while *pr-9* was induced only locally in KB inoculated with US-1. Our results demonstrated that the increase in LB severity on potato may be related to the observed changes in the activation of plant defense responses after infection.

B7. Fusarium II

B7.1 Application of DNA methods to identify and detect *Pythium, Phytophthora* and *Fusarium* species associated with soybean root rot in eastern Ontario and Québec.

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A survey of soybean root rot was conducted across 116 soybean commercial fields in eastern Ontario and Québec in spring season of 2001 and 2002 in order to determine the major root rotting fungi. Seedlings representing the various levels of root rot from each field were plated onto Petri dishes containing selective media for *Fusarium* or *Pythium/Phytophthora*. The remaining portions of the roots were frozen at -20 C for later molecular detection. A field located at the Central Experimental Farm, Ottawa, and naturally infested with *Phytophthora sojae* was selected as a positive control for the detection of this species. During the two years, we frequently isolated *Pythium sylvaticum* and rarely *Phytophthora sojae*. In 2002, we noted a higher level of root colonization by *Pythium* and *Phythophthora* than in 2001. We routinely detected *P. sojae* in Quebec (36% of fields) and in Ontario (28% of fields) and additional species of *Pythium* reported as pathogenic on soybean and cereals were found. Using DNA sequencing of the EF1-alpha gene, a set of 219 *Fusarium* isolates was selected and classified into eleven main groups. Some isolates of *Fusarium* from Ontario and Quebec were highly pathogenic on soybean seeds in plates and significantly reduced root and shoot growth in growth chamber experiments. For the first time, we developed a *Fusarium* DNA array assay to consistently identify and detect *Fusarium* species directly in soybean roots. A low incidence of false negative results was found with the DNA array technique when compared to root plating. The *Fusarium* species implicated in soybean sudden death syndrome disease was not found in eastern Ontario or Québec.

B7.2 Effect of seed treatments on control of dwarf bunt of winter wheat in Ontario.

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Dwarf bunt, caused by *Tilletia controversa Kühn*, is a destructive disease of winter wheat in Ontario. To determine the effect of seed treatments, trials were conducted with eight cultivars of winter wheat at two locations each year from 2002-2003 in central Ontario, under *T. controversa* inoculated field conditions. The seed treatments including three rates of Dividend XL RTA (difenoconazole + metalaxyl-M, 0.13, 0.26, and 0.39 g a.i. kg^{-1} seed), Vitaflo 280 (carbathiin + thiram, 0.83 g a.i. kg^{-1} seed), and bioagent ACM941, a strain of *Clonostachys rosea* (*C. rosea* ACM941, 2.5 x 10^6 cfu kg^{-1} seed). Untreated seed was used as controls. Dwarf bunt was observed in the untreated controls for all trials, at average of 12% in 2002 and 18% in 2003. All seed treatments significantly ($P < 0.05$) reduced disease incidence. The three rates of Dividend XL RTA were all highly effective, providing 98-99% control. Vitaflo 280 and ACM941 were partially effective, reducing disease incidence by 51 and 31%, respectively. All seed treatments increased emergence by 5-8%, which was significantly different from the untreated control. Although the data indicate a trend of higher yield (0.6-2.6%) and lower thousand seed weight (0.4-1.5%) with seed treatments over the untreated controls, the differences were not significant at $P = 0.05$. The wheat cultivars differed in susceptibility, but all were considered moderately susceptible to the disease. There were no significant cultivar × treatment interactions in both disease incidence and emergence, suggesting that Dividend XL RTA is an effective fungicide for controlling dwarf bunt for all winter wheat.

B7.3 Corn transformation for *Fusarium* resistance.

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Gibberella ear rot in corn, caused by *Fusarium graminearum* can result in severe reduction in crop yield and quality of Ontario corn. After infection, this fungus releases several mycotoxins, such as deoxynivalenol (DON). *RPL3* (Ribosomal Protein Large Subunit 3) gene product is the target for DON toxin. A single nucleotide mutation that leads to the substitution of cysteine for tryptophan at position 255 in wild-type yeast protein confers resistance to *RPL3* for DON toxin. The same mutation was engineered into rice *RPL3* gene (Harris and Gleddie, AAFC Ottawa) and was named as *RPL4*. Two vectors, PML14 and PML16 were constructed**,** with *RPL4* gene under the control of enhanced 35S promoter and a silk specific promoter, respectively. Both vectors were individually introduced into *A. tumefaciens* and were then used to transform corn inbred line CG65. Putative transgenic plants were obtained after selection on phosphinothricin and selfed to produce T_1 seeds. The presence and incorporation of the transgene into genomic DNA was confirmed by PCR and southern blotting. A field trial of the T_1 generation was planted to test the effects of *RPL4* on resistance to infection by *F. graminearum*. At silking, corn silk or kernals were artificially inoculated with *F. graminearum* spores and the severity of ear rot symptoms were evaluated using visual rating scales based on the percentage of infected kernels. Some individuals in the population had ears with low disease ratings. The current studies are testing for the presence of *RPL4* gene in the plants with low disease ratings.

B7.4 Diversity of *Gibberella zeae* isolates from Manitoba

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Gibberella zeae (anamorph *Fusarium graminearum*) causes fusarium head blight, one of the most important diseases of cereals in the Canadian prairies for the last two decades. In 2002, 60 single-spored isolates of *G. zeae* were collected from naturally infected spikes of winter wheat from Carman and Winnipeg in Manitoba. These isolates were compared using vegetative compatibility analysis and PCR-based sequence related amplified polymorphisms (SRAP). Sixteen vegetative compatibility groups (VCG) were found among the 50 isolates tested. Eleven SRAP primer pairs amplified 90 polymorphic DNA fragments from 60 isolates and identified 59 distinct haplotypes within the dataset. Among seven pairs of isolates, each pair from a distinct spike, three had isolates with different VCGs and six comprised different SRAP haplotypes. Principal component analysis and UPGMA separated the dataset into two main groups, each with isolates from both locations. The analysis of molecular variance also revealed that 99% of the variance was associated to differences between individual isolates. According to the Mantel test, there was no significant relationship between VCG and SRAP data. This study showed that though the high diversity among *G. zeae* isolates, Carman and Winnipeg collections have a similar genetic makeup and are likely part of a same population.

B7.5 Introgression of Fusarium head blight (FHB) resistance in Canadian wheat germplasm by *in vitro* and marker-assisted selections.

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Fusarium head blight (FHB), incited by *Fusarium graminearum*, causes important yield and quality losses in wheat in Canada and worldwide, The development of wheat cultivars with resistance to FHB is difficult to achieve due to the laborious screening methods and the complex inheritance of resistance, both of which are subject to environmental variability. QTLs for FHB severity and DON level provide the basis for marker-assisted selection of resistant lines. We have developed *in vitro* selected doubled haploid (DH) lines from different crosses between Canadian cultivars as AC Superb (CWRS), AC Crystal (CPS), and candidate sources of resistance from China, Brazil and CIMMYT. The response of the DH lines and their parental lines to FHB infection was also evaluated using point inoculation method in greenhouse and spray method in field conditions. Molecular analyses were conducted on the parental lines and the genetic material derived from, using simple sequence repeat (SSR) markers linked to FHB resistance QTLs identified from the literature.

We will present data on FHB assessment of parental and doubled haploid progeny, and the use of four polymorphic QTLs in Canadian germplasm. Molecular analyses have allowed us to identify resistant DH lines with the same haplotype as the resistant parent. The application of these QTLs in Canadian wheat germplasm will be discussed. Based on these results, we believe that *in vitro* selection combined with marker–assisted selection, SSR markers, could offer a faster and more efficient way of developing FHB resistant wheat cultivars.

C1. Biochemistry I

C1.1 Differential biological availability of iron chelated with N,N'-di(2-hydroxyethyl)-ethylenediamine-N,N'-diacetic acid (HBED) to two Strategy I green algae.

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N,N'-di(2-hydroxybenzoyl)-ethylenediamine-N,N'-diacetic acid [HBED] is a very strong Fe^{3+} chelator. Strategy I vascular plants, which use a reductive system for iron acquisition, similar to many green algae, are able to access iron from HBED (Chaney RL [1988] J Plant Nutr 11:1033). However, iron-limited cells of the Strategy I green alga *Chlamydomonas reinhardtii* Dangeard were unable to access iron present as Fe^{3+}-HBED. In contrast, Fe^{3+} chelated with HEDTA (a weaker chelator) was rapidly taken up by iron-limited *Chlamydomonas* cells. *Chlamydomonas* ferric chelate reductase activity with Fe^{3+}-HBED was approximately 15% of the rate observed with Fe^{3+}-HEDTA, suggesting that low reductase activity with Fe^{3+}-HBED might be one factor in the low rate of iron acquisition. By contrast, iron-limited cells of the Strategy I green alga *Chlorella kessleri* Fott et Nováková were able to rapidly assimilate Fe^{3+} chelated by HBED, although ferric chelate reductase activity with Fe^{3+}-HBED was approximately 38% the rate of activity with Fe^{3+}-HEDTA. Similar differential iron uptake rates for the two algal species were obtained using the strong Fe^{3+} chelator DFB-mesylate. These results suggest that there are differences among Strategy I green algae in their abilities to acquire Fe^{3+} from various chelates: not all Strategy I algae can access tightly complexed Fe^{3+}. *Chlamydomonas* appears to be the first documented Strategy I organism that is unable to acquire iron from Fe^{3+}-HBED. These results may have implications for algal iron acquisition from bacterial siderophores (which also have high stability constants), and suggest that iron acquisition from Fe^{3+}-HBED might serve as an assay for an organisms' ability to access tightly complexed iron.

C1.2 LexA: A novel function in *Synechocystis* sp. strain PCC 6803: Redox responsive gene expression

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Expression of the *crhR* RNA helicase gene in *Synechocystis* sp. strain PCC 6803 is regulated by the redox poise of the electron transport chain between carriers Qa of plastoquinone and Qo of cytochrome b_6f [1]. A change in electron flow through these carriers allows response to light-dark transitions via alteration of *crhR* transcript accumulation. The isolation and identification of *Synechocystis* LexA as the *crhR* regulatory protein provides initial clues to the mechanisms responsible for differential *crhR* transcript accumulation in response to the predominating redox/light environment.

Northern analysis of *lexA* and *crhR* transcript levels under a variety of environmental conditions including light, dark, glucose and cold stress suggests that LexA functions as a repressor of *crhR* expression. Confirmation of an interaction between the LexA protein and the *crhR* gene by EMSA analysis, together with Western analysis showing constitutive expression of the LexA protein suggest LexA binding to the *crhR* gene may be post-translationally modified potentially via a redox-dependent phosphorylation/dephosphorylation cascade. Enhanced *lexA* transcript levels in the presence of glucose suggest glucose/carbon may also play a role in *lexA* transcript accumulation. Interestingly, *lexA* insertion and deletion mutants created by insertional mutagenesis exhibit light sensitivity. Work in

progress is directed towards a greater understanding of the *lexA*/carbon relationship. Due to their common association with DNA repair and the *Escherichia coli* SOS response, further transcript analysis was performed looking specifically at *lexA* and *recA* levels under DNA damaging conditions. Conclusions drawn from transcript accumulation following ultraviolet irradiation and mitomycin-C treatment suggest that *lexA* and *recA* are not DNA damage inducible genes, contrary to the established dogma in *E. coli*.

Taken together, these results suggest a novel and unique role among prokaryotes for *Synechocystis* LexA protein in the regulation of redox-responsive but not DNA repair gene expression.

[1] Kujat,S.L., and Owttrim,G.W. **(**2000) *Plant Physiol*., **124**, 703-713.

C1.3 The Expression of Quinol Terminal Oxidases in the Cyanobacterium *Anabaena variabilis* and the Marine Centric Diatom *Thalassiosira pseudonana*

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Plant chloroplasts contain a protein that shares some sequence similarity with mitochondrial alternative oxidase. This oxidase (called the IMMUTANS terminal oxidase or plastid terminal oxidase) is associated with the photosynthetic electron transport chain and catalyzes the oxidation of plastoquinol with reduction of O_2 to H_2O. We uncovered PTOX genes in the sequenced genomes of several different cyanobacteria (Plant Mol. Biol. 53, 333-341, 2003) as well as on cyanobacterial scaffolds present in the recently released metagenome of the Sargasso Sea environmental samples (Gene 349, 15-24, 2005). The presence of PTOX in cyanobacteria supports an endosymbiotic origin for this protein in eukaryotes via the event that led to chloroplast genesis. This hypothesis is also supported by the presence of PTOX in *Mesostigma viride* (a representative of the earliest branch of green plant evolution), as well as its presence in green and red algae (eg. *Chlamydomonas reinhardtii* and *Cyanidioschyzon merolae,* respectively). PTOX has apparently also entered additional lineages via secondary endosymbiosis as evidenced by its presence in the haptophyte *Emiliania huxleyi* and the diatoms *Phaeodactylum tricornutum* and *Thalassiosira pseudonana*. To date, PTOX has not been found in any non-photosynthetic organism. Toward understanding the role of PTOX in aquatic photosynthetic organisms, we have investigated PTOX expression in the cyanobacterium *Anabaena variabilis* PCC 7120 and the marine centric diatom *T. pseudonana*. In both organisms, PTOX expression increases in response to an increase in light intensity and decreases in response to dark. In addition, we are comparing the changes in PTOX expression to changes in expression of other quinol oxidases in these organisms (a bacterial-like cyt bd oxidase in *A. variabilis* and a mitochondrial alternative oxidase in *T. pseudonana*). Our hypothesis is that all or a portion of these oxidases may be involved in short-term adjustment of metabolism to changes in photosynthetic electron flux.

C1.4 A quinone redox cycle active during powdery mildew infection of wheat

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At least two types of quinone reductases are present in plants: [1] the ?-crystallin-like quinone reductases (QR1, EC 1.6.5.5) that catalyze the univalent reduction of quinones to semiquinone radicals, and [2] the DT-diaphorase-like quinone reductases (QR2, EC 1.6.99.2) that catalyze the divalent reduction of quinones to hydroquinones. QR2s protect cells from oxidative stress by making the quinones available for conjugation, thereby releasing them from the superoxide-generating one electron redox cycling catalyzed by QR1s. Two genes, putatively encoding a QR1 and a QR2, respectively, were isolated from an expressed sequence tag collection derived from the epidermis of a diploid wheat *Triticum monococcum* L. 24 h after inoculation with the powdery mildew fungus *Blumeria graminis* (DC) EO Speer f. sp. *tritici* Em. Marchal. Northern analysis and tissue specific RT-PCR showed that *TmQR1* was repressed while *TmQR2* was induced in the epidermis during powdery mildew infection. Heterologous expression of *TmQR2* in *Escherichia coli*

confirmed that the gene encoded a functional, dicumarol-inhibitable QR2 that could use either NADH or NADPH as an electron donor. The localization of dicumarol-inhibitable QR2 activity around powdery mildew infection sites was accomplished using a histochemical technique based on tetrazolium dye reduction.

C1.5 Significance of $P700^+$-reduction by electron donors generated through alternative pathways in *Hypogymnia physodes* L., and *Cucumis sativus* L.

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The driving force for the dark reactions of carbon metabolism in plant leaves is the generation of the reductant, NADPH and energy rich phosphate molecule, ATP through linear electron transport involving photosystem II (PSII) and photosystem I (PS I). When crop plants experience severe stress conditions like high temperatures or high light regimes or drought, PSII is down-regulated as a strategy to protect the function of photosynthetic apparatus against over-reduction of the electron transport intermediates. We have shown previously that the above mechanism operates in cucumber plants if subjected to a combination of low light and chilling temperature stress. In such leaves, the rate of electron donation from stromal reductants for $P700^+$ dark-reduction, an alternate electron transport route, was accelerated. In the present study, we used epiphytic lichens, Hypogymnia physodes L. to study the kinetics of $P700^+$ re-reduction during severe water deficit stress as this species encounters repetitive cycles of dehydration and re-hydration. Our results suggest that $P700^+$ reduction in dehydrated thalli is diminished as the electron donation pathways are curtailed. A large fraction of $P700^+$ dissipates the absorbed energy as heat, a protection mechanism that is much similar to one observed in photoinhibited cucumber leaves at low temperature. In re-hydrated state, $P700^+$ re-reduction in the dark is predominantly mediated by stromal reductants as PSII is unable to reduce plastoquinones. The physiological relevance of the different routes of electron donation to $P700^+$ will be discussed in detail.

C1.6 Towards understanding the role of PR10 proteins.

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Pathogenesis-related (PR) proteins are expressed in response to pathogen infection as well as under abiotic stress and are grouped under 17 well-characterized families based on their biological activity. These include the relatively under-characterized PR 10 protein family composed of small, acidic proteins with sequence similarities to RNases and cytokinin-binding proteins. Very little is known about the actual biological function of the PR 10 proteins as cytokinin-binding activities have never been demonstrated and RNase activity has only been established in some cases. Characterization of salinity induced changes in pea root proteome in our laboratory revealed significant increases in the levels of various members of the PR 10 protein family prompting us to investigate whether PR 10 proteins have an important role in mediating plant stress responses. Experiments conducted with transgenic Brassica napus plants constitutively expressing the pea PR 10.1 gene revealed that they were able to germinate and subsequently grow better under saline conditions compared to the non-transgenic controls. Results from these experiments will be presented along with those from mechanistic studies aimed at understanding the precise role of PR 10 proteins in mediating plant stress responses.

C2. Forest biodiversity

C2.1 Floods, fire and ice: disturbance ecology of riparian cottonwoods.

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Cottonwoods, *Populus* tree species that occur in riparian or streamside zones, are well adapted to physically dynamic floodplain environments. They are inundation-tolerant and not only survive flood flows but are dependent on periodic over-bank floods for bank erosion and channel change, and for supplemental moisture. At higher latitudes and elevations, rivers freeze and ice break-up in

spring-time provides powerful geomorphic disturbance. Since floodplain zones are relatively moist, riparian woodlands were less influenced by fire but fires have become more common since river valleys are favored areas for human uses including recreation.

This presentation analyses the life history and ecophysiological characteristics of riparian cottonwoods that enables their adaptation to physical disturbance. We recognize the natural coordination between river hydrology and cottonwood phenology and consider the consequences of disrupting the seasonal hydrograph by river dams and flow regulation. The influence of ice-events is poorly understood and we present observations from many rivers that provide insight into ice-associated disturbance. We further present observations from fires and burned cottonwood groves for a number of rivers across western North America.

Following from these observations, we analyze the roles of physical disturbance on riparian cottonwoods and assess the different responses of *Populus angustifolia, P. balsamifera, P. deltoides, P. fremontii and P. trichocarpa,* the five cottonwoods that are adapted to different ecoregions throughout North America. We conclude that the differing mechanisms and balances of clonal versus seedling reproduction of these cottonwoods reflects differences in local disturbance regimes.

C2.2 Un-deserted islands: Conserving forest plant biodiversity using aggregated variable retention harvesting.

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Forest understory plants, including both bryophytes and vascular plants, are susceptible to the physical and microclimatic alterations associated with forest management practices. Individual species may depend on the presence of particular substrates and/or particular microclimatic conditions found within forests. Variable retention harvest, retaining aggregates of standing trees ('tree islands') within clear-cut harvest blocks, may constitute areas of preharvest forest conditions to preserve source populations on site. This provides a nearby source of plant diaspores to encourage recolonization, thus conserving forest plant biodiversity. In order to offer conservation potential, tree islands must first contain the species at risk to forestry-related disturbance. This study evaluates the potential of tree islands to conserve forest plant species across different spatial scales.

One hundred ten $1m^2$ plots were established in each of five 1.0 ha tree island blocks and adjacent forest prior to harvest. Bryophyte and vascular plant species compositions as well as environmental variables (substrate, canopy coverage and tree density) were assessed for each plot. The conservation potential of tree islands was evaluated by comparing species and environmental variables of tree islands to those of adjacent areas (to be harvested), at a range of spatial scales and proximities. On average, individual tree islands contained 86% of the vascular species found within their local harvest block, 68% of those at the watershed scale, and 48% of those within the entire study site. Bryophytes showed a different pattern, having greater richness within individual islands (n=50 vs n=38 for vasculars), but a lower proportion of the block and watershed floras represented within the island (72%, 56% respectively). Such differences between vascular and bryophyte species richness and representation across spatial scales will influence the strategies used to conserve these taxa on site.

C2.3 Spatial pattern of forest floor bryophytes in mixedwood stands of northern Alberta.

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Bryophytes (mosses, liverworts, and hornworts) constitute an important yet often overlooked component of understory plant diversity in northern mixedwood forests. In these forests, bryophyte diversity and abundance are largely controlled by the number, types, and properties of substrates available for colonization. Considering the forest floor contains a variety of microhabitats, it is conceivable that multispecies assemblages, and the individual bryophyte species that comprise them, are patterned at different scales in response to changing conditions. Spatial pattern of species can provide valuable insight into processes influencing forest floor diversity. This study employs Multiscale Ordination (MSO) to

investigate the multispecies pattern of forest floor bryophytes along transects of contiguous quadrats in stands of mixedwood boreal forest. MSO uses the quadrat variance technique Three-Term Local Quadrat Variance (3TLQV) to detect the average size of multispecies phases. Spatial autocorrelation of transect data was determined using multivariate Mantel correlograms to examine community structure. The investigation of multispecies pattern using MSO was complemented by a pattern analysis of individual species using 3TLQV, and also Two-Term New Local Variance (2NLV) for detecting smaller phases of pattern. Bryophyte abundance was enumerated along three transects each consisting of 301 15x15 cm quadrats, in stands of mixed-coniferous forest in the Clearhills Upland Ecoregion of northern Alberta. Species that occurred in >5% of all quadrats within a given transect were entered into MSO. Results demonstrate that MSO was effective at summarizing the spatial pattern of multispecies transect data. Multispecies pattern consistently occurs across a wide range of spatial scales, corresponding to block sizes of approximately b=40, 20, and 5 quadrats. Individual species also exhibit several scales of pattern, with 2NLV frequently detecting phases at <1 m. Multispecies data exhibit significant positive spatial autocorrelation at the smallest and largest of 10 distance classes along transects (Mantel r, alpha = 0.005 after correcting for multiple tests). Results provide a unique perspective for assessing the potential effects of forest harvesting on the local persistence of bryophyte diversity.

C2.4 Patterns in terrestrial bryophyte and lichen species in young and old sub-boreal spruce forest

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This study examined the effect of forest age on the diversity, biomass and abundance of terrestrial moss, lichen and liverwort species. The study compared old-growth and young second-growth sub-boreal spruce forest sites at the Aleza Lake Research Forest in central British Columbia. Terrestrial lichen and bryophyte diversity patterns, and the processes affecting them, have not been extensively studied in the Sub-Boreal Spruce biogeoclimatic zone in B.C. Forest harvesting is a major landscape altering process central B.C. and clearcut logging can affect terrestrial bryophyte and lichen communities through changes to substrate and microclimate and through habitat fragmentation. The effects that extensive forest management is having on bryophytes and lichens in sub-boreal spruce forests is not well documented.

In total, 116 species of bryophytes and lichens were identified in both stand ages. Major differences in species composition were found between forest ages with 49% of lichen and bryophyte species found in common between the forest ages, 30% found only in old-growth forest and 21% found only in young second-growth. Liverwort species were much more diverse in old-growth with half of the liverwort species found only there and 96% of the recorded liverwort cover occurring in old-growth forests. Lichen cover was greater in second-growth stands and different assemblages of lichen species were common in young second-growth compared with old-growth stands. Moss abundance and diversity was similar between forest ages, however, species compositions varied. Second-growth stands were largely dominated by a single moss species.

This study highlights the need for additional study of the impacts of logging practices and rotation times on lichen and bryophyte diversity. The length of time required to accomplish the transition from second-growth to old-growth non-vascular and lichen floristics is unknown for this region.

C2.5 Tracking bryophyte community reassembly in the Acadian forest 9 years after forest harvest

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Clearcutting and site preparation (ground scarification) for tree planting are common forestry operations in the Acadian forest of New Brunswick, but responses of understory vegetation, particularly bryophytes, are poorly understood. Impacts range from death by scraping, crushing or burial, to gradual decline or immediate death when simple canopy removal alters microclimate. The nature of clearcutting creates a mosaic within which any or all of these stresses may be experienced at

the fine scale. This study documents the responses of forest bryophytes to two forestry operations: cut (i.e. harvested) vs. cut-and-scarified (i.e. harvested with site preparation).

Responses to these two operations were measured in 169 permanent quadrats as changes in bryophyte species abundances 4 and 9 years post harvest relative to those before harvest. Most trends noted 4 years post harvest continue 9 years post harvest. Greatest community change, and highest species losses and abundance declines occurred in the cut-and-scarified areas. Mean richness in these areas remains lower than in cut areas. Species composition in cut areas shows signs of recovery to pre-harvest composition: the 9 year post-harvest community shows higher similarity to the pre-harvest community than to 4 year post-harvest community. However, communities in cut-and-scarified areas continue to diverge from the pre-harvest community. Initial increases in moss species richness in both cut and cut-and-scarified areas reflect invasion by colonizing species, however these have since declined. Liverwort species richness declined significantly immediately after harvest, and continues to decline.

This study underscores the vulnerability of liverwort populations, and reinforces the relative importance of substrate disturbance in structuring bryophyte communities. Limiting scarification and other ground disturbance caused by forest harvesting methods may help to maintain bryophyte diversity in the Acadian forest.

C2.6 Responses of understory plants to selection harvest in an Acadian forest

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Given evidence that total forest canopy removal, as with clear-cuts, results in changes in understory communities and potential loss of biodiversity, forest managers are considering a shift to selection harvest, in which a mosaic of individual trees is removed. We investigated patterns of forest floor plant community change (vascular plants and bryophytes) in relation to partial canopy removal associated with selection harvest, and compared it to that in adjacent intact forest. Using a BACI design, we set up two study sites designated Control and Impact in 67 ha of Acadian forest. Permanent sample units were established in each: four quadrats ($1m^2$) nested within each of 24 plots ($100m^2$). Species cover and environmental features were recorded for 2 yrs Before and 2 yrs After harvest. Using univariate and multivariate approaches, we evaluated the impact of selection harvest by: [1] quantifying changes in the understory community and in environmental characteristics, and [2] relating changes in the plant community to those in the environment, especially to those created by the selection harvest. Not surprisingly, bryophytes and vascular plants in the impact area underwent greater change in composition, richness, and evenness than those in the control area, however there was more annual turnover in the latter than expected. Degrees of change differed among functional groups, with inherently infrequent species (e.g. many liverworts) at greatest risk of extirpation. Although the most intense disturbances were equivalent to those found with other harvest methods, the finer scaled mosaic of disturbance intensities created by selection harvest may reduce loss of species related to substrate damage. These findings will contribute to evaluate relative impacts of different forest operations on vulnerable components of plant communities.

C3. Bioproducts and alternative crops

C3.1 Microarray analysis of flax bast fibre development

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Flax (*Linum usitatissimum* L.) is an agricultural crop which produces phloem (bast) fibres in the outer layers of the stem. Bast fibres develop through a complex process involving cell fate establishment and pattern formation, cell elongation, and secondary cell wall biosynthesis. To study these developmental processes specifically in relation to bast fibres, the outer (fibre-containing layers) were removed from the stem by a process called scutching. mRNA was extracted from the scutched tissue and was used to create a cDNA library representing expressed sequences from epidermal, cortical, and bast fibre tissues. 9600 clones were cultured into glycerol stocks, inserts were

amplified by colony PCR and gel electrophoresis was conducted to confirm positive clones. PCR-amplified inserts ranging in size from 500 bp to 3 kb were purified and spotted onto a microarray slide. Average size of insert was estimated to be 1 kb.

Microscopic analysis has shown that flax bast fibres contain small amounts of cytoplasm at mature stages, possibly indicating that fibres are alive and metabolically active at maturity. Tissue samples were isolated corresponding to the three main tissues represented by the library: epidermis, cortex, and bast fibres. RNA from tissue fractions was extracted, amplified, and used for microarray analysis to designate tissue-specificity of genes represented in the cDNA library.

To enrich the microarray analysis results, target genes with putative involvement in cell identity, cell elongation, and cell wall development were cloned from arabidopsis by RT-PCR. These target genes were used as probes to obtain corresponding flax homologs from the cDNA library. Once isolated, these target genes will be used as flax-specific marker genes for studying bast fibre developmental processes.

C3.2 The effects of altered carbohydrate allocation and metabolism on plant growth and cellulose yield

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Increasing harvestable plant yield is a priority in many plant breeding efforts worldwide. In fiber producing crops, like forest trees, the economic value of this yield is directly related to the amount and quality of the cellulose-based fibers. As the availability of carbohydrates derived from photosynthesis is believed not to be a major limiting factor in cellulose biosynthesis, it should be possible to manipulate cellulose production through the over-expression of one or more key enzymes involved in the production of UDP-glucose (UDPG), the precursor to cellulose. This paper will discuss the effects of over-expressing two genes, sucrose synthase (SuSy) and UDP-glucose pyrophosphorylase (UGPase) on growth and carbohydrate allocation of transgenic plants. Both enzymes play integral roles in the synthesis of UDPG: SuSy catalyzes the breakdown of sucrose to UDPG and fructose, while UGPase produces UDPG from UTP and glucose-1-phospate.

Tobacco (*Nicotiana tabacum* L. cv. Xanthi) plants expressing either gene under the regulation of a constitutive promoter (*2X35S*) or a xylem-localized (4CL) promoter were generated by agrobacterium-mediated transformation. The plants carrying single transgenes were reciprocally crossed to generate T_2 double transgene plants, harboring both the SuSy and UGPase genes. Expression profiles of the transgenic lines were consistent with the expression patterns of the promoters, with higher expression of *2X35S* in leaves, and higher expression of 4CL in the stem tissue. Many single transgene lines and all double transgene lines showed a significant increase in height relative to controls. Single and double transgenic lines also showed increases in soluble sugar contents. Fibre characteristics were consistent with increased growth rates, with slightly shorter and thicker cell walls, representative of reaction wood formation. While there was no difference in starch and/or cellulose content, the increased growth rates and altered soluble sugar concentrations provide evidence for the role of SuSy as a marker in sink strength, and lend credit to the function of UGPase in a similar role. Despite not altering cellulose content, the up-regulation of these two genes was effective in altering total plant biomass, and as a result, total cellulose yield from a single plant.

C3.3 Polyhydroxybutyrate production in transgenic plants

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There has been a recent surge of interest in producing plastics from materials that can be readily eliminated from our biosphere in an "environmentally friendly" fashion. Polyhydroxybutyrate (PHB) belongs to a class of biodegradable polymers, which has properties similar to conventional plastics. Flax (*Linum usitatissimum* L.), a major oilseed crop grown in Canada, is an obligate self-pollinator

and has no viable weedy relatives in the Prairies. Thus, flax represents an attractive plant system to overcome the cross- pollination problem encountered with other genetically engineered crops. Flax accumulates a relatively large quantity of seed oil (~ 40% fresh weight). One strategy for the synthesis of PHB in developing flax seed could involve the diversion of acetyl-CoA from oil synthesis to bioplastic production. Three genes, which encode the enzymes responsible for PHB synthesis, were isolated from *Ralstonia eutropha*. Two gene constructs were prepared, driven by seed-specific promoters, to drive the synthesis of PHB in the cytosolic and leucoplast subcellular compartments of the oil-forming cells of developing *Arabidopis* and flax seed. Currently, the constructs are being introduced into these plants using *Agrobacterium tumefaciens*-mediated transformation.

C3.4 Seed yield improvement in fenugreek (*Trigonella foenum graecum* L.) using mutation breeding.

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Fenugreek is an annual legume widely grown as spice. It can produce high quality, bloat free forage and is a source of diosgenin. The first North American forage cultivar "Tristar" has indeterminate growth habit and needs 120 days to produce mature seed. To generate mutants that would produce high quality seed we used Ethyl Methane Sulphonate (EMS) on Tristar fenugreek. Seed were presoaked in distilled water and treated with varying EMS concentrations between 10 and 300 mM for 2, 4, 6, 8, 12, 16 & 24 hours. Plants were allowed to grow for 85 days, treated with a desiccant, allowed to dry before seed yield and quality was determined. Plants with high seed yield and uniform golden yellow colour seed were selected for producing the next generation. Progenies of some large seeded high yielding M_2 plants have shown the ability to mature early. One M_3 line derived from a selected plant has grown well under greenhouse condition with determinate growth habit and high seed yield. Since fenugreek is a self pollinated crop this line has the potential to become a new cultivar. This work indicates usefulness of mutation breeding in fenugreek improvement.

C3.5 Jerusalem artichoke development as a multipurpose crop.

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Jerusalem artichoke (*Helianthus tuberosus*) is a perennial plant, native to the North American prairies, that has vast potential as a multipurpose crop for the future of Western Canadian agriculture. Its high yielding capability of more than 100 tonnes of biomass per hectare, coupled with its ability to serve as raw material for food, forage and industrial material markets, make Jerusalem artichoke a perfect candidate for a novel agricultural crop. We are endeavouring to develop Jerusalem artichoke as a unique and ideal bioreactor through the discovery of tissue-specific promoters, initially from tubers. We have instigated three distinct approaches to find novel promoters; total protein isolation and characterization via HPLC, 2-Dimensional gel electrophoresis and SDS-PAGE analysis, genomic DNA isolation and analysis by GenomeWalking (BD Biosciences) and RNA isolation for cDNA library construction and screening. The protein analysis visualized three major protein bands (~45, 34 and 27 kDa) common to all Jerusalem artichoke varieties tested but unique to the tuber samples. We have also isolated three genomic DNA targets and have some promising initial results from the GenomeWalking procedure. The cDNA library contains approximately 60,000 clones and has yielded results among the relatively limited number of clones screened to date. I will be discussing the ongoing research and goals of this project.

C3.6 Is improvement in non-bloat causing sainfoin possible?

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Sainfoin is a valuable forage legume because it lowers incidence of bloat in grazing

ruminants when grown in mixed stands with alfalfa compared to pure alfalfa and production of greenhouse gasses when consumed. It is however, a challenge to maintain sainfoin in mixed stands in western Canada. Present cultivars selected for pure stand performance, yield less than alfalfa and get eliminated from mixed stands rapidly. To determine yield and quality of new accessions, tests were conducted at Lethbridge Alberta. Tests were also initiated to determine their ability to survive in alfalfa mixed stands. The pure stand tests indicated that some accessions out yielded the western Canadian check (Nova) by over 20% and had similar nutritional quality as the check cultivar. The high yielding accessions also showed better ability to survive in mixed stands than the check cultivar. These results indicate that sainfoin cultivars with improved yield and ability to survive in alfalfa stands in western Canada can be developed. A 3-year old mixed stand test allowed us to select germplasm for development of new and improved sainfoin cultivars.

C4. Hormones in abiotic stress

C4.1 Interactions between abscisic acid and ethylene in salt-stressed tomato roots

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Alpha-dioxygenases (α-DOX) catalyze the oxygenation of fatty acids to yield a newly identified group of oxylipins that protect tissues from oxidative damage and cell death. In tomato *a-DOX* was identified as a salt-regulated gene and is represented by a small family of which only one member, *LEa-DOX1,* is salt responsive. In roots LE*a-DOX1* expression is abscisic acid (ABA)-responsive. However, in salt-stressed roots with low ABA content, LE*a-DOX1* expression was enhanced relative to that in salt-treated roots with normal ABA levels. An explanation for this is provided by the role of ABA in suppressing ethylene accumulation in osmotically-stressed roots. *LEa-DOX1* expression was markedly responsive to the ethylene generating agent ethephon and the ethylene precursor 1-aminocyclopropane-1-carboxylic acid (ACC). However, ethylene responsive *LEa-DOX1* expression was reduced when ACC was combined with either salt stress or exogenous ABA suggesting that ABA suppresses ethylene effects and hence influences *LEa-DOX1* expression. Current research to dissect the interaction between ABA and ethylene utilizes mutants with altered ethylene production or signaling in which to explore the regulation by ABA and ethylene of LE*a-DOX1* and genes that encode key enzymes for the synthesis of ABA and ethylene. Finally, given the well-documented effects of both ABA and ethylene on root growth, we are also examining root growth in these plants and how it is influenced by ABA and ethylene.

C4.2 Analysis of the *Arabidopsis* cell suspension phosphoproteome in response to low temperature and abscisic acid treatment.

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One of the major signaling events that occurs in response to abiotic and biotic stresses is protein phosphorylation. Plants recognize stress at the cellular level, which then triggers an initiation of a stress response. Signal transduction pathways that transmit information throughout the plant induce this response. Stress can regulate gene expression at different levels, including posttranscriptional regulatory mechanisms, enhanced translation, and stabilization of proteins. In this phosphoproteomic study, we used different techniques to study changes in the phosphorylation status of the Arabidopsis cell suspension proteome in response to different conditions. We used both low temperature (4°C and 12°C) and abscisic acid (ABA) treatment at different time periods (0, ½ min, 1 min, 2 min, and 5 min) as different conditions for this study. We hypothesized that these two treatments would stimulate the phosphorylation of numerous proteins, some of which would be common between the two treatments. We used radioactive orthophosphate for pulse-labeling of Arabidopsis cell suspension culture to identify proteins that are phosphorylated directly in response to low temperature and ABA treatment. Total proteins were extracted and separated by two-dimensional

polyacrylamide gel electrophoresis, and phosphorylated proteins were identified using phosphor image analysis. Furthermore, we used Pro-Q Diamond phosphoprotein gel stain to study the steady-state protein phosphorylation under the same treatments. To confirm the specificity of the Pro-Q Diamond stain, we treated protein extracts with calf-intestinal phosphatase and did not observe any clear binding of the stain with proteins, when compared to untreated extract. In conclusion, we were able to identify several proteins that changed in their phosphorylation status in response to low temperature and ABA treatment as a first step for identifying candidate genes for the goal of producing stress tolerant crop varieties.

C4.3 Brassinosteroid functions in several abiotic stress responses of *Arabidopsis thaliana* and *Brassica napus*

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Brassinosteroids (BRs) are a group of naturally occurring plant steroidal compounds that posses growth-promoting and stress-response modulating properties. Previously we have shown that *Brassica napus* and tomato seedlings grown in the presence of 24-epibrassinolide (EBR), a BR, are more resistant to heat stress than are seedlings grown in the absence of EBR. We now demonstrate that EBR treatment increases the drought and cold tolerance of *Arabidopsis thaliana* and *B. napus* seedlings, as well as increasing the basic thermotolerance of *A. thaliana* seedlings. To understand how EBR promotes thermotolerance, we analyzed the expression levels of heat shock proteins in wild type *A. thaliana,* and in *det2-1* and *dwf4* BR-deficient mutants. In wild type seedlings, the hsp90 and hsp101 transcripts and proteins were present at slightly higher levels in EBR-treated *vs* untreated seedlings, whereas in *det2-1* and *dwf4* seedlings, the levels were higher in untreated seedlings. These results suggest that although exogenous EBR enhances hsp accumulation, the lack of endogenous BRs does not suppress the heat stress response. Similar to the increase in hsp expression, EBR also enhanced the expression of a subset of known drought- and cold-responsive genes in both *A. thaliana* and *B. napus* seedlings. Overall, the results suggest that EBR treatment increases resistance in plants to a variety of environmental stresses. A study of global gene expression in *A. thaliana* in response to EBR treatment is underway to understand how BR confers stress tolerance.

C4.4 Morphological and hormonal studies of transgenic canola (*Brassica napus*) plants expressing the gene 1-aminocyclopropane-1 –carboxylic acid (ACC) deaminase

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We studied the morphological (plant height, leaf area, root and shoot dry weight, flowering time, pod size, number of seeds per pod and seed weight) and hormonal (ethylene (of both shoot and pod), ABA, IAA and GA) characteristics of transgenic canola plants expressing the bacterial gene 1-aminocyclopropane-1 –carboxylic acid (ACC) deaminase, compared to canola cultivar Wester. Transgenic plants had significantly higher shoot height, pod size, number of seeds per pod and seed weight. Transgenic plants also flowered early (on 28th day) compared to Wester plants (34th day). They also had significantly lower leaf and pod ethylene production throughout the experiment. Transgenic plants also produced significantly lower amount of ethylene under induced wounding and gravitropic conditions. Transgenic plants exhibited significantly higher amount of ABA in shoot tissues on fourth and seventh week compared to Wester type while no significant differences were observed in IAA and GA content.

C4.5 The effect of ethylene and temperature on suberin lamella deposition in the exodermis and endodermis of *Zea mays* L. roots.

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Previously, roots of *Zea mays* grown in stagnant conditions developed aerenchyma and more suberin lamellae in their exodermis compared to roots grown in an aerated hydroponic medium. It is well known that aerenchyma development is controlled by ethylene. Is this also the case for suberin lamella development, and if so, is its effect consistent over a range of temperatures? To answer these questions two experiments were conducted, one with a hydroponic temperature of 23 C and the other with temperatures ranging from 16 to 19 C. Three-day-old seedlings were transferred to nutrient solution aerated with a blend of O_2, CO_2 and N_2 (control), or with these same gases plus ethylene. After six days, the roots were segmented according to age (which was proportional to daily growth rates), sectioned and stained with Sudan red 7B for suberin lamella. Root sections were scored based on suberin lamella staining intensity in exo- and endodermal cells. Control roots from both temperature treatments were long, thin, straight and white, while roots exposed to ethylene were short, thick, coiled and slightly browned. In the trial in which roots were grown at 23 C, ethylene enhanced exodermal suberin deposition; it was heavily encrusted in these cells very early in root development and through its entire length. At lower temperatures, exodermal suberization was slightly increased in the control roots. However, ethylene did not promote additional suberization of the exodermis in this case. In contrast to the exodermis, endodermal suberin lamella development was similar between the control and ethylene treatments in both temperature trials. In conclusion, ethylene can induce early and intense suberin lamella deposition in exodermal cells at 23 C, but this effect is suppressed at lower temperatures. Lower temperatures alone seem to induce some exodermal suberization but not nearly to the same extent as ethylene at the higher temperature. Since aerenchyma was formed in ethylene-treated roots at both temperature regimes and not in the control roots, it is clear that aerenchyma development and exodermal suberization (which is known to prevent radial oxygen loss from the cortex) are not controlled in the same way.

C4.6 Rhythm in the expression of a *PIP2* gene in roots of *Pisum sativum* and its response to $HgCl_2$ and ABA treatment imply AQPs are critical for radial water entry.

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Aquaporins (AQPs) are a highly conserved family of water channel proteins that play a significant role in the permeability of the plasma membrane. An AQP gene has been identified in pea (*Pisum sativum*) and its level of expression was studied in response to various factors to gain insight into radial water movement through roots. Diurnal variation of a *PsPIP2* AQP gene, its response to treatments with $HgCl_2$ (a known inhibitor of AQPs) and abscisic acid (ABA; a hormone critical in water movement) were investigated using real-time PCR (qPCR). It has been shown that *PsPIP2* mRNA is upregulated in fine roots at 9:00h and subsequently in tap roots at 11:00h. Following this, levels of the gene in both fine and tap roots decline to the end of the photoperiod to a background level maintained throughout the night. *PsPIP2* expression was consistent with later increases in root conductivity (Lp) that reached a maximum at 16:00h. The data suggests that the *PIP2* AQP is made later in the morning in order to accommodate the large flux of water movement seen in the afternoon. Exposure of roots to increasing concentrations of $HgCl_2$ for one hour caused an increase in *PsPIP2* while three hour exposure downregulated expression of the gene. *PsPIP2* may increase following one-hour $HgCl_2$ treatment to compensate for those AQPs blocked by Hg^{2+}. ABA exposure caused a decline in *PsPIP2* transcript levels. We hypothesize that closure of stomata induced by ABA treatment (as seen by a decline in transpiration) decreases the tension on water in the xylem and mitigates the need for a large number of water channels. This research emphasizes the importance of radial water entry in meeting the water requirements of the whole plant.

C5. Diseases of horticultural species

C5.1 Effect of 1-Methylcyclopropene treatment on postharvest decay in apple cvs. Empire and McIntosh.

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In recent years, 1-methylcyclopropene (1-MCP) has shown tremendous potential in maintaining fruit quality in apples during storage. The objective of this study was to determine the effect of 1-MCP on decay in stored apples. 'Empire' and 'McIntosh' apple fruits that had been wounded immediately after optimum harvest for long-term storage (as determined by internal ethylene content and starch staining) were treated with/without 1 $\mu L \cdot L^{-1}$ 1-MCP for 24 h at $0^{o}C$. Fruit were then stored for up to 120 days either in air at 0-1°C or in standard controlled atmosphere (SCA; 'McIntosh' at 3°C, 2.5% O_2 + 2.5% CO_2 for the first 30 days, then 4.5% CO_2 thereafter; 'Empire' at $2^{o}C$, 2.5% O_2 + 2.0 % CO_2). Decay incidence was recorded after 30, 60, 90, and 120 days after 1-MCP treatment in apples that were kept in air. No decay was observed at 30 and 60 days after treatment in both cultivars held in air. A higher incidence of decay was observed in wounded plus 1-MCP treated 'McIntosh' apples than in wounded only apples after 90 and 120 days in air and in SCA storage. 1-MCP had a variable effect on decay in 'Empire' apples, where higher decay incidence was observed in wounded plus 1-MCP treated apples than in wounded only apples at 90 days, but the reverse trend was found at 120 days after treatment. In summary, 1-MCP had a variable effect on decay incidence in different apple cultivars, and this variability in response is an important consideration in any program utilizing 1-MCP treatment.

C5.2 Internal Fruit Rot of Greenhouse Sweet Peppers in Alberta

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Internal fruit rot of sweet pepper (*Capsicum annuum* L.) is a new disease in Alberta greenhouses. The white mycelium of the fungus covers the internal surfaces of infected fruits, and some fruits, but not all, also develop external lesions. Because affected fruits can not be culled based solely on visual symptoms, many suspicious fruits are destroyed, thus reducing marketable yield. Research was undertaken to study the etiology and epidemiology of this new disease and to develop a prevention program. Eight greenhouses in central and southern Alberta were surveyed in 2004, fungi from symptomatic fruits were isolated, identified, and their pathogenicity confirmed following Koch's Postulates. More than 50 *Fusarium* isolates were collected, mainly belonging to two genera, *F. solani* and *F. proliferatum.* A few isolates of *F. verticillioides* and *F. oxysporum* were obtained from one greenhouse. *Fusarium solani* was the major cause of fruit and stem rot on red and yellow peppers, while *F. proliferatum* was mainly associated with orange peppers (cv. Sympathy) showing internal fruit rot symptoms. Only *F. proliferatum* caused internal fruit infection on red pepper (cv. Early California Wonder) when flowers were artificially inoculated. *Fusarium verticillioides* was also pathogenic when artificially inoculated to pepper flowers. *Fusarium* infection was associated with high temperature and humidity levels in greenhouses. Molecular detection using PCR with species-specific primers on all *F. proliferatum* and *F. verticillioides* isolates is in progress.

C5.3 The Effect of Different Rates of Nitrogen and Calcium Fertilizers on Incidence and Severity of Septoria Late Blight and Celery Yield

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Septoria late blight (*Septoria apiicola*) is a common and economically important disease affecting celery (*Apium graveolens*) leaves and petioles. Integrated management of the

disease could be improved by further understanding the relationship between calcium (Ca) and nitrogen (N) fertilizer and disease. Field trials were conducted in 2003 and 2004 to determine effects of calcium chloride (Ca 36%) and ammonium nitrate (N 34%) on disease and yield of two celery cultivars. Trials were inoculated with diseased tissue spread between the rows, and nutrients were applied as a percent of the recommended rate. Disease and yield were assessed at harvest. In 2004, disease progress was monitored weekly by assessing % leaf area diseased, and plant tissue was analyzed for N and Ca content at midseason and harvest. In 2003 disease incidence was significantly lower in the 150%N + 100%Ca treatment except those that received 100 or 200 % N plus Ca. In 2004, harvest weight and trimmed weight of both celery cultivars decreased with increasing area under the disease progress curve (AUDPC). There were significant differences in tissue N and Ca between cultivars at midseason and harvest. Increasing rates of N resulted in higher tissue concentration of N midseason, but not at harvest, and higher concentration of N at midseason was related to higher yields. For cv. 'Florida 683', higher tissue concentrations of Ca midseason were related to lower leaf blight severity ($r = -0.44$, $P < 0.018$) and higher weight and height, possibly because of lower disease levels.

C5.4 Important Pathological Problems of *Agave tequilana* in Mexico

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There are at present, three principal, important pathological problems of the *Agave tequilana* Weber var. *Azul.* The plant is important, because from its sap is produced an alcoholic drink known as Tequila and also a sweet extract which is exported to Canada and added to maple syrup. The plant is grown on an extension of approximately 62,000 ha with the present population of 250,000.000 plants in the states of Jalisco, Guanajuato, Michoacán, Nayarit and Tamaulipas. *Agave tequilana* suffers from attacks of *Thielaviopsis paradoxa, Cercospora* type fungus and a Coryneform bacterium besides few other bacteria. The first fungus is widely distributed and affects around 23% of the plants usually from four years on, producing yellow irregular areas on its leaves. The leaves roll inwards. The fungus affects the roots, turning them black, but in young plants the fungus inhibits rooting, because of gas production. Mother plants and suckers die in few months. Some leguminous plants are affected the same way. The Cercospora type fungus affects leaves and stems (up to 55% of the same age of plants as the previous ones), produce grayish, oval lesions, usually on the lower leaf parts, kills the tissue, turning it red and producing usually complete chlorosis and death of the plant. The Coryneform bacterium produces soft rot of leaf tissue and has killed up to 53% of young plants in coastal Jalisco region. *T. paradoxa* and the bacterium can be introduced into the plant by the weevil *Scyphophorus acupunctatus* Gyllenhal (Coleoptera: Curculionidae).

C5.5 Blueberry anthracnose in BC – weather conditions required for infection, overwintering strategy, and potential biological control of disease.

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Blueberry anthracnose (ripe-rot) is a yield-reducing disease that severely affects post-harvest fruit quality of highbush blueberries (*Vaccinium corymbosum* L.) in British Columbia and other growing regions. The epidemiology of this disease was investigated by determining the causal organism, the weather conditions required for successful infection, the host tissues that served as inoculum overwintering sites, as well as evaluating two potential biological control agents. Isolates were collected from BC blueberry fields over a 3-year period (2002-2004) and identified as *Colletotrichum acutatum* using fungal plate colony growth morphology, growth rate comparisons, and PCR analyses with species-specific primers. Artificially inoculated fruit were infected over a wide range of temperatures (7° to 30°C) in growth chamber trials. Field experiments on trap plants exposed to natural levels of inoculum, repeated over 3 years (2001-2003), as well as on potted blueberry plants artificially inoculated in 2004, showed that plants were

susceptible to infection at all stages of development (bloom to ripe berry) and that inoculum presence and weather conditions determined disease incidence. Infection occurred when specific infection periods (10 hours of leaf wetness with simultaneous temperatures above 11°C) occurred during the season, suggesting that prediction of disease may be possible based on weather conditions. An investigation of overwintering inoculum in host tissues revealed that survival occurred mostly within flower buds, followed by twigs and fruit trusses. *C. acutatum* was not recovered from field-collected mummified berries, or artificially-inoculated berries left in the field during 2003 and 2004. Infection of developing flower buds in May-June of the preceding growing season resulted in the greatest inoculum recovery the following year, suggesting a target for appropriate timing of disease control methods. Two biological control agents, *Gliocladium catenulatum* (Prestop, Verdera Oy, Finland) and *Trichoderma harzianum* (PlantShield, BioWorks Inc., USA) reduced disease by 30-45% on potted plants (2002) and in field trials (2003 and 2004) when applied three times during the growing season.

C5.6 Identification of *Botrytis mali* Ruehle existing within the *Botrytis* population causing fruit decay on apple in the orchards of British Columbia.

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DNA sequence analysis of the β-tubulin gene, for 36 *Botrytis* isolates consisting of nine species, was used to aid morphological identification of species involved in fruit decay of apple. The DNA analysis placed the various isolates into groupings that generally reflect the morphological based model for *Botrytis* classification. Isolates of *B. porri* and *B. aclada* grouped together in a minor clade, as did isolates of *B. tulipae* and an unidentified *Botrytis* species, while *B. streptothrix*, and *B. squamosa* both formed individual subgroups. *Botrytis cinerea* isolated from apple and other host plants grouped together in a single clade along with *B. fabae*. However, isolates of a second putative *Botrytis* species, *B. mali*, also isolated from apple and two *B. mali* herbarium Type-specimens formed a distinct clade. Morphological characteristics including growth rates and sclerotial development were also recorded for *B. mali* isolates, and the results closely resembled those described for this species by Ruehle (1931). The morphological data along with the DNA sequence analysis reported in this study support the initial work by Ruehle, describing the apple pathogen *B. mali* as a unique species, separate from *Botrytis cinerea*. This is the second report of *B. mali* causing decay of apple since first being described in 1931.

C6. Molecular aspects of pathogenesis II

C6.1 An importin-a homolog, MOS6, is required for *snc1*-dependent constitutive disease resistance signaling in Arabidopsis

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A constant evolutionary arms race is underway between pathogens and their hosts. Plant disease resistance is the consequence of an innate defense mechanism mediated by *Resistance* (*R*) genes. The Arabidopsis *snc1* mutant contains a mutation in a TIR-NBS-LRR-type *R*-gene that leads to constitutive activation of resistance reponses. The structure of this R-protein is reminiscent of immune response perception modules in animal cells, such as Toll-like receptors (TLRs) and Nucleotide-binding oligomerisation domain (NOD) proteins. We eployed a *modifier of snc1* (*mos*) suppressor screen to identify components of the downstream signaling network activated by *snc1* – few of these are presently known.

Three alleles of the mutant *mos6* partially suppressed constitutive *pathogenesis-related* (*PR*) gene expression, salicylic acid (SA) accumulation and resistance to virulent pathogens in the *snc1* and *snc1 npr1* genetic backgrounds. The single mutant *mos6-1* exhibited enhanced disease susceptibility (EDS) to a virulent oomycete pathogen. *MOS6*, identified by positional cloning, encodes

importin-a3, one of eight a importins identified in Arabidopsis. a-Importins, a conserved family of mobile targeting receptors, mediate the import of nuclear localization signal (NLS)-containing proteins across the nuclear envelope. There is evidence from animal and other plant systems that different importin-a homologs are essential for the nuclear import of specific proteins. We previously reported that MOS3, a protein homologous to human nucleoporin 96, is required for constitutive resistance in *snc1*. The identification of *MOS6*, highlights an essential role for nucleo-cytoplasmic trafficking, especially protein import, in plant innate immunity.

C6.4 Ubiquitination is required for plant disease resistance signaling

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Plants are fending off attacking pathogens using a complex network of sophisticated defense mechanisms. In gene-for-gene resistance, pathogen elicitors are recognized by cognate *Resistance (R)*-genes that initiate various physiological reactions, culminating in the containment of the pathogen and systemic resistance against subsequent infections. Although the physiological responses are well studied, the underlying genetic mechanisms are only recently being elucidated.

To understand *R*-gene downstream signalling, we are taking advantage of a unique gain-of-function mutation in a TIR-NB-LRR *R*-gene. A single amino acid substitution renders the *RPP5* homolog *suppressor of npr1-1 constitutive 1 (snc1*) constitutively active, resulting in increased resistance to bacterial and oomycete pathogens, constitutive expression of *Pathogenesis Related* (*PR*)-genes, elevated endogenous salicylic acid levels and stunted growth of the mutant. In order to dissect the signaling pathways induced by *snc1,* we conducted a screen for suppressors of the constitutive resistance in *snc1* and identified several *modifier of snc1 (mos*) mutants.

Here, I am presenting *mos5*, a mutant that suppresses the morphological phenotype of *snc1* partially, abolishes constitutive *PR*-gene expression and resistance and shows reduced endogenous levels of salicylic acid. The mutation in *mos5* was cloned using a map-based approach and found to be a 15 bp deletion in *AtUBA1,* one of two Ubiquitin activating enzymes (E1) in *Arabidopsis thaliana*. The *mos5* single mutant displays slightly enhanced disease susceptibility, indicating a role of the ubiquitin/26S proteasome machinery in both gene-for-gene and basal resistance. Furthermore, our data indicate that the perturbation in the ubiquitin system caused by the *mos5* mutation does not affect all tested *R*-genes, implying an even wider array of possible signaling mechanisms. The mutation in *mos5* lies outside the highly conserved regions important for E1 function and might affect substrate specificity. A hypothesis for the role of *mos5* in *R*-gene signaling will be presented.

C6.3 Reduced Expression of Eukaryotic Translation Initiation Factor 5A2 Imparts Strong Pathogen Resistance to *Arabidopsis thaliana*

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Previous studies in our laboratory have demonstrated that eukaryotic translation initiation Factor 5A (eIF5A) regulates programmed cell death (PCD). Specifically, eIF5A appears to act as a shuttle protein, translocating specific mRNAs encoding death proteins from the nucleus to the cytosol for subsequent translation. In Arabidopsis, there are two death isoforms of eIF5A. One of these, eIF5A1, regulates PCD accompanying senescence. In this study, we show that the second isoform, eIF5A2, regulates PCD accompanying infection by necrotrophic bacterial and fungal pathogens.

Within 72 h of infection with virulent Pseudomonas syringae pv Tomato DC 3000, eIF5A2 protein is strongly up-regulated in the leaves of Arabidopsis wild-type plants. The up-regulation coincides with visible symptoms of disease progression. Moreover, eIF5A2 is the only eIF5A isoform to show increased expression following infection. In addition, the up-regulation appears to be post-transcriptionally regulated. To further test the role of eIF5A2 in disease development, transgenic Arabidopsis plants with constitutively suppressed eIF5A2 expression

were developed using antisense T-DNA insertions. The suppressed plants showed marked resistance to virulent *Pseudomonas syringae* pv Tomato DC 3000, with some lines exhibiting a 99 % decrease in bacterial load, relative to wild-type plants. This suppression of pathogen growth correlated with the absence of disease symptoms. The same lines also showed strong resistance to infection by the necrotrophic fungal pathogen, *Sclerotinia sclerotiorum*. The results have been interpreted as indicating that eIF5A2 regulates PCD induced by necrotrophic pathogens, and that suppression of eIF5A2 inhibits host cell death upon infection and the ensuing release of nutrients that normally support growth of the pathogen.

C6.4 Genetic transformation of interspecific *Exacum* hybrids with a thaumatin-like protein (tlp) construct for enhanced fungal resistance

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Interspecific hybrids of *Exacum* L. (Gentianaceae), native to Sri Lanka, have great potential as a new ornamental crop. However, these hybrids are very susceptible to root fungal pathogens including the vascular wilt, *Fusarium* sp. To address this problem, we aim to generate plants with increased resistance to fungal pathogens by introducing an antifungal gene (i.e., tlp; thaumatin-like protein) through *Agrobacterium*-mediated gene transfer. The tlp gene is reported to confer broad-based resistance to several fungal pathogens when incorporated into non-native genomes.

Preliminary studies concentrated on refining multiplication and organogenesis protocols for this germplasm. Use of published protocols for *Exacum* micropropagation resulted in the development of chlorotic and necrotic tissue unacceptable for this research. Healthy micro-shoot production required modification of the published micropropagation media by the addition of charcoal and higher concentrations of plant growth regulators (PGR). Shoot organogenesis from leaf explants required genotype specific medium. However, the most effective PGR combination (across genotypes) was MS salts plus 0.1 mg/l NAA + 2 mg/l BA.

Our tlp construct was modified to include a selectable marker (hygromycin resistance), and a new promoter site (2 tandem CMV35S) driving the tlp gene. Following construct modification, we evaluated both the floral-dip and co-cultivation *Agrobacterium* mediated transfer methods. Due to difficulties on establishing the optimal stage of flower development, we decided to use the co-cultivation transformation system.

Based on the literature for Gentianaceae transformation and preliminary experiments, two *Agrobacterium* strains have been evaluated (i.e., C58 and EHA-105). Subsequent experiments evaluated various concentrations of bacterial solutions (i.e., OD_{600} 0.1-0.9). Results suggest higher concentrations are more effective for this system. Additional variables evaluated included pre-cultivation treatments (i.e., leaf explants placed on shoot organogenesis medium 2-3 days before *Agrobacterium* infection), two different co-cultivation media (i.e., 2 mg/l BA+ 0.1 mg/l NAA, 2 mg/l 2-4D), and explant type (i.e., calli, internodal sections, leaf discs, hypocotyls). Currently, co-cultivated explants are growing on selection media and will be evaluated for the presence of the transgene when sufficient tissue is available.

C7. Fusarium III

C7.1 Characteristics of *Fusarium graminearum*-positive wheat fields in Alberta, 2002 and 2003.

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Over the past several years the Canadian Grain Commission (CGC) has observed an increasing frequency of *Fusarium graminearum* (Schwabe) (*Fg*) associated with fusarium damaged kernels collected from Alberta wheat samples submitted as part of their yearly harvest survey. Characteristics of these *Fg*-positive (+ve) fields were collected anonymously from Alberta producers to develop a better understanding of factors contributing to the appearance and development of *Fg* in Alberta. In 2002 and 2003, 40 and 45 Alberta producers with *Fg*+ve grain samples were contacted, respectively, and fields were classified according to wheat class, irrigation, tillage regime, previous crop,

and frequency of host crops in the previous 4 years. In 2002, 17 of the producers contacted responded to the survey questions that were sent out, while in 2003, 19 of the producers responded. Over 90% of the *Fg*+ve fields were planted to wheat cultivars with either a poor or very poor rating for fusarium head blight resistance. In 2002 and 2003, 41% and 70%, respectively of the *Fg*+ve fields were amber durum. Considering only the irrigated region of Alberta, irrigation was reported for 8 of 11 and 16 of 17 fields in 2002 and 2003, respectively. Over 80% of the fields with *Fg*+ve samples were under either conventional or minimum tillage. A slightly higher percentage of fields had a non-host versus host (56.2 vs. 43.8%) as the previous crop in 2002, while in 2003 a similar trend was observed. In 2002 and 2003, most of the *Fg*+ve fields (>80%) had 2 or 3 host crops planted during the previous 4 years. In general, susceptible wheat classes/cultivars, irrigation and rotations with less than 2 years between host crops were common practices for producers with *Fg*+ve grain samples.

C7.2 *In vitro* evaluation of *Fusarium spp.* and fusarium head blight resistance in barley at two temperatures.

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Agar amended with ground grain of barley (*Hordeum vulgare* L.) genotypes 'AC Lacombe', 'Stander', 'Chevron' H94051001, I92130, Penco/Chevron, and CI4196 was used *in vitro* to evaluate genotype resistance or tolerance to fusarium head blight (FHB) as indicated by mycelial growth of *Fusarium graminearum* Schwabe (PW027) using two temperature regimes (17 and 23±1ºC). Twenty grams of surface-sterilized grain were ground in 100 ml of sterilized distilled water, and then mixed with plain agar at a ratio of 1 agar: 9 grain, and solidified in Petri dishes. *Fusarium graminearum* showed larger colony diameter (mm) at 17ºC and room temperature (23±1ºC) on the mixture of agar and ground grain for susceptible genotypes, 'AC Lacombe' and 'Stander' compared with most resistant/tolerant genotypes except for resistant CI4196. Colony growth at 17^{o}C was slower compared with room temperature, but the magnitude of the difference in colony diameter was similar between susceptible and resistant genotypes for the two temperature regimes. Detached leaves of the same barley genotypes except CI4196 were inoculated with single isolates of *F. graminearum* and *F. culmorum* (PWOT6) for evaluation of partial disease resistance (PDR) components at 10^{o}C and room temperature. Both *Fusarium* isolates were pathogenic at room temperature with shorter incubation and latent periods and greater lesion sizes on detached leaves compared to 10^{o}C. PW027 caused larger lesions on susceptible cv 'AC Lacombe' and 'Stander' compared with resistant 'Chevron', H94051001, I92130 and Penco/Chevron at room temperature. PW027 was more virulent than PWOT6 at both temperatures causing shorter incubation and latent periods and larger lesions for all barley genotypes. PW027 produced more macroconidia at room temperature in susceptible genotypes compared with resistant ones. The PDR components were poorly correlated between the two isolates evaluated under the two temperature regimes. Results suggest that both *in vitro* assays may be alternate selection methodologies for FHB resistance. The detached leaf assay has the advantage of measuring specific disease components, allowing elucidation of the potential nature and genetic components of resistance to FHB. *Fusarium graminearum* appeared to better differentiate between resistant and susceptible barley genotypes at room temperature compared with *F. culmorum.*

C7.3 Influence of soil type on incidence and severity of fusarium wilt (*Fusarium oxysporum* f. sp. *conglutinans*) of canola (*Brassica napus*) in Alberta.

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Fusarium wilt is an important disease in canola that can cause up to 30% yield loss in canola. Fusarium wilt has been documented in some, but not all, regions of Alberta. Therefore, the objective of this research was to determine the incidence, severity and distribution of this disease in canola-growing regions across Alberta. As *Fusarium oxysporum* f. sp.

conglutinans is a soil-borne fungus, edaphic factors, as encapsulated in soil classifications, may be important predictors of wilt severity or incidence. Surveys were conducted in 2003 and 2004, encompassing 250 sites across Alberta. One hundred plants were randomly collected from each field along a "W" shaped pattern. Each plant was evaluated for symptom severity on scale ranging from 1 to 9, where 1 represented no disease and 9 represented complete necrosis. Incidence was calculated as the percentage of symptomatic plants in each field. Fields with symptomatic plants were found throughout Alberta, but the disease was most severe and prevalent in the east-central area. Overall severity and incidence were higher in 2003, a drier and warmer year, than in 2004. This research identified an association between soil type and disease incidence. Disease incidence was highest on plants found in Chernozemic, followed by Solonetzic, then Luvisolic soils. Incidence in fields with black and dark brown Chernozemic soils was greater than in those with dark grey Chernozemic soils.We hypothesize that a negative interaction between available soil water and disease incidence overrides other soil characteristics, because high disease incidence was associated with high rainfall areas, and because the disease was more severe in the drier, warmer year of the study.

C7.4 Relative susceptibility of cereal grains to fusarium head blight

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Fusarium head blight (FHB) is the most damaging and pervasive disease of barley and wheat in the eastern Canadian prairies. However, FHB also affects other cereals, such as oat, and has been reported on rye and triticale. Casual observations indicate that crops vary in their general susceptibility to FHB, but this has not well-documented. In 2004, representative cultivars of common and durum wheat, barley, and oat, and single examples of rye, triticale and canary seed were planted in southern Manitoba to compare reactions to FHB, based on *Fusarium* spp., fusarium damaged kernels (FDK), and deoxynivalenol (DON) levels in seed. The cultivars chosen were largely recently-registered and not been tested comprehensively for their FHB responses. Trials were established at 3 locations, with 4 replications. Background *Fusarium* was supplemented with *F. graminearum*-infested 'corn spawn' inoculum spread on the soil surface prior to heading. Severity of FHB varied among the 3 locations, e.g. DON levels for the 12 wheat cultivars tested were 2.3, 3.6 and 7.2 ppm., and varied among crops across all sites, e.g. DON levels of 2.3 ppm for canary seed, 12.1 ppm for triticale. Similar variability was found for *Fusarium* spp. and FDK. Among crops, when averaged over all locations or based on East Selkirk ( ), where disease was highest, DON levels were 4.4 (7.2) for wheat, 2.7 (5.4) barley, 2.9 (4.0) oat, 2.4 (2.9) rye, 12.1 (18.5) triticale, and 2.3 (3.0) canary seed. The results suggest that triticale is particularly susceptible to FHB (similar to durum), followed by wheat, and that barley, oats, rye and canary seed develop somewhat lower, but similar levels of FHB. Among the major cereal crops, FHB severity (as DON) among the cultivars tested was most diverse in wheat (1.1 - 13.0 ppm), less so in barley (1.8 - 4.6 ppm) and least in oat (2.2 - 3.1 ppm). The potential variability in FHB reactions among additional cultivars of rye, triticale and canary seed should be examined further.

C7.5 Role of *Fusarium* species in rusty root development on ginseng roots in British Columbia

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Rusty root of ginseng is characterized by reddish-brown spots and discoloured areas on roots, usually near the crown. These areas become dark brown, raised and corky over time and slough off, exposing the underlying tissues. The cause(s) of rusty root are presently unknown. Rusty root tissues collected from several farms during 2003 and 2004 were plated onto different culture media to recover microbes. *Fusarium* was the most frequently isolated genus, with *F. solani*, *F. equiseti*, *F. culmorum*, *F. avenaceum* and *F. sporotrichioides* recovered in order of decreasing frequency. All species except *F. solani* were also recovered from barley and wheat straw mulch from ginseng gardens. *In vitro* pathogenicity tests and greenhouse inoculations of ginseng roots revealed that *F. equiseti* induced reddish-brown lesions resembling rusty root symptoms. Other species pathogenic to a lesser extent were *F. culmorum*, *F. avenaceum*, and *F.*

sporotrichioides, while *F. solani* and *F. graminearum* were non-pathogenic. Populations of *F. equiseti* in soil in 12 ginseng fields were highly correlated with rusty root incidence. Histopathological studies revealed that rusty root tissues contained mycelium and the upper 8-10 cell layers of the root that were invaded became necrotic. Levels of phenolic compounds, peroxidases, polyphenoloxidase and phenylalanine ammonia lyase were significantly higher in the affected tissues. Rusty root of ginseng is the outcome of root defense responses following infection by *Fusarium* species, notably *F. equiseti*, in attempts to restrict pathogen colonization. This is the first report of the completion of Koch's postulates for ginseng rusty root.

D1. Shoot apical meristems (part 1)

D1.1 The shoot apical meristem: an historical perspective

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Although much of the current investigation of shoot apical meristems is in the realm of molecular genetic analysis, it is important that previous structural and functional studies not be overlooked. Since Caspar Friedrich Wolff described the shoot apical meristem in 1759, many and varied interpretations have arisen. In the early 19th century the apical cell was recognized in vascular cryptogams and this interpretation was extended to seed plants. However, by the 1860s this view was replaced in seed plants by the histogen concept which recognized specific meristem layers giving rise to specific tissues. In 1924 the tunica-corpus interpretation of angiosperm shoot apices became widespread, the two regions being distinguished by different planes of cell division. However, in the 1950s the meristeme d'attente concept appeared in France which argued that the central region of the apex remained essentially inactive until the onset of flowering. Meanwhile the recognition of zonation patterns in angiosperm and gymnosperm apices assumed growing functional importance. Clonal analysis based on chimeral structures in the meristem recognized the presence of initial cells but also their replacement. Surgical experimentation and nutrient culture of excised apices have stressed the autonomy of the shoot apex and its role in shoot development. Present molecular genetic analysis may help to resolve some of the persistent questions.

D1.3 The shoot apical meristem and organization of primary vascular architecture.

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The shoot apical meristem is a dynamic system, continuously producing new leaf primordia and the primary tissues of the stem. The close correspondence between the positions of leaf primordia and the early appearance of the procambial strands that will later become their leaf traces has led to the suggestion that vasculature in the older parts of the shoot could provide an inductive signal for the placement of leaf primordia (Esau, 1965; Larsen, 1983; Lyndon, 1998). Analyses of the spatial patterns of expression of genes that appear to function early in the development of procambium provides evidence for the formation of vascular pattern elements prior to detection of the corresponding leaf primordium: the *PINHEAD/ZWILLE* gene is expressed in narrow strands of tissue below the site of incipient primordia at least two plastochrons before leaf initiation (Lynn et al., 1999), while the *Arabidopsis thaliana HOMEOBOX-8* (*AtHB-8*) gene is expressed in the position of leaf trace procambial strands one plastochron before leaf initiation (Kang et al., 2003). The *AtHB-8::GUS*-expressing strands form a reticulate pattern within the shoot, with each leaf trace derived from two adjacent vascular sympodia. Shoot phase change is marked by alterations of this fundamental pattern at the juvenile-adult and vegetative-reproductive transitions. The longitudinal discontinuity in *AtHB-8::GUS* expression mirrors the later pattern of xylem differentiation and indicates that one function of this gene might be to define the xylem components of vascular radial pattern The strong correlations between the three-dimensional architecture of the vasculature within the shoot and phyllotaxis generated on the shoot apical meristem suggest, at the least, that mechanisms regulating phyllotaxis also organize shoot vascular pattern in a highly coordinated manner.

D1.5 In-vitro development of shoot meristems of conifers

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Studies conducted during organogenesis and somatic embryogenesis of spruce (*Picea glauca)* have revealed that development of shoot apical meristems (SAMs) in culture is accompanied by precise structural and metabolic changes. These changes are generally similar to those observed during in-vivo formation of SAMs, although differences are sometimes observed. In spruce somatic embryos, aberrations in the architecture of the SAM are related to poor culture conditions and result in low regeneration frequency. Optimizations of the culture environment will certainly benefit from a broader understanding of the molecular mechanisms regulating the formation and maintenance of SAMs. Our understanding of the gene interactions governing SAM development in conifers is very limited compared to the flowering plants, where some of the components required for the formation and maintenance of the SAM have been identified,. The expression of a member of the KNOX class of homeobox genes (*HBK1*) identified in spruce is restricted to the apical cells of the SAM and can be used as a good molecular marker for estimating meristem quality in spruce somatic embryos. We have conducted a series of cDNA microarry experiments in spruce lines differing in embryogenic potential in a continuing effort to find novel molecular markers for SAM development. Among the potential candidate genes we have isolated a full length ZWILLE-like gene, which is only expressed in the meristematic cells of developing spruce embryos. This gene is homologous to the ZWILLE genes of rice and *Arabidopsis* which play an important role during SAM development and maintenance. This homology suggests that meristem regulation in angiosperms and gymnosperms may be under the control of a similar set of genes and the supporting data will be presented.

D2. Phytochemicals in human health research: bioactivity vs. physiological relevance.

D2.1 Characterization of anthocyanins in saskatoon fruit to determined the antioxidant potential of the fruit

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The objective of this research was to identify and quantify the level of the major anthocyanins (antioxidants) present at fruit maturity in order to determine the nutraceutical potential of Saskatoon fruits. Antioxidants protect cells from damaging free radicals and from stress in general, and they've been implicated to be beneficial to patients with heart disease and cancer. Liquid chromatography-mass spectroscopy (LC-MS) along with high pressure liquid chromatography (HPLC) techniques were used to identify the specific types of anthocyanins that occur in the fruit of 5 different cultivars of saskatoon during development and at maturity. A number of anthocyanin- and flavanoid-type compounds were detected by HPLC analysis. Confirmation of compound identity was obtained using LC-MS. All cultivars examined (Thiessen, Northline, Smoky, Pembina, and Honeywood) showed similar anthocyanin developmental profiles. Four anthocyanins were identified with three of these anthocyanins present mainly at fruit maturity in all 5 cultivars. Although saskatoons have a relatively simple anthocyanin profile compared to that of blueberries, they contain a substantial amount of total anthocyanins per fruit. This work was supported by AVAC.

D2.4 Human Health and Medicinal Plants: Quality and Efficacy of Plant Based Medicines

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Health is a state of complete physical, mental, spiritual and social well-being and not merely the absence of disease or infirmity. Plants are by far the most important source of natural therapeutics, and their role in enhancing the longevity and quality of life is gaining wider acceptance in the Western world. This increased interest in natural remedies has also brought about the great challenge of maintaining a balance between the demand of expanding markets for plant-based medicines and the need to protect medicinal biodiversity. The development of effective cultivation technologies that define plant yield in terms of both biomass and medicinally active

phytochemicals is, therefore, extremely important for long-term conservation of medicinal plants and their sustainable use. A series of problems with medicinal plant products have prompted the introduction of regulations to ensure the quality and safety of medicinal plant products in Canada. The problems have included contamination with biological and environmental pollutants, quantitative and qualitative variations of bioactive compounds, adulteration with misidentified species, and the unsustainable harvest. A variety of protocols for the selection of superior clones, in vitro conservation and multiplication have been established for an international collection of unique medicinal species maintained at the University of Guelph. Our technology of in vitro - in vivo culture systems provides large quantities of medicinal plant tissues with optimized medicinal content. Phytochemically distinct Germplasm lines developed for widely used medicinal species, such as Huang-qin (*Scutellaria baicalensis*) and St. John's wort (*Hypericum perforatum*), have been evaluated for inhibition of aflatoxin mutagenicity in rat model systems and human cancer cell lines. As such, the technologies developed are expected to provide consistent and effective commercial products of medicinal plants in the near future.

D3. Conservation and diversity I

D3.1 Dammed Rare Plants! An exploration of factors on dam reservoirs limiting survival and establishment of two Atlantic Coastal Plain flora species at risk.

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The Atlantic Coastal Plain flora (ACPF) is a unique assemblage of plants requiring infertile shorelines maintained by fluctuating water levels. 11 of the 66 ACPF species found in Nova Scotia are listed under COSEWIC as species at risk. ACPF habitat has been greatly reduced throughout its range from the Gulf Coast of Texas to southern New England and Nova Scotia due to damming and lakeshore development. Damming results in the creation of head ponds and reservoirs, the latter emptied periodically to keep head pond levels constant. Though the static head ponds are not suitable habitat for ACPF, as water fluctuations are necessary to maintain appropriate shoreline, reservoir lakes do appear to be good habitat. Transplant experiments have been conducted to examine survival and establishment requirements of two at risk ACPF species *Coreopsis rosea* Nutt. (endangered) and *Hydrocotyle umbellata* L. (threatened). Preliminary results suggest that dispersal may be the main limitation to colonization of reservoir lakes for these species, however differences have also been found in fall and winter conditions on reservoir lakes which may influence over-wintering success at these sites. Preliminary results from an experiment on cold tolerance of these species suggests that *H. umbellata* is more susceptible than *C. rosea* and may not be able to survive the cooler fall and winter conditions experienced on reservoir lakes that have a later rise in lake levels. Altering the management of reservoir lakes may improve their suitability as habitat for ACPF communities and in particular for these at risk species.

D3,2 More Than One Way to Grow a Plant: Establishing Viable *Ex Situ* Populations of 3 Rare and Endangered Plants.

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The Limestone Barrens of the Great Northern Peninsula of Newfoundland is a hotspot of plant diversity. This critical habitat contains three world endemics; *Salix jejuna* (endangered), *Braya longii* (endangered) and *B. fernaldii* (threatened). The high level of threat these plants are faced with, coupled with their low numbers and the restricted nature of the populations, make them very vulnerable.

Several methods are being used to establish viable populations and maintain the genetic diversity of all three species in off site (or *ex situ*) facilities. These methods include:

1. Developing protocols for the persistence of live plants at the Memorial Botanical Garden. Experimentation with substrate characteristics, drainage and environmental conditions is ongoing in order to optimise growth of these plants.

2. Seed banking. Both *Braya longii* and *B. fernaldii* seeds remain viable for several years

if stored properly. Originally *Salix jejuna* seed was assumed to have low longevity and viability. However, preliminary work on *S. jejuna* indicates that seeds remain viable for much longer; this will play a critical role in our ability to develop a seed bank that preserves individuals from their entire distribution.

3. Developing tissue culture protocols for all three species. Not only does tissue culture allow these plants to grow in a sterile environment, (free of bacteria, fungus and insects) it allows the mass propagation of individuals from a very small amount of tissue.

The development of these protocols and the building of an *ex situ* collection will be invaluable as a failsafe in the event that the natural populations are extirpated. They will also be used for experimentation to reduce impact on natural populations and for reintroduction trials at suitable sites. *Ex situ* populations will arrest the erosion of the genetic diversity that is occurring in nature.

D3.3 Assessing levels of Genetic Diversity in Threatened Populations of Western Spiderwort (*Tradescantia occidentalis)*

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Tradescantia occidentalis (Western Spiderwort) grows in the dry sand hills of the mixed grass prairies on partially stabilized sand dunes. This species is widely distributed throughout the central United States but is restricted to four localities in Canada, two of which are isolated from the U.S. distribution. Western Spiderwort is listed as endangered in Saskatchewan and threatened in Canada. This systematic research proposal is designed to: 1) Assess habitat and population status of Western Spiderwort and 2) Investigate genetic diversity among and between populations in an effort leading to better management and conservation practices of this species. *In-situ* information will be obtained from field surveys to estimate population status and primary plant association. *Ex-situ* investigation will involve herbarium specimens and leaf samples collected from each locality. DNA will be extracted from selected herbarium and field-collected specimens and used in molecular studies. Various methods of data generation will be explored to determine genetic divergence. Molecular techniques will include amplified fragment length polymorphisms (AFLP's), inter-simple sequence repeats (ISSRs), and microsatellites. Inter- and intrapopulation genetic diversity will be estimated using Nei's and Dice's genetic similarity coefficients together with analysis of molecular variance (AMOVA). The knowledge generated in this study will contribute to a better understanding of endangered species, in particular the Western Spiderwort, and could be used as a model in long-term conservation and biodiversity studies in Saskatchewan.

D3.4 Interactions between plant competition, herbivory, and abiotic stress in an arctic-alpine community

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Interactions among plants vary from facilitative to competitive. Prevailing theory suggests facilitation will be most prevalent under stressful conditions, but there can be significant variation in the net effect of interactions. The local-scale factors that cause this variation are unknown. We investigated how local-scale herbivory, productivity, and abiotic stresses affect the net effect of plant interactions in a single arctic-alpine community.

The alpine meadows of the southwest Yukon are ideal for studying these interactions. Small mammal herbivores graze primarily close to talus rock piles, creating a strong herbivory gradient. Productivity and abiotic gradients also occur within short distances.

Individuals of two species, *Artemisia norvegica* and a *Carex consimilis*, were subjected to all combinations of herbivory, competition and water availability. These treatments were imposed using the natural herbivory gradient, along with experimental manipulations of neighbour density and water. Plant growth was followed for two growing seasons by measuring leaf demography and plant biomass.

Both species had increased leaf births and numbers of live leaves when released from competition. Distance from talus had strong effects. *Artemisia* individuals far from talus increased in stem mass with competition release, those near the talus did not. *Carex* individuals increased numbers of leaves more close to talus than far. Distance effects may be due to herbivory or abiotic differences; NO_3 flux and soil depth are greater, and snowmelt

averages 6 days earlier far from talus. Watering effects were subtle. Relationships between competition, productivity and community composition will be analyzed.

Predicting the outcome of multiple interactions on plant growth is challenging, especially with the small-scale variability present in arctic-alpine systems. It is clear that in this system, for these two species, that competition and its interactions are limiting growth of plants.

D3.5 Genetic diversity of *Arabidopsis thaliana* populations alters stand characteristics and feeding by *Trichoplusia ni*

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Although the relationship between species diversity and ecological function has been well studied over the last decade, only recently has attention turned towards understanding the impact of intraspecific genetic diversity on basic ecological processes. A variety of ecologically important plant traits, such as competitive ability, vary among genotypes within a species. As a result, it is reasonable to expect that population level responses to ecological challenges such as herbivory and N-stress will be influenced by the genetic diversity of the population. To test this idea, we conducted a large experiment in which we planted either genetic monocultures or mixtures (9 ecotypes per population) of *A. thaliana* at three densities (1, 9, 36 plants/pot) and two levels of fertilization (+/-). Germinating seed of known genetic identity was placed in known locations within each population (4" pots; sand/soil mix), with all populations placed outside during the summer. After four weeks, an herbivore treatment was imposed upon some populations, consisting of adding five second instar *T. ni* individuals to each population. The insects were removed after approximately one week of feeding. In total, this experiment consisted of 98 treatments, 880 pots, 2,000 insects, and 12,000 plants. Genetic diversity had significant effects on a variety of key measures. Plant genetic diversity increased herbivore performance (*T. ni* growth), with the magnitude of this diversity effect increasing with fertilization of the plant population. At low plant density, genetic diversity had no effect on overall plant population growth, regardless of fertilization or the presence of herbivores. At high density, genetic diversity enhanced the positive effects of fertilization in the absence of herbivores. However, genetically diverse populations did worse than monocultures when fertilized in the presence of herbivores. Overall, we found strong evidence that genetic diversity of the plant population has functional consequences for stand-level processes, as well impacting herbivore performance. Additionally, the impacts of diversity appear to be dependent upon other key ecological factors, such as plant density and resource availability. These findings suggest that intraspecific variation in plant traits will have significant impacts on ecological function.

D3.6 Measuring plant competition: applying yield-density models to multi-species systems.

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When plants compete for environmental resources, mean size per plant tends to decline as population density increases. The rate of decline is a measure of the plants' ability to compete, and this concept was first exploited by Kira's group in Japan in the 1950s. Their work was done with monocultures, and in the early 1980s others applied this approach to two-species associations. Since that time, inverse yield-density relationships have been used to measure competition in many binary species mixtures. Here we explore the potential to extend this procedure to three- and four-species associations, grown in separate experiments. The three-species mixtures were composed of barnyardgrass (*Echinochloa crus-galli*), green foxtail (*Setaria viridis*) and redroot pigweed (*Amaranthus retroflexus*). The four-species mixtures contained corn (*Zea mays*), wheat (*Triticum aestivum*), bean (*Phaseolus vulgaris*) and beet (*Beta vulgaris*). For both the three- and four-species associations, inverse yield-density relationships were able to describe the response of each of the component species to crowding. All 9 within- and between-species influences were significant in the three-species associations; all but one of the 16 within- and between-species influences were significant in the four-species system. Interactive effects of species densities were not significant, so

competitive influences were separately and independently assessed through this procedure. In the four-species association, three positive between-species influences were detected, indicating the occurrence of some facilitation. Since very large experimental arrays would be required, it is unlikely that this approach can be used for associations containing more than four species.

D4. Salinity stress

D4.1 Probing the mechanism underlying PR 10-mediated enhanced germination of *B. napus* under saline conditions.

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Salinity is a major abiotic stress that leads to a number of cellular and molecular changes eventually culminating in plant death. Studies of salinity-induced proteome-level changes in pea roots lead to the identification of several proteins including several members of a pathogenesis-related (PR) 10 protein family. Furthermore our studies showed that constitutive expression of a pea PR 10.1 gene in canola (*Brassica napus*) enables the transgenic plants to germinate and grow better under saline conditions. Our current studies are aimed at understanding the mechanism underlying this PR 10-mediated enhanced germination/growth under salinity stress. In this poster we present results from comparative studies at the morphological-, proteome- and biochemical-levels that may lead to a better understanding of the role(s) of PR 10 proteins in mediating salinity stress responses.

D4.2 Functional analysis of sodium transport genes expressed with both spatial and temporal control in rice.

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Development of salinity tolerance in crop plants has remained elusive due to the multi-genic nature of the trait and its physiological complexity. Two major problems hampering efforts to produce salt-tolerant plants through expression of transgenes include:

1. Cell-types must respond individually to salt stress to minimise the amount of Na^+ delivered to the shoot; and

2. Transgenes are typically expressed in plants constitutively, which is likely to be counter-productive to developing salt tolerance and means, in low-stress conditions, expensive metabolic processes may be active, thus reducing photosynthate available for grain production.

To address these issues, 824 rice (*Oryza sativa* L. cv. Nipponbare) GAL4-GFP enhancer trap lines were screened with a mild (i.e. agriculturally relevant) salt stress (40 mM NaCl) to identify enhancer elements which are up- or down-regulated by salt stress in specific cell-types. Expression of *gfp* (and therefore, enhancer element expression) was regulated by salt stress in 18 lines (12 up-regulated, 6 down-regulated). Salt transport transgenes (e.g. *AtSOS1, AtHKT1, AtNHX1, ScNHA1, AtAVP1, ScENA1*) will be fused to the GAL4 UAS (upstream activating sequence), in a modified Gateway® destination vector, and transformed into the lines previously identified in the screen, to allow their regulation by salt stress in specific cell-types. Lines expressing salt transport transgenes will be characterized by Na^+ accumulation in the shoots and roots (via flame photometry), $^{22}Na^+$ flux experiments, fresh weight measurements; and lines will be documented by digital photography and confocal microscopy.

D4.3 Changes in the physiology and metabolome of *Thellungiella salsuginea* in response to osmotic stress

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Thellungiella salsuginea is a subarctic crucifer closely related to *Arabidopsis thaliana* and *Brassica napus*. In contrast to *A. thaliana* and *B. napus*, Thellungiella is highly tolerant of salt, drought and freezing conditions and so provides an excellent model for the study of osmotic stress tolerance mechanisms. Using measurements of plant water status

(water/solute potentials and relative water content), transpiration, stomatal conductance and photosynthesis we have found that Thellungiella can withstand salt levels in excess of 300 mM NaCl and will recover and grow following severe water deficits. Leaf water and solute potential measurements from salt-stressed plants and plants rewatered following drought shows that Thellungiella accumulates solutes under osmotic stress and undergoes "osmotic adjustment", a response characteristic of plants that tolerate drought and saline conditions. Metabolic profiling is being used to identify compatible organic solutes associated with osmotic stress tolerance in this plant. We are using gas chromatography-mass spectrometry (GC/MS) to profile polar metabolites present in leaf tissues of well-watered and osmotically stressed Thellungiella. Plants grown under comparable conditions yield similar and reproducible profiles with over 300 components routinely detected in each chromatographic run. Among solutes that increase in abundance under stress are proline and galactinol; both have been implicated as osmoprotectants in other plants. Proline levels increase in a salt-responsive manner when Thellungiella is irrigated with 100 to 500 mM NaCl providing correlative evidence for a protective role in this species. Profiling metabolites by GC/MS can provide insight into the traits underlying the abiotic-stress tolerant phenotype of Thellungiella.

D4.4 Ribosomal Protein S15A: Heterogeneity within the small ribosomal subunit of *Arabidopsis thaliana*

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As a ribonucleoprotein complex, the plant ribosome consists of four ribosomal RNAs (rRNAs) and 75-92 ribosomal proteins (r-proteins), depending on the species. *Arabidopsis thaliana* r-protein genes exist in multi-gene families ranging in size from two to seven transcriptionally active members. The cytosolic *RPS15A* gene family consists of four members that, at the amino acid level, share 87-100% identity. Using semi-quantitative RT-PCR we have shown that transcript abundance differs both spatially and temporally among the four *RPS15A* genes in non-treated *A. thaliana* tissues and in seedlings following a variety of abiotic stresses. To determine the level of ribosome heterogeneity with respect to r-protein RPS15A an initial series of *CaMV 35S* :: *RPS15A-1, -2* or *-4* cDNA :: *fluorescent reporter gene* (i.e. *GFP*, *YFP* and *RFP*) constructs will be used to confirm functionality in tobacco. A second series of *RPS15A-1, -2* or *-4* cDNA :: *florescent reporter gene* constructs under respective *RPS15A* upstream regulatory region transcriptional control will then be used to generate stably transformed *A. thaliana* lines. Each differentially labeled RPS15A isoform will be visualized *in planta* using the Zeiss LSM 510 Meta confocal microscope; results will be discussed.

D4.5 Salt-responsive expression of *α-Dioxygenase* genes in *Arabidopsis*

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Alpha-DIOXYGENASES catalyze the conversion of linolenic and other fatty acids into their 2(*R*)-hydroperoxy derivatives, which are further converted to 2-hydroxy fatty acids and other oxylipins. The oxylipin products generated by action of a-DOX1 may act as signaling compounds for the induction of genes and enzymes of importance for plant defense. *Arabidopsis* possesses two genes that encode a-DOX enzymes. The expression of *a-DOX1* is associated with the local response to pathogen infection, where it may play a role in protecting cells from oxidative stress. Both *a-DOX1* and *2* are responsive to salt stress in *Arabidopsis* shoot and root tissues. Regulation of salt-responsive *a-DOX1* and *2* expression, as indicated by analyses of hormone deficient or response mutants and exogenous hormone treatments revealed that abscisic acid, salicylic acid, and ethylene play a positive regulatory role. T-DNA insertion mutants with reduced or knocked out expression of *a-DOX1* and *2* are being analysed to ascertain whether *a-DOX1* lays a role in the salt stress response in *Arabidopsis*.

D4.6 The influence of salt stress on genomic stability

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While the negative physiological effect of salt stress on plant is well documented its influence on genome is not yet well

understood. Effect of salt stress on genomic stability was estimated using transgenic *A. thaliana* plants containing *ß-glucuronidase* based substrate for homologous recombination (HR). It allowed us to score HR events and calculate rate of HR per single cell genome, recombination rate (RR). RR was related to the abundance of DNA double-strand breaks (DSBs) – the substrate for HR, and to the level of *AtKu70* and *AtRad51* transcripts – two key DSB repair genes. When we analyzed the effect of different concentrations of NaCl we found linear correlation between the salt concentration and RR. This finding was consistent with 20% increase of DSBs level in 75mM NaCl treated plants as compared to untreated control. While application of NaCl in range of 0 – 100mM triggered 30% increase of level of transcription of *Rad51* the level of *Ku70* remained steady. Further analysis showed that effect of salt stress on genome and HR was mainly due to Cl^- ions. Similar concentration of Cl^- and Na^+ induced HR 5- and 0.75-fold, respectively. Analysis of progeny of control (PofC, no NaCl) and progeny of plants exposed to 25 or 75mM of NaCl (PofS) showed that salt-induced HR increase was inherited. While level of DSB was comparable between PofS and PofC, RR was induced in absence of stress by factor 1.9 and 2.4 in progeny of 25 and 75mM treated plants respectively. Similarly, *Rad51* was upregulated and *Ku70* was down regulated in PofS as compared to PofC. Finally we observed significant increase in salt tolerance of PofS plants coupled with 15% higher global hypomethylation of the genome. Together our data showed that salt stress does not only change the genome stability but also contributes to the fast adaptation to new environmental conditions by precise management of DNA repair machinery and generation of epigenetic changes within the genome.

D5. Biochemistry II

D5.1 Genetic engineering of fatty acid desaturation in *Arabidopsis thaliana* and *Brassica napus*

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Development of vegetable oils with ultra-low saturated fatty acid (FA) content will provide a competitive edge to the oilseed extraction and processing industry due to the notion that low saturated FA content is associated with increased health benefits. During seed development, desaturases catalyze the formation of both monounsaturated and polyunsaturated FAs. We attempted to reduce the saturated FA content of canola oil and that of the model plant, *Arabidopsis thaliana*, by engineering FA desaturase action during seed development. Introduction of a cDNA encoding palmitoyl (16:0)-acyl carrier protein desaturase from a forest vine (*Doxantha unguis-cati* L.) resulted in reduction of 16:0 and a concomitant increase in palmitoleic acid (16:1) in both *Arabidopsis* and canola. In addition, several other unusual FAs were formed in the seed oil of the transgenic plants. Although 16:0 was reduced in transgenic plants, an increase in other saturated FAs, especially stearic acid (18:0), resulted in no change in the overall saturated FA content. Expression of a non-specific FA desaturase from blue-green algae, however, did result in a significant reduction in the saturated FA content of canola oil. These investigations set the foundation for the development of vegetable oil with ultra-low saturated FA content for the oilseed industry.

D5.2 Enhancing growth and seed yield in canola by suppression of deoxyhypuysine synthase expression via vacuum-infiltration of *Agrobacterium*.

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A full-length cDNA clone encoding canola (*Brassica napus* cv Westar) deoxyhypusine synthase (DHS) was isolated from a cDNA expression library prepared from senescing leaves. DHS mediates the first of two enzymatic reactions that convert inactive eukaryotic translation initiation factor-5A (eIF-5A) to an activated form, thought to facilitate selective mRNA transportation from the

nucleus to the cytoplasm. Transgenic canola seeds expressing the antisense 3'-UTR canola DHS cDNA under regulation of the constitutive cauliflower mosaic virus 35S (CaMV-35S) were obtained by vacuum infiltration of canola inflorescences by modifying the protocol developed for *Arabidopsis*. The efficiency of transformation was enhanced by removal of new flowers that formed after the plants had been vacuum-infiltrated, and transformation rates of 50 to 60% were routinely obtained. Transgenic plants had reduced levels of leaf DHS protein and exhibited delayed natural leaf senescence. Suppression of DHS also increased leaf size by 1.5- to 2-fold and resulted in increases in seed yield of up to 65%. This was attributable in part to an increase in the size of the siliques, which were on average 18% to 26% longer than wild-type siliques depending on the line. When wild-type and transgenic plants were grown in 6-inch pots, the increase in seed yield accruing from suppression of DHS was ~4.5-fold greater than when the plants were grown in 12 inch pots. Thus suppression of DHS appears to ameliorate the effects of sub-lethal stress engendered by growth in small containers. The increase in seed yield for transgenic plants translates into a corresponding increase in seed oil content based on measurements of triacylglycerol, and there was no change in the fatty acid composition of the oil in transgenic seeds.

D5.3 Cloning and biochemical characterization of a novel *Arabidopsis thaliana* methyltransferase involved in phosphocholine synthesis

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Choline is a ubiquitous metabolite in plants as the membrane phospholipid phosphatidylcholine (PtdCho) or as free choline. Many plants can oxidize choline to produce the osmoprotectant glycine betaine in response to environmental stresses such as cold, salinity, and drought. In Arabidopsis, the enzyme phosphoethanolamine *N*-methyltransferase (PEAMT) sequentially methylates phosphoethanolamine three times (through intermediates phosphomethylethanolamine (PMEA) and phosphodimethylethanolamine (PDEA)) to yield phosphocholine (PCho); each transfer requiring *S*-adenosyl-L-methionine (SAM) as the methyl donor. Arabidopsis PEAMT is encoded by the gene at locus At3g18000. A BLAST analysis of the Arabidopsis genome reveals two highly similar genes annotated as encoding putative PEAMTs, both on chromosome 1 (At1g48600 and At1g73600). A cDNA library from Arabidopsis prepared in the expression vector pFL61 was used to identify clones that functionally complement a yeast mutant that is auxotrophic for choline. cDNAs corresponding to the genes at the loci At3g18000 and At1g48600 rescued the yeast mutant. We inferred that At1g48600 encodes a methyltransferase with biochemical properties similar to PEAMT. The substrates used by the gene products corresponding to At3g18000 and At1g48600 were compared in vitro using cell-free extracts. In contrast to PEAMT encoded by At3g18000, the methyltransferase encoded by At1g48600 does not methylate PEA; it only methylates PMEA and PDEA. Hence we designated this enzyme as phosphomethylethanolamine *N*-methyltransferase or PMEAMT. The properties of the two enzymes differ: for PEAMT apparent Kms were 0.3 mM and 0.14 mM for PMEA and PDEA, respectively, while for PMEAMT the apparent Kms were 0.16 mM and 0.03 mM for PMEA and PDEA, respectively. The physiological role of the product encoded by At1g48600 remains to be determined.

D5.4 Calcium and pH sensitivity of phospholipase D in tomato fruit protoplasts and sub-cellular preparations

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Phospholipase D (PLD), a key enzyme involved in membrane phospholipid catabolism, exists in several isoforms designated alpha, beta and gamma with reportedly distinct calcium- and pH- stimulatory characteristics. PLD alpha was the major distinguishable isoform present in the mitochondrial and microsomal membrane fractions, as well as the cytosol, isolated from tomato fruit. All enzyme preparations were stimulated by calcium at low micromolar levels (1-10), though further stimulation was observed at millimolar levels. Maximal

stimulation was observed at pH 4 for both mitochondrial and microsomal PLDs. A much higher stimulation of activity was observed if a pH gradient was established across the membrane. Tomato protoplasts were labelled with fluorescent phospholipid analogues and examined using confocal microscopy. Incorporation of phospholipids into the plasma membrane resulted in a ring shaped fluorescence emission, which was stable for over 30 minutes. Addition of calcium at 10 micromolar in the presence of a calcium (A23187) ionophore resulted in immediate quenching of fluorescence indicating the catabolism of fluorescent phospholipids by enhanced PLD activity. Thus, at physiological levels of calcium, PLD can become membrane bound catalyzing the hydrolysis of phospholipids.

D5.5 Expression analysis of select *PREPHENATE DEHYDRATASE-LIKE* (*PDL*) genes in *Arabidopsis thaliana*.

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A family of six genes identified in *Arabidopsis* have been designated *PREPHENATE DEYHYDRATASE-LIKE* (*PDL*), since they have very similar sequences as those described for bacterial *PREPHENATE DEHYDRATASE*s (*PDT*s). PDT is a catalytic enzyme which acts in the classical pathway of phenylalanine biosynthesis. Based on biochemical evidence, however, it is expected that phenylalanine in higher plant species is synthesized via the arogenate pathway. Therefore one would predict the presence of an arogenate dehydratase (ADT) instead of PDT as a catalytic enzyme. Unfortunately, all annotated sequences in any sequence database list PDT but no ADT sequences for plants. One can only assume at this point that both enzymes are encoded by genes having very similar sequences.

The PDLs for *Arabidopsis* and other plants have been grouped into three different subfamilies based on sequence comparisons. All PDLs have two conserved regions: a proposed catalytic domain that is highly similar to bacterial PDT domains, and an ACT or ligand binding domain predicted to regulate enzymatic activity by feedback inhibition. In addition all plant PDLs have a more variable sequence at the 5' end, a putative signal peptide sequence allowing for subcellular targeting to various organelles (most likely chloroplasts and mitochondria).

To date it is unclear why *Arabidopsis* needs six *PDL* genes to fulfill its requirements for phenylalanine synthesis. However, phenylalanine can either been used directly for protein synthesis, or as a precursor for secondary metabolites involved in stress response or the formation of structural components. These diverse needs might be met by individual *PDL*s and one can therefore anticipate that the expression patterns for the six *PDLs* will be distinct and different.

To analyze the expression patterns of *PDLs* in *Arabidopsis* an RT-PCR approach was chosen. In such an approach it is essential to differentiate between amplifications which use RNA or DNA as a template. Typically, by choosing primers which span a region containing an intron, RNA and DNA products can be easily distinguished by size. However, only two of the six *PDL*s contain introns making this approach problematic. We will present data using a modified RT-PCR protocol to study the expression of individual *PDLs* in various tissues types.

D5.6 Modulating the papain inhibitory activity of a tomato cystatin by single mutations at a positively selected amino acid site

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Proteinase inhibitors of the cystatin superfamily regulate cysteine proteinases in various biological processes. In plants, cystatins play several important functions, including notably the inhibition of exogenous cysteine proteinases during herbivory or infection. We used here a maximum likelihood approach to assess whether plant cystatins, like other proteins implicated in host-pest interactions, have been submitted to adaptive evolution –or positive selection– during the course of their evolution. Several amino acid sites were identified as being under positive selection in cystatins from either Poaceae and Solanaceae. These hypervariable sites are located at strategic sites on the protein, each side of the conserved glycine residues in the protein's N-terminal trunk; surrounding the

larfav motif, a sequence of unknown function, conserved among plant cystatins; and within the first (...qxvxg...) and second (...pw...) inhibitory loops entering the active site of target enzymes. Supporting the assumption that positively selected sites are indicative of amino acid sites implicated in functional diversity, mutants of the 8th cystatin unit of tomato multicystatin, including alternative residues at a positively selected site in the N-terminal trunk, exhibited variable affinities for the model cysteine proteinase, papain. In contrast, changing an adjacent, neutrally-selected site for the same residues did not alter papain inhibitory activity of the cystatin, suggesting a negligible impact for this amino acid site at the functional level. Overall, these observations suggest that cystatins in plants have been under selective pressure to evolve in response to predatory challenges by herbivorous enemies. They also point out the potential of site-directed mutagenesis at positively selected sites for the rational design of cystatins with improved binding properties.

D6. Crop resistance to disease

D6.1 Evidence for a second seedling leaf rust resistance gene in the Thatcher-*Lr1* near-isogenic wheat line RL6003

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It was previously thought that the Thatcher-*Lr1* near-isogenic wheat (*Triticum aestivum*) line RL6003 only had a single gene, *Lr1,* for seedling resistance to wheat leaf rust (*Puccinia triticina*). However, a number of *P. triticina* isolates collected from across Canada during 2004 produced an unusual mesothetic resistant host response when inoculated onto RL6003. The isolates were classified as avirulent to the *Lr1* differential line RL6003 but virulent to the *Lr2a*, *Lr2c* and *Lr3* differential lines which resulted in a code of "K" on this first set of four differential lines. The mesothetic reaction was produced by these "K" isolates on most plants in the RL6003 line but some plants were fully susceptible to these isolates. RL6003 however was uniformly resistant, with an infection type of "0;" which is characteristic of *Lr1*, when inoculated with other *Lr1* avirulent isolates. The "K" isolates were fully virulent when inoculated onto other lines reported to carry *Lr1* such as Glenlea, Roblin, and RL6028, which is a Thatcher near-isogenic line carrying *Lr1* from a different source than that used to create RL6003. Based on these results the "K" isolates are virulent to *Lr1*, but are avirulent to a second gene in most plants of the RL6003 line. This evidence suggests that most plants in the RL6003 line contain two seedling leaf rust resistance genes, *Lr1* plus a second unidentified resistance gene. This second gene may be linked to *Lr1,* since it is not found in Thatcher but was retained through five backcrosses to Thatcher, while selecting for *Lr1* at each generation during the creation of RL6003. This second gene may have simply been undetected for many years of testing RL6003 because isolates with virulence to *Lr1* and avirulence to the second gene may have been rare.

D6.2 Identification of an Amplified Fragment Length Polymorphism (AFLP) marker linked to a spot blotch resistance gene in barley using bulked segregant analysis.

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Spot blotch, caused by *Bipolaris sorokiniana* (Sacc.) Shoemaker is one of the major leaf diseases of barley in western Canada, especially in the eastern prairie region. TR 251 is a barley line with a good level of resistance to all current pathotypes of spot blotch. A doubled haploid (DH) population derived from the cross (TR 251/CDC Bold) was used to determine the genetics of resistance to spot blotch in TR 251. The DH population was assessed for infection response to different Manitoba pathotypes of *B. sorokiniana*. Based on phenotypic data, the population was classified into 3 classes: resistant, moderately susceptible, and susceptible. The most resistant and most susceptible lines were selected for the genetic study using the bulked segregant analysis method. A partial linkage map of the genome was constructed using polymorphic microsatellite and AFLP markers. AFLP markers were assigned to chromosomes by anchoring them with locus-specific microsatellite markers. An AFLP marker was mapped 15 cM from a resistance gene on the short arm of chromosome 3H. If the presence

of this gene for spot blotch resistance can be confirmed in further studies, it could be used for marker assisted selection in Canadian barley breeding programs.

D6.3 Influence of Rootstock, Incubation Temperature and Duration of Incubation on Bacterial Canker Severity Caused by *Pseudomonas syringae* pv. *syringae* in Peach

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In one field experiment using peach trees (*Prunus persica* (L.) Batsch, cvs. Flavorcrest and Loadel) grafted on three different rootstocks, Nemaguard, K119-50 (*P. dulcis* × *P. salicina* hybrid), and P30-135 (*P. persica* × *P. salicina* hybrid), significantly different size lesions occurred after inoculation with *Pseudomonas syringae* pv. *syringae* van Hall. Peach stems on Nemaguard developed the longest lesions and stems on K119-50 the shortest lesions among all three rootstocks. Peach stems on Nemaguard had significantly lower bark calcium and higher bark nitrogen content than stems on K119-50 and P30-135. A negative correlation was found between lesion length and bark calcium content and calcium/nitrogen ratio. Laboratory experiments with excised peach stems (cv. Ross) on Nemaguard, K119-50, P30-135, Lovell, and Guardian rootstocks growing in a different orchard showed results that were inconsistent with the first orchard. Stems on Nemaguard in this orchard did not significantly differ in bark calcium and nitrogen content compared to those from trees on K119-50 and P30-135 rootstocks, but developed significantly smaller lesions than those on K119-50 and P30-135 rootstocks. Stems on Guardian and Lovell rootstocks also developed significantly smaller lesions than stems from trees on K119-50 and P30-135 rootstocks. Temperature fluctuation during incubation had no clear effect on decreasing lesion length compared to those incubated constantly at 14.4^{o}C, but had significant effects on promoting lesion lengths compared to stems incubated constantly at 0^{o}C.

D6.4 Potential impact of a stem rust race in Kenya with wide virulence on Canadian wheat production

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Stem rust of wheat (*Triticum aestivum*), caused by *Puccinia graminis* f. sp. *tritici*, is a major disease that has been controlled in Canada since the mid-1950's through the use of resistant cultivars. Although the specific gene pyramids present in most Canadian wheat cultivars are unknown, it is likely that a combination of 3 to 5 resistance genes confers the resistance. Recently, a race with virulence on *Sr31*, the predominant gene used by CIMMYT for stem rust resistance, was initially identified in eastern Africa and is likely to spread to other regions. This study evaluated the virulence of this race on 44 designated Sr genes and 26 Canadian cultivars using seedling tests. Preliminary results indicate that this African race has broad virulence to many *Sr* genes and also to most Canadian wheat cultivars. If this race spread to North America, severe stem rust infection could occur on many Canadian wheat cultivars. The potential impact on Canadian spring wheat production in western Canada under severe infection could be significant. However, several cultivars were found to be resistant at the seedling stage. Field testing of predominant Canadian wheat cultivars is currently in progress in Kenya to assess the field reaction to this race with virulence on *Sr31*.

D6.5 Screening for Resistance to Tan Spot, Septoria Nodorum Blotch and Septoria Tritici Blotch in Wheat, Durum and Wild Relatives.

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Leaf spot of wheat in North America consists of a group of diseases involving tan spot, septoria tritici blotch and stagonospora nodorum blotch. A complex of these diseases occurs in nature hence managing leaf spots is difficult. The use of resistant cultivars is the most effective and economical means of controlling leaf spot. None of the present grown wheat cultivars show high level resistance to all the leaf spotting diseases. Hence, to identify new sources of resistance to the leaf spotting diseases, 1100 accessions of durum, wheat and wild relatives were evaluated under controlled environments to virulent races 1, 2, 3, and 5 of *Pyrenophora tritici-repentis* found in North America and two exotic races 10 and 11 found in South America. In addition genotypes were tested with foliar pathogens *Stagonospora nodorum* and *Mycosphaerella graminicola.* New sources of resistance were identified against the three leaf spotting pathogens, *P. tritici-repentis, S. nodorum* and *M. graminicola,* in accessions of *Triticum monococum* L., *T. turgidum* L., *T. dicoccum* L., *T. dicoccoides* Koern., *T. timopheevii* Zhuk., and *T. aestivum* L. including synthetic wheat. Presently breeding efforts are being made to transfer the leaf spot resistance into adapted wheat and durum cultivars.

D6.6 Screening maize for resistance to common rust, eyespot, and northern leaf blight

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Common rust, eyespot, and northern leaf blight are three major leaf diseases on maize in Canada. In a search for new sources of resistance to these diseases, from 1997 to 2004, we screened the resistance of 983, 932, and 969 corn genotypes, including 711, 695, and 694 Canadian genotypes, 272, 237, and 275 exotic genotypes to common rust, eyespot, and northern leaf blight, respectively. All genotypes were planted in two replicates at Agriculture and Agri-Food Canada, Central Experimental farm, Ottawa, Ontario, unless exotic seed supply was limited. Each replicate consisted of 3.8 m long individual rows (76 cm row spacing) for each genotype with 15 plants per row. All genotypes were inoculated with the respective pathogen and the fields were irrigated afterwards to promote disease development. Inoculation techniques and disease severity assessments were developed and standardized for each disease. Pustule and lesion types were used to record the specific resistance to common rust and northern leaf blight. Resistant genotypes were screened again in the following year. As the results, 25, 38, 128, 253, 331, 208; 3, 69, 152, 337, 289, 82; and 10, 44, 100, 188, 310, 317 genotypes had the disease severity at 1.0-2.0, 2.1-3.0, 3.1-4.0, 4.4-5.0, 5.1-6.0, 6.1-7.0 to common rust, eyespot, and northern leaf blight, respectively. All 53 genotypes had the lower severity (=3.0) to rust expressed hypersensitive or resistance pustule types, most were exotic germplasms from United States and Mexico. Canadian genotypes, CF08, CG80, CG95, CO390, CO391, CO420, CO428, CO439 showed lower severity from 3.5-4.0, but without specific resistance to rust. Exotic genotypes Chile 301, NIDERA AX884 (from Argentina), CHN7 (from China), POB.86C5 (from Mexico), Narino 521, MO45, B37, B113, A661, A679, A681, MO47, ND278, and NYLB31 (from USA) showed low severity (=3.0) to eyespot, some Canadian genotypes, such as CF08, CF24, CM105P, CM174, CO353, CO354, CO424, CO439, and CG103 exhibited relatively low severity (=4.0) to eyespot. All genotypes with Ht2, Ht3, and Htm1 showed resistant or moderately resistant lesion types and lower severity (=4.0) to northern leaf blight. The Canadian inbred CO428 exhibited both specific (resistant lesion) and general resistance (3.0) and was identified as a new source of resistance source to northern leaf blight.

D7. Pathogens of Brassica species

D7.1 Comparison of single spore isolation techniques for *Plasmodiophora brassicae*

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Populations of *Plasmodiophora brassicae*, causal agent of clubroot of crucifers, often consist of a range of different pathotypes, which can make their characterization difficult. A number of different techniques for the isolation of single spores of the pathogen were compared, and a simple and efficient protocol was developed to obtain relatively large

numbers of single spore isolates of *P. brassicae*. Spore suspensions were diluted with phosphate buffer (pH 8)to a concentration of 10^3 spores mL^{-1}, and 1 µL drops were visualized under a microscope. Upon confirmation of the presence of only a single resting spore, the roots of susceptible Chinese cabbage 'Granaat' seedlings (germinated on filter paper) were used to soak up the drop directly from the slide surface. Following incubation for two days under darkness in a Petri dish, the seedlings were transplanted to pots filled Metro-Mix soil, which were placed in standing water (pH 6) for a period of 19 days. Subsequently, the pots were removed from the standing water and watered as required. Infection rates as high as 50% were obtained using this procedure.

D7.2 Induction of systemic acquired resistance by *Leptosphaeria biglobosa* in canola to blackleg disease caused by *Leptosphaeria maculans*

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Blackleg disease of canola (*Brassica napus* L.), caused by virulent strains (pathogenicity groups [PG] 2, 3, 4) of *Leptosphaeria maculans* (Desm) Ces. & De Not, was reduced on cv. Westar pre-treated with an avirulent strain of *L. biglobosa* n. sp. (PG1). In greenhouse studies, percentage of lesion area to leaf (PLAL) on cotyledons was less than 1.0% when PG1 was co-inoculated or pre-inoculated with PG2, PG3 or PG4 at 12, 24 and 48 h. PLAL in absence of PG1 was 1.9 to 6.6%. PLAL on the plants pre-inoculated with PG1 and challenged with PG2 on neighboring leaves, at six leaf stage, ranged from 2.9 to 3.9% at 24, 48, 72, 96 and 120 h, which is similar to salicylic acid (SA) pre-treated positive control and significantly less than 5.2% of water pre-treated control. Compared to the water pre-treated control, increases in total activities of chitinase (9.1 fold at 96h), ß – 1, 3 glucanase (4.9 fold at 96h), phenylalanine ammonia lyase (1.8 fold at 72h) and peroxidase (2.6 fold at 96h) were found in leaf tissues pre-inoculated with PG1 and challenged with PG2. The expression of defense-related genes induced by PG1 is under investigation using northern hybridization and real-time PCR. The research indicates the induction of systemic acquired resistance (SAR) by *L. biglobosa*.

D7.3 Investigating the molecular basis of resistance to the necrotrophic fungus *Alternaria brassicae*

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World production of canola (*Brassica napus*) has been increasing rapidly in several countries largely in response to the continuing demand for edible oils and its products. Alternaria blackspot of canola (causal organism *Alternaria brassicae*) severely affects canola production and has caused extensive yield losses in recent years in Western Canada. Resistance to this pathogen in Brassica species has been reported however, the genetic basis for resistance is currently unknown. The characterization of proteins involved in an incompatible *B. napus-A. brassicae* interaction may be an important step towards understanding the molecular basis for disease resistance. We have analyzed the two-dimensional leaf protein profile of a canola line displaying high degree of tolerance to *A. brassicae* and established the identities of nine proteins that are differentially expressed using tandem Mass Spectrometry. The potential roles of some of these proteins in mediating resistance are discussed.

D7.4 Mapping genes for resistance to *Leptosphaeria maculans* in *Brassica juncea*

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Blackleg disease of crucifers, caused by the fungus *Leptosphaeria maculans*, causes extensive yield loss to oilseed rape producers worldwide. *Brassica* species containing the B genome have high levels of resistance to blackleg. *Brassica juncea* F_2 and first backcross (B_1) populations segregating for resistance to a PG2 isolate of *L. maculans* were created. Segregation for resistance to blackleg in these populations suggested that two genes, one dominant and one recessive in nature, were each capable of conferring resistance to blackleg. A map of the *B. juncea*

genome based on segregation in the F_2 population of 325 restriction fragment length polymorphism (RFLP) and microsatellite marker defined loci was constructed and subsequently aligned with previous maps of the Brassica A and B genomes. The gene controlling dominant resistance to *L. maculans* was positioned on linkage group J13 based on segregation for resistance in the F_2 population. This position was confirmed in the B_1 population where the resistance gene was definitively mapped in the interval flanked by pN199RV and sB31143F. The provisional location of the recessive gene controlling resistance to *L. maculans* on linage group J18 was identified using a subset of informative F_2 individuals.

D7.5 *Pseudomonas chlororaphis* strain190 and biological control of Blackleg in Canola: Understanding the array of potential mechanisms involved.

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Pseudomonas chlororaphis 190 (strain Pc190) isolated from canola stubble reduces blackleg of canola (*Brassica napus* L.) caused by *Leptosphaeria maculans* (Desm.) Ces & de Not). Strain Pc190 may use an array of mechanisms as antibiotics, siderophores and induction of plant defense enzymes in biocontrol of blackleg disease. Significant disease control was achieved at cotyledon and 3-4 leaf growth stages of canola with strain Pc190 application in the greenhouse. Strain Pc190 inhibited pycnidiospore germination on canola leaves. Polymerase Chain Reaction analysis showed the presence of biosynthetic genes of phenazine and pyrrolnitrin, which are common bacterial antibiotics involved in biocontrol of several plant pathogens. Assays of *Cladosporium* spore inhibition on Thin Layer Chromatography plates, and agar-diffusible antifungal activity indicated the presence of antifungal compounds in the broth extract of strain Pc190. Gas Chromatography-Mass Spectrometry confirmed the presence of phenazine, hydroxy phenazine and 2-acetamidophenol in the broth extract. Strain Pc190 induced the expression of plant defense enzymes, β-1,3 glucanase and peroxidase. When co-inoculated with the pathogen, strain Pc190 induced 1.5, 7 and 5 fold increase in glucanase activity at 72, 96 and 120h respectively, and peroxidase activity increased over time and peaked at 120h, as compared to strain Pc190alone and pathogen control. Strain Pc190 is a good candidate for biocontrol of blackleg in canola.

D7.6 Susceptibility of *Brassica napus* at different growth stages to *Leptosphaeria maculans* and its relationship to weather conditions.

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Leptosphaeria maculans (Desmaz.) Ces. & De Not. (anamorph: *Phoma lingam* (Tode:Fr.) desmaz.) causes blackleg disease of canola (*Brassica napus* L.). To investigate the susceptibility of different stages of canola to blackleg (PG2), plants at the cotyledon, 3-leaf and rosette stages were placed in a blackleg-infested canola field for one week from the first of June through the 7th of July. The plants were then returned to the greenhouse and allowed to grow to maturity. The plants were rated for disease severity using a 0-5 scale. Disease severity of the plants infested at cotyledon stage was significantly higher than those infested at the two other stages in all 5 weeks of observations. However, the weekly monitored disease severity of the plants infested at cotyledon stage was highly variable, suggesting that the environment is an important factor in disease progress. Also, the susceptibility of the plants infested at the 3-leaf stage was slightly higher than the plants infested at the rosette stage. Disease severity of the plants at different growth stages was positively correlated with total rainfall in each week (r = 0.88, 0.82, 0.66 for cotyledon, 3-Leaf and rosette stages, respectively). Total rainfall was more important than the frequency of the rain events. The importance of temperature was less than that of total rainfall during infestion at different growth stages.

D8. Teaching in the plant sciences

D8.1 Plant biology for non-believers.

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We need new approaches in our teaching to encourage undergraduate students to enter programs in plant biology. This is based on the Univ. of Alberta, but the same situation

may occur in other universities. A big problem is that most of us no longer have an introductory 1st-year botany course. What we probably have is a large introductory biology course that includes some plant biology. At the Univ. of Alberta, we have 2 such courses (cell biology and biological diversity); there is plant biology in both, but more in the diversity course. Our department now offers a 2nd-year course, Fundamentals of Plant Biology, but its enrollment is about 50 and most of the students are already in other programs. We also offer courses specifically designed to attract students; they do not seem to work—most of the students in those courses (as well as the Fundamentals course) are "traffic" rather than students who might become plant biologists or are already in other programs. We have to appeal to some of the students in the large (1st-year), introductory courses—this may be our only opportunity. What can we do to capture their interest? Here are some suggestions. 1) Do not apologize for plants and plant biology. 2) Recognize (but do not say it) that some of your students hate plants or are simply not interested in plants. 3) Be clear and concise in your lectures. 4) Try to exhibit enthusiasm. 5) Use animated images to jazz up your lectures where appropriate. 6) Try to make connections with the laboratory part of the course. 7) Try to use physical models in your lectures (e.g., nucellus, tracheary element, carpel, endodermal cell). 8) Try to avoid unnecessary discussion of plant life cycles during your lectures. 9) Occasionally remind your students that there are many plants used for food, fibre, wood, and pharmaceutical drugs.

E1. Shoot apical meristems (part 2)

E1.1 Visual models of shoot apical meristem development

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The presentation will outline our recent results pertinent to the simulation modeling of shoot apical meristem development. Emphasis will be put on:

- A geometric model of phyllotactic patterns, which can account for transitions between different patterns during development. This model is related to the experimental results of Kwiatkowska and Florek-Marwitz [1999];
- Molecular-level models of phyllotaxis, based on the conceptual model by Reinhardt et al. [2003];
- A model of primordium growth at the shoot apical meristem, inspired by the experimental results of Dumais and Kwiatkowska [2001];
- A model of primary vascular patterns in shoots, related to the results of Kang et al. [2003].

The presentation will be concluded with a discussion of the role of simulations and visualizations in the studies of shoot apical meristem development.

Some of the results have been obtained in collaboration with Enrico Coen, Cris Kuhlemeier, and Didier Reinhardt.

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E1.2 An investigation into the mechanism of shoot bending in a clone of *Populus tremuloides* exhibiting 'crooked' architecture

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Populus tremuloides Michx. (trembling aspen) is a tree species native to much of North America, characterized by an excurrent crown with horizontal to ascending branches and a dominant terminal leader. An unusual clone of trembling aspen was discovered in the 1940s near Hafford, Saskatchewan. This clone demonstrates abnormal crown morphology, in

which elongating shoots bend down, ultimately leading to an overall twisted or crooked appearance. The objectives of the present study were to investigate the mechanism of shoot bending by [1] characterizing the process and timing of bending, and [2] comparing anatomical aspects of bending shoots with those of wild-type shoots. Developing, vigorous, current-year shoots of crooked aspen were 3-D digitized at regular intervals through an entire growing season. Subsequent L-system reconstruction models revealed a dramatic bending of the shoot after exceeding a certain length. In an attempt to identify the mechanism of shoot bending, current-year leader shoots from both wild-type and crooked trees were sampled throughout one growing season. Anatomical studies revealed differences between developing wild-type and crooked shoots, as well as differences within sections of crooked shoots. Structural components, including primary phloem fibres, were different between the upper and lower sides of bending shoots. Fibre cells on the upper side maintained relatively large lumens while those on the lower side were fully lignified, like those of wild-type shoots. As a control, several crooked aspen leader shoots were forced to grow in a vertical manner, comparable to wild-type leader shoots. These shoots maintained differences in phloem fibre wall thickness, similar to that of bent shoots. These results suggest that young shoot bending in the crooked aspen may be caused, at least in part, by differences in phloem fibre lignification, leading to uneven mechanical support and ultimately bending due to self-weight.

E1.3 Chasing the golden angle of needle trace-divergence in lodgepole pine.

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Transverse sections of 1st-year immature stems of lodgepole pine (*Pinus contorta* Dougl. ex Loud. var. *latifolia* Engelm.) were examined to see if relationships between Fibonacci (F) numbers (5,8,13,21) of resin canals (RCs), numbers of vascular bundles, and the angle and direction of needle trace divergence could be confirmed. The measured average angle of the needle trace divergence for lodgepole pine immature stems was found to be ~137.64^0 (n=89), very close to the expected golden angle (137.507…°) that can be approximated by dividing any F_n# by F_{n+2}# and multiplying by 360°. The pattern and angle of needle trace divergence in stems with 8, 13, and 21 resin canals were determined. The pattern of trace divergence confirmed a 1:1 relationship between number of primary resin canals and number of vascular bundles. The mean measured angles of trace divergence for stems with 8, 13, and 21 resin canals were very close to the theoretical average angle of needle trace divergence for phyllotactic fractions 3/8 (135°), 5/13 (138.46°), and 8/21 (137.14°). Finally, the angles oscillate around the golden angle in the predicted way giving rise to an oscillation in the direction of leaf trace divergence for a given ontogenetic spiral direction. These results corroborate our previous studies on the spiral phyllotaxis of lodgepole pine.

E1.4 Wound-Associated *de novo* Meristem Generation in *Arabidopsis* Foliar Explants

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Many species of plants have a remarkable ability to generate meristems from somatic cells. To investigate this process at the molecular and cellular levels, we have developed a system to study shoot & root organogenesis from somatic cells in foliar explants of *Arabidopsis*. As an initial step in investigating this process we have conducted a natural variation screen and characterized the *de novo* generation of shoot- & root meristems in 60 ecotypes. Mapping has subsequently identified loci associated with the ability to regenerate shoots in some of these ecotypes. With regard to organogenesis and cell proliferation in 5 selected ecotypes, we have discovered critical periods of sensitivity to hormone application, light exposure and explant age. Utilizing these critical conditions in specific combinations, we have conducted a mutant screen of 20,000 M2 explants to identify key genes regulating shoot organogenesis in response to these conditions. From this screen we have isolated 12 EMS mutants with consistently enhanced shoot

organogenic abilities under normally non-permissive conditions. To examine to roles of various hormone-response and developmental pathways in *de novo* meristem generation, we are comparing cell proliferation and shoot organogenesis in ~100 previously characterized mutants and transgenics with altered developmental, hormone- and light-responses. Further, we are utilizing a variety of GUS and GFP markers to dissect the developmental process and to characterize the effects of a variety of conditions on cell proliferations and the different stages of shoot organogenesis. Additionally, we have studied the responses of 5 selected ecotypes to the exogenous application of various phytohormones and hormone response inhibitors. Guided by these observations we have begun to elucidate the mechanisms underlying the differing abilities of the ecotypes to generate *de novo* shoot meristems and identified treatments that promote shoot-organogenesis in normally recalcitrant ecotypes.

E1.5 Determination of the shoot apical meristem in microspore derived embryos of *Brassica napus* cv Topaz

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Maximization of the efficiency of the canola microspore derived (MD) embryo system has been achieved through the enhancement of embryogenesis via the promotion of synchronous development using the glutathione synthesis inhibitor buthionine sulfoximine (BSO). Applications of BSO to the culture medium created a more oxidized environment through alterations in the glutathione redox system and resulted in a 3.5 fold increase in conversion frequency. Treated embryos showed improved shoot apical meristem structure and an increased accumulation of storage products, similar to those observed in zygotic embryos. These events were directly correlated to the ability of BSO-treated embryos to produce viable and vigorous plants. Changes in ABA metabolism were also observed after BSO treatments. During the late phases of embryogenesis ABA turnover increased in treated embryos, which showed higher levels of endogenous ABA, as well as increased levels of several ABA catabolites, i.e. phaseic acid, dihydrophaseic acid and ABA-glucose ester. Exogenous applications of ABA mimicked the effect of BSO, resulting in improved embryo conversion frequency and alterations in embryo structure similar to those observed after BSO treatments. Overall, these results suggest that proper development of the shoot apical meristem and improved embryogenesis in canola are dependent upon the interaction of the glutathione redox state and ABA metabolism.

E2. Phytochemicals in human health research: bioactivity vs. physiological relevance (part 2)

E2.1 Designer Fruits with Enriched Phytochemicals for Human Health

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High dietary intake of fruits and vegetables rich in phytochemicals, particularly those with antioxidant activity, has been linked to reduced risks of many chronic diseases including cancer and cardiovascular diseases. Nutraceuticals containing such bioactive phytochemicals have been popular and made available in the market; however, supplementation of these extracted, and some times purified phytochemicals can pose new health concerns such as: 1) the toxicological effect of these phytochemicals at highly concentrated level is largely unknown; 2) the extraction/purification process may eliminate important synergistic minor components. Non-processed fruits and vegetables that are known to be rich in bioactive phytochemicals are therefore advantageous for the intact and balanced phytochemical contents. Fruits with elevated concentrations of known antioxidant phytochemicals, for example, may set a new trend for the breeding programs. However, this is a challenging task and it requires close

multi-disciplinary collaborations among scientists. In this presentation, the author is prepared to show the chemistry, biochemistry and health benefit of different antioxidant phytochemicals in fruits, and how breeding and other factors can help obtain safe designer fruits in which phytochemicals known for lowering different chronic diseases are enriched.

E2.4 Plants and Neurochemistry

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One of the most common uses of plants is the manipulation of human brain function and human culture is profoundly influenced by those plants which affect the function of our brains. Different plant species contain human neurotransmitters, neuroregulators and neurotoxins. The human neurotransmitter melatonin (N-acetyl-5-methoxytryptamine) is a ubiquitous, highly conserved molecule associated with timing of circadian rhythms in bacteria, yeast, mammals and perhaps higher plants. Compounds such as hyperforin, once thought to be characteristic of St. John's wort (*Hypericum perforatum* L.) but now isolated from several plant species, are effective selective serotonin reuptake inhibitors in the human brain. A different group of neuroregulatory molecules produced by plants overstimulate human neurons resulting in neuronal cell damage and death. Excitotoxins such as ß-methylamino-L-alanine not only effect human health but are also regulatory molecules redirecting plant growth. The accumulation of specific compounds in plant tissues is dependent on the growing conditions, nutrient availability and micro-environment. The highly conserved nature of some neurologically active compounds may indicate a greater role in signal transmission and the regulation of plant cell growth.

E3. Conservation and diversity II

E3.1 Parasitic-host plant interaction as a mechanism facilitating horizontal transfer of mitochondrial genes in land plants

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Horizontal gene transfer (HGT) is widely recognized as a major force in bacterial evolution. In eukaryotes, however, it is prevalent only in certain phagotrophic protists and limited largely to the ancient acquisition of bacterial genes. Moreover, HGT is unknown *within* the evolution of animals, plants, and other groups of multicellular eukaryotes except in the special context of mobile genetic elements. Recently, it has been demonstrated that standard mitochondrial genes, encoding ribosomal and respiratory proteins, are subject to evolutionarily widespread HGT between distantly related land plants. These transfers have created a variety of genomic outcomes, including gene duplication, recapture of genes lost through transfer to the nucleus, and chimeric, half-monocot/half-dicot, genes. Taken together, these results implied the existence of mechanisms for the delivery of DNA between unrelated plants and prompted numerous speculations about possible nature of those mechanisms. The evidence for two new cases of HGT, involving the same mitochondrial gene (*atp1*) and the same genus of recipient flowering plants (*Plantago*; Plantaginaceae) is presented here. Both transfers most likely occurred by direct plant-to-plant transmission of DNA to host plants from distantly related parasitic plants, *Cuscuta* (Convolvulaceae) and *Bartsia* (Orobanchaceae), respectively. This is indicated by strong phylogenetic evidence that the parasitic plants, which penetrate hosts intracellularly as part of their normal life cycle, served as donors in both transfers. This inference is further supported by the striking biogeographic concordance of donors and recipients as well as by the well documented exchange, in both directions, of macromolecules, viruses, and phytoplasmas between parasitic plants and their hosts.

E3.2 Phylogeographic genetic analysis of Pitcher's thistle (*Cirsium pitcheri*)

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Thistles are common throughout Canada and those belonging to the *Cirsium* genus have been known to exist not only as troublesome weeds, but also as endangered species. Pitcher's thistle is endemic to the shorelines of the Great Lakes and is currently limited to approximately four main sites within Canada. In 1999 these plants were listed as endangered by the Committee on the Status of Endangered Wildlife in Canada (COSEWIC). We have applied organelle DNA sequences [mitochondrial DNA (mtDNA) and chloroplast DNA (cpDNA)] and nuclear sequences to assess the phylogeography and levels of genetic diversity of Ontario Pitcher's thistle populations. The Pitcher's thistle habitat is limited to disturbed dune systems and as a result local populations likely undergo extinction and recolonization events. The modes of dispersal of this species are largely unknown; assessing the above Ontario populations will test the hypothesis that long-distance dispersal has occurred through dormant seeds that are moved with sand and the water currents of the Great Lakes. Identifying distinct populations has implications for the number of conservation units to be considered under COSEWIC, the Species-at-Risk Act (SARA) and associated Recovery Team. Furthermore, identifying connectivity and the evolutionary relatedness among provincial sites will provide insight for recommending source populations for translocations as part of potential management strategies. Future studies will examine fine-scale metapopulation dynamics and the role of local recolonization and gene flow within the provincial sites.

E3.3 Characterizing Plant Biodiversity in Naturalized Pastures of the Maritimes

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Pastures in the Maritimes often contain native species of forage such as bluegrasses, bentgrass and white clover. Although these resilient species play an important part in the nutrition of the region's ruminant livestock, limited data exists on the dynamics of species composition and sward nutritional quality over the grazing season. The objective of this project is to determine changes in the botanical composition, yield and nutritional quality of naturalized pasture swards over the grazing season. Two pastures have been studied; an upland field at Nappan, Nova Scotia and a Salmon River flood plain at Truro, Nova Scotia receiving only dung from grazers as the managed nutrient source. The flood-plain soil was high in organic matter and extremely fertile resulting in a steady production of grass pasture, even though the legume content was small. From May 31 to Sept 29, 35-82% of pasture forage dry matter was composed of bluegrass, 7-49% was couchgrass, 4-18% timothy, 1-5% legume, and 0.1-18% other species. On the upland pasture, from May 1 to Oct 31, 18% of pasture dry matter was bluegrass, 16% white clover, 5% timothy, and 4% meadow fescue. The species composition of these pastures is probably reflects equilibrium under the conditions of current grazing intensity. Forage species composition may indicate how cultivated stands naturalize, and the degree of compatibility between species in a pasture sward. Forage samples will be analyzed and the research scope will be expanded by adding another pasture type in 2005.

E3.4 Horizontal and vertical seed banks from Ritchie's prisere on the Churchill River Estuary, MB.

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In 1957, Ritchie identified five successional vegetation stages that had developed between the mudflats and the relatively stable lichen woodland adjacent to the Churchill River estuary, Churchill, MB. He named these zones: meadow, shrub, invading forest, closed forest, and open black spruce forest (with mound topography). The purpose of this project was to describe soil seed banks from the first four of Ritchie's vegetation zones and investigate any relationship between them and the standing vegetation. The fifth zone is now heavily disturbed and was not included in our study. Twenty-five soil samples were taken from the

substrate surface of each zone. Additional samples were extracted at one-centimeter depth increments from four cores per zone to the depth of the water table. The germination method was used to assess soil seed content of fresh and cold-stratified soil samples. Standing vegetation was described from 25 50 x 50 cm quadrats per zone. Attempts to germinate seeds from surface samples resulted in 448 seedlings of 16 taxa (to date). The majority of seedlings (70%) were those of *Juncus arcticus.* This species dominated the seed banks of all zones except the meadow, even though it was only a rare component in the standing vegetation. *Spergularia marina* was the most prevalent species in the meadow zone seed bank but it persisted to a lesser extent into the shrub zone. One hundred and fifty seedlings emerged from the soil cores, 97% occurred in the top 5 cm but a few were found to a depth of 11 cm. The majority (57%) were those of *Juncus arcticus*. In conclusion, the seed banks did not reflect the standing vegetation but only components of it, or that of an earlier successional stage.

E3.5 Geographic distribution of chloroplast DNA haplotypes of *Oxyria digyna* (Polygonaceae) and implications for the postglacial history of western Canada.

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The distribution of genetic variation in plant populations can provide important insights into patterns of migration and dispersal in response to past climatic and geological events. We examined genetic patterns in western North American populations of the circumboreal Arctic-alpine plant *Oxyria digyna,* using restriction site variation in the psbA-trnH and trnT-trnL spacer regions of the chloroplast genome. Pronounced north-south differences in haplotype composition were found. Of the 10 chloroplast haplotypes identified, three are predominantly or entirely northern (north of 56° N). One of these is widely distributed, occurring also in Norway, Russia, and the Canadian Arctic. Three other haplotypes are largely southern (central British Columbia and southward). At least two haplotypes occur entirely within the boundaries of the most recent glacial advance in western Canada. Regional haplotype diversity is greatest in north-central B.C., a region of great topographic relief generally thought to have been entirely covered by Late Wisconsin glaciers. The distribution of haplotypes of *O. digyna* is consistent with postglacial recolonization of B.C. from both Beringian and northwestern United States unglaciated regions, but it also suggests that populations of *O. digyna* may have persisted in refugia within the Cordilleran Ice sheet.

E3.6 Co-occurrence of rare vascular plants in the Northern Rocky Mountains of Alberta

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Traditional approaches to the assessment and conservation of rare plants in Alberta and Canada have focused on individual species. Funding is limited and with the current rates it will take over 200 years to conduct formal assessments on vascular plants currently listed as rare in Alberta by the Alberta Natural Heritage Information Centre. We tested the fidelity of rare species to habitat and vegetation types in the northern Rocky Mountains of Alberta and co-occurrence of rare species within these habitats in a three year period from 2001 to 2003. The largest numbers of rare species occur at high elevation and many species within these alpine sites have similar responses to environmental gradients as noted by an analysis of their attribute plots from a constrained ordination (distance-based redundancy analysis). Subsequent association and indicator species analyses indicate that some taxa, for example *Juncus biglumis* and *Koenigia islandica*, do co-occur and this, when combined with an examination of fidelity to habitat and distribution along environmental gradients, may provide a useful tool for the conservation of these rare species. Other species are poorly represented within the study area and thus their fidelity to habitat and co-occurrence with other rare species cannot be determined. Species specific approaches to conservation are thus appropriate for these taxa.

E4. Rhizosphere

E4.1 Impacts of fertilization on fine roots and ectomycorrhizas under young lodgepole pine and interior spruce stands in the interior of British Columbia

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We studied fine roots and ectomycorrhizas in 20-year-old lodgepole pine and interior spruce stands in central British Columbia 10 years after the initiation of annual fertilizer treatments. Three treatments were studied: unfertilized control, ON1 (650 kg N, 400 kg P, 400 kg K added in total plus other nutrients), and ON2 (1350 kg N, 400 kg P, 400 kg K plus other nutrients). We determined ectomycorrhizal morphotypes, fine root length, and active and dead fine roots. ON2 had less lodgepole pine fine root length, fewer ectomycorrhizal roots, fewer active fine roots, and more non-mycorrhizal fine roots than ON1 and the unfertilized control. *Suillus* sp. and *Amphinema* sp. disappeared from the highest fertilizer treatment while *Wilcoxina* sp. increased. For interior spruce, the trends were different with fine roots and ectomycorrhizas responding positively to fertilizer. These dynamic changes to fine roots and ectomycorrhizas reflect differences in the two conifer species as well as changes to the plant community as a result of treatment.

E4.2 A comparison of fungal communities associated with nodules and roots of gray alder (*Alnus incana*) and the roots of paper birch (*Betula papyrifera*).

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To test the effects of root function on communities of root-associated fungi, we compared the composition and physiology of fungal communities on the roots and nodules of a nitrogen-fixing plant with those on the roots of a non nitrogen-fixing plant. Roots of *Alnus incana*, and of the comparable, related, and cohabitating *Betula papyrifera*, were collected from several sites in central Alberta, Canada. Standardized homogenates prepared from surface-sterilized root-tips and nodules from these samples were streaked on selective media to obtain culturable fungal associates and pipetted into Biolog microtitre plates to obtain a crude estimation of substrate utilization profiles of the associated fungal communities. Preliminary analyses of these data show that while the profiles of all sample types overlap, the diversity of these profiles is greater for the roots of either species than that of the nodules. Preliminary community composition data support these findings: fungal communities associated with nodules were less variable among populations and were more distinct taxonomically than the communities associated with the roots of either plant species. Such a narrow suite of fungi may be indicative of unusual physiological conditions in and around nodules. These preliminary observations also suggest that the functional diversity of a plant community could drive the diversity of associated fungal communities.

E4.3 Interaction between wheat cultivar and bacterial isolate for rhizosphere colonization and root accumulation of *Pseudomonas*-derived 2,4-diacetylphloroglucinol

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Biological control of soilborne pathogens by 2,4-diacetylphloroglucinol (DAPG)-producing isolates of *Pseudomonas fluorescens* (rhizobacteria) offers a sustainable means of controlling root diseases, but efficacy of the isolates is subject to environmental factors. Our laboratory is identifying host traits that govern interactions with rhizobacteria, with the view to improving biological control in the field. We measured populations of two isolates of *P. fluorescens* on soil-grown roots of 28 cultivars of *Triticum aestivum* L. (hexaploid wheat). Isolate Q8r1-96 persisted in the rhizosphere of most cultivars longer than isolate Q2-87, but certain cultivars supported high populations of both isolates, whereas others supported low populations of both. Differential rhizosphere colonization was not due to differences in aggressiveness, as both Q8r1-96 and Q2-87 rapidly colonized wheat roots within four days of seed inoculation. Using three cultivars representing each colonization phenotype, we examined the relationship between cultivar and

bacterial isolate in accumulation of DAPG, a pathogen-suppressive polyketide metabolite known to exert phytotoxic effects. Accumulation of DAPG was higher on roots colonized with Q8r1-96, compared to Q2-87, four days after seed inoculation and growth in Petri plates. These results were consistent with the relative production of DAPG by the isolates in culture. However, DAPG production by Q8r1-96 was much greater than Q2-87 on cv. Tara, whereas no difference was observed on cv. Buchanan. The accumulation patterns could not be attributed to differences in rhizoplane population densities, or to the greater root surface area of Tara. Our findings indicate that DAPG accumulation in the wheat rhizosphere is independent of population density, but dependent on an interaction between host and bacterial genotypes. Progress in identifying the host determinants of differential accumulation using micorarrays and other approaches will be presented.

E4.4 Combined effects of elevated atmospheric CO_2 and rhizobial strains on nitrogen fixation and cold acclimation in alfalfa

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The predicted rise in concentration of atmospheric CO_2 could affect directly a variety of plant processes, including photosynthesis, reserve accumulation and, ultimately, tolerance to environmental stress. Elevated atmospheric CO_2 could also alter the biotic environment of the root system. Perennial alfalfa (*Medicago sativa* L.) is a sustainable crop with a deep root system, living in symbiosis with rhizobia for atmospheric nitrogen (N) fixation and, as such, is a good candidate for carbon (C) sequestration in agriculture. The objectives of the research project were to determine 1) the effects of elevated concentration of atmospheric CO_2 on growth, N fixation, net photosynthetic rate (Pn), the accumulation of C and N reserves and cold acclimation in alfalfa, and 2) whether soil microflora is modified under high atmospheric CO_2. Plants inoculated with two different strains of rhizobia (*Sinorhizobium meliloti*) were grown two months at optimal growing temperature and then cold acclimated at 2°C for two weeks under either 400 (ambient) or 800 (elevated) ppm of CO_2. Pn rates were increased under high CO_2 and were higher when plants were inoculated with rhizobial strain A2 than with strain NRG34. Growth of both roots and shoots of alfalfa were increased under 800 ppm of CO_2 with a higher root weight for alfalfa inoculated with strain A2 as compared to strain NRG34. Nitrogenase activity was also stimulated under 800 ppm of CO_2. Plants acclimated under ambient CO_2 were shown to be more freezing tolerant. Cryoprotective sugars linked with freezing tolerance such as sucrose, stachyose and raffinose increased in taproots during cold acclimation but did not differ between ambient and high- CO_2 treatments. Furthermore, the expression of key genes, linked with freezing tolerance and reserve accumulation in alfalfa, was modified by both the CO_2 treatments and the rhizobial strains. We observed a modification of the soil microflora under elevated CO_2 with a significant increase in fungi population. This multi-disciplinary experiment clearly shows that it is possible to identify rhizobial strains to improve plant performance under high CO_2. The study of the synergistic plant response to both atmospheric CO_2 and rhizobial strains is a promising emerging research avenue.

E4.5 Multiple isolates of species of *Monodictys* from the roots of *Saxifraga oppositifolia* from the Canadian High Arctic.

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Species of *Monodictys* (Hyphomycetes) have teleomorphs in the Dothideomycetes and have been reported as wood decomposers, plant pathogens, and soil and water inhabiting fungi in temperate regions. Recently, we obtained nine isolates assignable to this genus from the roots of seven different collections of *Saxifraga oppositifolia* collected from Ellesmere Island in the Canadian High Arctic (79º N, 76º W). Colonies were melanized, produced smooth, straight, unbranched, and darkly pigmented conidiophores, and bore multicellular (4-9 celled), melanized, globose to

oblong conidia. Conidial size varied from 20–50 ?m in length and from 20-30 ?m in width. Prior reports of dark septate endophytes (DSE) in the roots of arctic plants refer to *Phialocephala fortinii* and *Leptodontidium orchidicola* but there are no reports of species of *Monodictys* in this habitat. Unlike other species of DSE which have a broad host range, our isolates of *Monodictys* were recovered exclusively from *S. oppositifolia*. These observations may represent the first report of species of this genus from plant roots.

E4.6 Microorganisms stimulating growth and development of greenhouse tomato plants

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Plant growth stimulation by microorganisms has been widely studied over the years. Microorganisms that stimulate plant growth by either improving the nutrient status of the plant or by producing plant growth regulators are often referred to as biofertilizers. In this study, eight microorganisms (*Penicillium brevicompactum* Dierckx, *Penicillium solitum* Westling, two strains of *Pseudomonas fluorescens* Migula, *Pseudomonas marginalis* (Brown) Stevens, *Pseudomonas putida* (Trevisan) Migula, *Pseudomonas syringae* van Hall and *Trichoderma atroviride* Karsten) showing antagonistic activity against *Pythium ultimum* Trow, causal agent of tomato (*Lycopersicon esculentum* Mill) root rot, were tested for their potential to promote the growth of greenhouse tomato plants. For the first experiment, tomato seeds were inoculated with the micoorganisms at the moment of sowing in perlite. After 30 days, the plants were evaluated and the results showed that, compared to the control, several of these microorganisms significantly increased the surface area of the root system as well as the fresh and dry weight of the plant. Furthermore, greenhouse assays on mature crops revealed that the inoculation of the growing media (rockwool or an organic medium made of peat, compost and pine sawdust) with some of these microorganisms significantly increased the marketable fruit yield and, in some cases, the plant growth. This study suggests that these antagonistic microorganisms may eventually be used for their biofertilizing properties.

E5. Reactive oxygen

E5.1 Visualizing reactive oxygen species formation in plant cells treated with stressors.

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Many environmental stressors are known or suspected to induce the formation of reactive oxygen species (ROS) in plant cells. For example, redox-active metals and polyaromatic hydrocarbons (PAHs) exhibit synergistic toxicity in exposed organisms. A mechanism for this toxicity is the blockage of electron transport in both chloroplasts and mitochondria by PAHs, such as 1,2-dihydroxyanthraquinone (dhATQ). By redox cycling, Cu^{2+} and similar metals can divert blocked electrons to oxygen, generating ROS. To monitor ROS formation in plant cells, Cu^{2+} and dhATQ were applied alone and in combination. Additional stress treatments with ultraviolet (UV) light, cold temperature and the electron transport inhibitor DBMIB were also tested. ROS production was monitored using fluorescent ROS indicator dyes, including 2',7'-dichlorodihydrofluorescein diacetate. ROS generated after short exposures (2 h) to Cu^{2+}, dhATQ, or a combination of Cu^{2+} and dhATQ was assessed in chlorenchyma cells isolated from *Asparagus densiflorus* (Kunth) Jessop. The effects of longer exposures (24+ h) were observed in mesophyll cells of intact *Lemna gibba* L. plants. Laser-scanning confocal microscopy was used to pinpoint subcellular locations of ROS generation. Indicator dyes revealed that the bulk of ROS was formed in chloroplasts and mitochondria, as expected. In *Asparagus* cells, three indicator dyes detected lower ROS in dhATQ treatments, while a fourth reported increased ROS with dhATQ. There was no qualitative increase in dye fluorescence with Cu^{2+} either alone or combined with dhATQ after 2 h. Differences in organelle fluorescence were noted between the indicator dyes and between control, dhATQ, and Cu^{2+} treatments. In intact *Lemna* fronds, Cu^{2+}, cold temperature, and UV increased ROS formation, while DBMIB and dhATQ reduced ROS formation compared to controls after 24

h. The results will be interpreted and discussed in the context of the current ROS generation model.

E5.2 Potential role of thiol-disulfide oxidoreductase-based antioxidant system in aluminum (Al) and cadmium (Cd) resistance

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Oxidative damage induced by metal ions has been widely recognized as a mechanism of metal toxicity in plants. Not surprisingly, various components of the antioxidant system have been shown to play a crucial role in metal resistance. We have focused on various components of the antioxidant system which possess a conserved dicysteine active-site motif. These thiol-disulfide oxidoreductases may play a major protective role during Al and Cd stress by maintaining cellular redox homeostasis, repairing oxidatively damaged proteins, and scavenging reactive oxygen species and lipid hydroperoxides. Currently we are using yeast (*Saccharomyces cerevisiae*) as a model system to identify key components of the antioxidant system for future research in *Arabidopsis*. From 15 single disruption mutants defective in thioredoxins, thioredoxin reductases, thioredoxin peroxidases or glutaredoxins, 4 were found to be hypersensitive under Al and Cd stress. Northern analyses showed increased expression of the genes identified in the wild-type strain under Al stress. Lipid peroxidation was also increased in the hypersensitive mutants compared to the wild-type strain under Al stress. Hypersensitivity to Al was shown to increase ~20% in double and triple mutants of the 4 identified genes. These results suggest that this antioxidant system based on thiol-disulfide oxidoreductases may play an important role in Al and Cd tolerance. These key components identified in yeast will be further studied in *Arabidopsis* to confirm the role of this antioxidant system in metal tolerance in plants.

E5.3 Class-1 hemoglobin and antioxidant metabolism in alfalfa roots

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In the course of nitric oxide (NO) scavenging, hemoglobin (Hb) is oxidized from the ferrous to the ferric (metHb) form. The NO scavenging rate of class-1 barley Hb is markedly facilitated by monodehydroascorbate (MDA) reductase. The latter mediates a coupled reaction involving ferric Hb reduction in the presence of ascorbate and NADH by removing MDA, which acts as a strong oxidant. The Hb turnover, therefore, is linked to antioxidant metabolism and affects cellular NADH/NAD ratios. The influence of Hb presence on the ascorbate-glutathione cycle enzymes and the levels of NO, H_2O_2 and ascorbate was investigated in alfalfa root cultures over-expressing (Hb+) or down-regulating (Hb-) barley Hb. Hb- lines had half of the activity of the NO- and H_2O_2- sensitive enzyme, aconitase, as compared to Hb+ lines. Low stability of this enzyme in Hb- lines upon the exogenous treatment with the NO-donor DEANO demonstrates that Hb protects plant cells from NO. The H_2O_2 levels in Hb+ and Hb- lines were not significantly different. Hb+ lines had almost twice the level of ascorbate, elevated MDA reductase and ascorbate peroxidase activities. Hb- lines showed significant increases in dehydroascorbate (DHA) reductase and glutathione reductase activities. The observed changes in ascorbate and ascorbate-glutathione cycle enzymes were pronounced both at high (40 kPa) and low (3 kPa) O_2 concentrations. The results indicate the importance of ascorbate in the maintenance of Hb in the reduced state and the modification of the ascorbate-glutathione cycle by the presence of Hb. They suggest a role for barley class-1 Hb in the improvement of the antioxidant capacity of plant cells.

E5.4 Characterization of the alternative oxidase of *Chlamydomonas reinhardtii* in response to nitrogen source and oxidative stress

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In addition to the ubiquitous cytochrome pathway of mitochondrial electron transport,

plants possess a second alternative pathway that is comprised of a single protein, alternative oxidase (AOX). AOX is encoded by a nuclear gene, *Aox1*, which is transcriptionally activated by a variety of stress conditions. Furthermore, recent research suggests that AOX may function to aid in programmed cell death.

In the described research we employed a variety of experimental procedures to characterize the response of AOX in *Chlamydomonas reinhardtii*. In particular we used the arylsulfatase (ARS) reporter gene system to study the transcriptional regulation of *Aox1*. Interestingly, while we find that the *Aox1:Ars* reporter system is activated in response to a shift in nitrogen source from ammonium to nitrate we find that the reporter is insensitive to the induction of *Aox1* triggered by antimycin A, H_2O_2 and cold stress. Furthermore, we provide data on the interactions between respiration and nitrogen metabolism gleaned from the analysis of a number of mutants which fail to express the *Aox1:Ars* reporter gene following a shift in nitrogen source. Taken together with other experiments we hypothesize that in *C. reinhardtii* there exist two pathways leading to transcriptional activation of *Aox1* - one sensitive to nitrogen status and a second that responds to an increase in intracellular ROS.

E6. Pathogen detection

E6.1 A PCR approach to detecting bacterial blight organisms in an epidemiological study in dry bean fields in southern Alberta.

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Diseases caused by pathogenic bacteria have a serious impact on dry bean production worldwide. In Alberta, the major bacterial diseases on beans are halo blight [*Pseudomonas syringae* pv. *phaseolicola* (Burk.) Young *et al.*], and common blight [*Xanthomonas axonopodis* pv. *phaseoli* (Smith) Vauterin *et al.*]. Bacterial blights can lead to significant crop damage and yield loss, especially when environmental conditions (i.e. warm humid periods) favour disease development. One of the primary methods of disease management is the use of certified disease-free seed. However, recent bacterial blight problems in southern Alberta have arisen in fields sown with certified seed stocks originating outside of Canada. Producers have questioned whether these disease outbreaks were the result of contaminated seed or originated from local sources. In order to pinpoint the origin of such infestations, a survey of seven bean fields with halo blight and/or common blight was done in southern Alberta in 2004. Samples of bean leaves and pods, as well as bean seed, weeds, soil, water, insects, and soil and crop residues from farm machinery, were cultured in order to isolate bacteria with colony types typical of those pathogens causing blight diseases on bean. Genomic DNAs extracted from each isolate were used as templates in PCR reactions to confirm the presence of *P. syringae* pv. *phaseolicola* or *X. axonopodis* pv. *phaseoli* from these samples. The implications of the PCR data on the epidemiology of bacterial blight diseases in Alberta will be discussed.

E6.2 Refinement of PCR-based methods for detecting and quantifying the bacterial spot pathogen on seeds and transplant seedlings

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Over the last few years, Ontario tomato and pepper growers have experienced significant yield losses due to the disease bacterial spot, which is caused by *Xanthomonas campestris* pv. *vesicatoria*. The primary source of infection is thought to be infested seed even though most commercial seed receives some form of antibacterial treatment. Once inside a transplant seedling greenhouse, the pathogen can spread very quickly reaching high populations in a short period of time on plants that may appear symptomless. Our objective has been to develop rapid, sensitive, quantitative methods for determining the inoculum potential, or disease threat, of these infested seeds and transplants. Both conventional PCR and quantitative PCR (qPCR) using the Roche LightCycler system formed the basis for our assays. The primer sets we used, BSX1/2 and BSX9/10, were derived from the sequence of the bacterial spot diagnostic DNA probe KK1750. A number of methods for extracting bacterial DNA from large seed and transplant samples were assessed. The sensitivity of the seed assay was enhanced significantly by first germinating the seed, allowing any viable bacteria present

to multiply. The optimal recovery method for seed differed from that used for transplants or field plant samples. The minimal detection level with qPCR for a 15-g plant sample was approximately 50,000 bacteria; the detection limit for samples containing 20,000 untreated germinated seed was approximately 20,000 bacteria.

E6.3 Discrimination of carrot diseases in storage using headspace volatile metabolite profiles

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Gas chromatography/mass spectrometry was used to profile volatile metabolites from the headspace gas of carrot cultivar Vita – Treat which was inoculated with water or four different pathogens: *Botrytis cinerea* Pers., *Erwinia carotovora* ssp *carotovora* (Jones) Begey et al., *Aspergillus niger* Tieghem and *Fusarium avenaceum* (Corda, Fries.) Sacc. A total of 137 different volatile metabolites were detected but only 44 of them were relatively consistent, including 13 metabolites that were specific to one or more inoculations/diseases. The metabolites namely 3-methyl-1- butanol, 1-pentanol, 2-methyl -1- propanol, 2,3-butanedione, ethyl - boronic acid, 1-methoxy-3-methyl-butane and ethoxy ethene were specific to *E. carotovora* ssp. *carotovora*. 1,2-dimethoxy-ethene was specific to carrots inoculated with *E. carotovora* ssp. *carotovora* and *B. cinerea*, while 3-chloro-4-hydroxy-1,4-diphenyl-2-butanone was common only to carrots inoculated with *E. carotovora* ssp. *carotovora*, *F. avenaceum* and sterile distilled water. Discriminant analyses of metabolite fingerprints based on mass spectra and metabolites correctly classified 35 to 85% and 20 to 60% of metabolite fingerprints, respectively, in to respective inoculations/diseases. Discriminant analyses models for the metabolite fingerprints from 3 d incubation period correctly classified (up to 90%) the unknown fingerprints into respective inoculations/diseases better than those from 6 d incubation period (60%). The potential application of early detection and discrimination of post harvest diseases of carrot Vita -Treat using disease-specific compounds and discriminant analysis models will be discussed.

E6.4 Characterization of *Plasmodiophora brassicae* populations from Alberta, Canada

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Clubroot, caused by the obligate parasite *Plasmodiophora brassicae* Woronin, was identified in a number of canola (*Brassica napus* L.) fields in central Alberta in 2003. In order to characterize the virulence of local populations of the pathogen, field isolates from a number of locations in the Edmonton region were tested on the two most widely used sets of differential hosts, i.e. those of P.H. Williams and the European Clubroot Differential (ECD) series. While the reaction of some host varieties could be clearly defined as either resistant or susceptible, other differentials showed intermediate disease index scores. If disease indices of 0 to 45% and 46% to 100% were regarded as resistant and susceptible, respectively, then races 3 and 5 were identified among six isolates tested from two fields in the Edmonton area. In contrast, an isolate obtained from Abbottsford, B.C., was designated as race 6. Other field isolates are presently being tested, from Alberta and other parts of Canada. In addition, preliminary resistance screening of 48 varieties included in the 2004 Prairie Canola Variety Trials with a race 3 field isolate indicated that they were all highly susceptible to the disease. This suggested that if clubroot were to become widespread in Alberta, it could have a major negative impact on yields.

E6.5 The development of multiplex real-time PCR to monitor biological control agents in the orchard

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Fire blight caused by *Erwinia amylovora* is one of the most destructive bacterial diseases in pear, apple and other rosaceous ornamental plants in Northern America. Control measures involve pruning and the use of antibiotics. The

appearance of streptomycin resistance in the United States and the potential loss of streptomycin registration in Canada has resulted in the development of biological control methods. Bacteriophages are bacterial viruses that specifically kill their bacterial host. In order to use phages as a biological control agent, we have isolated and screened for specific phages showed lysogenic activity on *E. amylovora*. Selected phages were applied to blossoms with newly developed phage carrier system. The efficacy of the carrier-phage system was evaluated under field conditions. We developed a multiplex real-time PCR detection system to simultaneously monitor three microorganisms (biological control agents and bacteria) in an orchard to understand the ecological impact of biocontrol agents in an ecosystem. Detailed results from biological control experiment and multiplex detection by real-time PCR will be discussed.

E6.6 Double antibody sandwich enzyme-linked immunosorbent assay testing for detection plum pox *potyvirus* using polyclonal antibodies.

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Plum pox *potyvirus* (PPV) is one of the most devastating diseases of stone fruits, including plums, peaches and apricots. In 2000, Dideron strain of PPV (PPV-D) was detected in Ontario, Canada. As a part of a program to improve detection of in field surveys, a two year study was conducted to determine the distribution of PPV-D in leaves and fruits on the PPV infected trees in Ontario, Canada. The detection of PPV was carried on leaf and fruit tissue samples by the double antibody sandwich enzyme-linked immunosorbent assay (dasELISA) using polyclonal antibodies. Results from the dasELISA on leaf samples showed that PPV-D strain was unevenly distributed in the trees of ten different peach cultivars in Ontario. The fruit from PPV-D-infected scaffolds were found to be positive: PPV-D was detected in the skin and in the tissues below the skin and the virus was unevenly distributed in these tissues. PPV-D could be detected in fruit that was kept at 22°C for up to10 days. The virus from peach fruits was found to be infective, as determined by its mechanical transmission to the herbaceous host, *Nicotiana benthamiana.* The resulting PPV-D -positive *N. benthamina* plants were confirmed by dasELISA.

F1. Resistance to diseases: Unraveling the plant responses (part 1)

F1.1 Systemic Acquired Resistance in Tobacco and Arabidopsis

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Systemic resistance to disease is exhibited in many plants after local infection by avirulent pathogens. NN tobacco infected with TMV and Arabidiopsis infiltrated with avirulent strains of *Pseudomonas syringae* provide two robust systems for the study of this systemic acquired resistance (SAR) phenomenon. At least in tobacco, salicylic acid (SA) is required for a successful hypersensitive response and initiation of the long distance signalling process triggering SAR. By cell specific expression of an enzyme able to degrade SA (salicylate hydroxylase) we show that SA in the phloem of infected leaves may be required for long distance SAR signal transmission in tobacco. The expression of benzoate 4-hydroxylase (a fungal p450 enzyme) in tobacco removes the availability of benzoate for SA synthesis and can also block establishment of SAR inferring a possible role for BA (either direct or indirect) in long distance SAR signal transmission. Comparative research in Arabidopsis has suggested that SA is not synthesised from benzoic acid and has also questioned the involvement of SA in SAR signal transmission. Using metabolomics approaches we demonstrate that local inoculation with avirulent pathogens triggers metabolome changes in local, non-infected tissues. These signals are currently being investigated to determine whether such tissues are involved in the propagation of the SAR signal in this species.

F1.4 Whirly: a new family of plant transcription factors involved in defence responses

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Plants have evolved complex recognition and response mechanisms to counter attack by microbial pathogens. These mechanisms involve perception of the invading pathogen and activation of signal transduction pathways that lead to transcriptional reprogramming of the host. We have recently identified a new family of transcription factors, called Whirly, that are involved in plant disease resistance. Members of the Whirly family of proteins are found throughout the plant kingdom and are predicted to share the ability to bind single-stranded DNA in a sequence specific manner. Resolution of the crystal structure of potato StWhy1 has provided insight into the DNA-binding mechanism of this family of proteins, their mode of action and possible autoregulation. We have shown that Arabidopsis and potato Whirly orthologs act as transcription factors that regulate defense gene expression, and that the Arabidopsis Whirly protein AtWhy1 contributes to both basal and specific defense responses against the oomycete pathogen *P. parasitica* and the bacteria *P. syringae*. Recently acquired evidence suggests that Whirly proteins might also play roles in processes other than defense responses and could function in the chloroplast as well as the nucleus.

F2. Alpine and arctic plants (part 1)

F2.1 Pattern and process at the alpine treeline

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The alpine treeline represents the boundary between lower elevation subalpine forest and upper elevation tundra communities. The controls on the position of this conspicuous landscape feature are primarily thought to be climatic although topographic, edaphic and disturbance conditions also play roles in determining the location of the contemporary treeline.

The variety of processes active at treeline lead to variations in pattern. Although often conceived as a sharp landscape boundary, treeline environments exhibit complicated patterns including linear extensions (fingers) into the tundra and patches. Pattern is often linked to positive feedbacks within the treeline system.In this presentation I outline the current thinking about treeline controls and consider the utility of treeline as an indicator of climate change at both long and short time scales. Furthermore, I discuss the importance of feedbacks and the ways in which patterns are generated in treeline environments. Current research utilizing field collected data and modeling are highlighted. Examples are drawn primarily from the Rocky Mountains and the Swedish Scandes.

F2.3 Top-down vs. Bottom-up regulation of vegetation in a Boreal Forest understorey

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Plants in the boreal forest are an important component of the ecosystem for two main reasons. First, the plants as vegetation form the physical surroundings for both herbivores and carnivores and are the basis of the physical structure of the community. Second, as primary producers, they provide the energy and nutrients to the herbivores on which higher trophic levels depend. Therefore, understanding the factors that limit the quantity and the quality of plants is fundamental. Our studies focus on the herbaceous vegetation but primarily on two grasses, *Festuca altaica* and *Calamagrostis lapponica*, which dominate small meadows scattered throughout the white spruce forest, and on the four herbs *Lupinus arcticus, Anemone parviflora, Mertensia paniculata* and *Achillea millefolium var. borealis* which are relatively abundant in the forest understory. These plants provide a source of relatively high quality food to the herbivores. Soil nutrients, especially nitrogen, often limit the productivity of boreal forest vegetation and may control vegetation standing crop. Plants differ in their abilities to respond to raised nutrient levels, and community composition usually changes following fertilization as more competitive species begin to dominate. The nutrient availability level will also influence a species' ability to produce defensive chemical compounds against herbivory and the ability to

regrow after herbivory. Conversely, herbivory may have a direct effect of vegetation quantity and quality. Herbivory has long been known to influence species composition in some plant communities due to differential plant palatability and differences in plants' abilities to tolerate herbivory. Many plants produce defensive chemical compounds in response to herbivory.

For the purposes of our study we considered the soil to be a 'trophic level.' The herbaceous vegetation, along with the soil nutrient pool, forms two of the four trophic levels recognized in this system. To understand some of the inter-trophic level linkages between components of the system, three hypotheses regarding the vegetation were tested: that vegetation was controlled by (i) nutrient availability alone (bottom-up, or donor control), (ii) by herbivores alone (top-down control), and (iii) by both nutrient availability and herbivores. This involved three major experimental treatments - fertilization, herbivore exclusion, and fertilization plus herbivore exclusion. These treatments allow us to make specific predictions about changes in plant biomass, or standing crop, under the three different hypotheses.

F2.5 Climate warming and the (in)stability of plant-herbivore interactions in subarctic alpine meadows.

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For the past decade my group has examined interactions between mammalian herbivores and alpine meadow vegetation in the southwest Yukon. This region lies in the rainshadow of the St. Elias Mountains and like many parts of the western Arctic, has experienced significant warming over the past decade, especially during winter. During this time the snow free growing season has varied from 8 to14 weeks duration, and annual measurements indicate, not surprisingly, that primary production of alpine vegetation was largely determined by the timing of spring melt. However, spring melt was not always correlated with winter temperatures, so that there is considerable uncertainty about the impacts of climate warming on alpine communities. Both multi-season and within-season experiments (passive warming, nutrient addition, snow addition, grazing) have demonstrated a wide range of potential growth responses by alpine vegetation, but that changes in species composition tend to occur slowly. In contrast, alpine mammalian herbivores are critically dependent on the timing of plant growth for successful reproduction, growth and survival. Four of the common species (collared pikas, Arctic ground squirrels, hoary marmots, Dall sheep) exhibit a wide range of physiological, behavioural and life-history adaptations for living in extreme and variable environments, but are adversely affected during poor seasons. If herbivores are unable to adapt to changes in climate, plant phenology or growth as a result of rapid warming in the Yukon, some of our evidence suggests that long-standing plant-herbivore interactions in the alpine may become less stable.

F3. Molecular biology of hormones

F3.1 Control of *Arabidopsis* transition to flowering: A novel route involving ABA binding to *FCA*

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Transition to flowering is a critical developmental step in plants controlled by multiple regulatory genes. The Arabidopsis floral promoter FCA is one of the most studied of the identified flowering genes. It encodes an RNA-binding protein with a WW protein interaction domain. FCA has at least two known functions: negative regulation of its own transcript and downregulation of the floral suppressor, FLC. Both functions require the interaction with the polyadenylation factor, FY to promote flowering. Here, we provide the first description of a function for a protein that binds abscisic acid (ABA). FCA binds ABA with high binding affinity that is stereospecific and follows receptor kinetics. Treatment of Arabidopsis with ABA increases FLC levels and delays flowering. An early flowering Arabidopsis ABA-deficient plant shows lower FLC levels. ABA inhibits FCA/FY interaction, thus regulating the GUS expression pattern of transgenic Arabidopsis, indicating an effect of the hormone on the autoregulation of FCA transcript. The specific mechanisms involved have a strong similarity with the initial events associated with hormone binding to nuclear membrane proteins in mammals, suggesting the possibility of a common mechanism for this

type of hormone signal transduction in higher organisms.

F3.2 Characterizing cytokinin oxidase (CKX) throughout the development of R50 (*sym 16*), a pea mutant accumulating cytokinins

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Cytokinins (CK) are an adenine-based family of hormones, the content of which is regulated by cytokinin oxidase (CKX). The pea mutant R50 (*sym-16*) displays several traits indicative of increased CK content. This accumulation was recently confirmed by LC-MS-MS and may be a result of either reduced activity or expression of CKX. Biochemical activity was detected through development of R50 and wildtype (WT) by a colorimetric assay (Liberos-Minotta and Tipton, 1995). Activity in mature, dry and imbibed seeds of R50 was low but comparable to that of the WT. In contrast, a significant decrease in CKX activity was noted in the roots and shoots of R50 seedlings and mature plants, which mirrored the previously obtained CK profile (Ferguson *et al.,* 2005). Using two recently published ESTs (Vaseva-Gemisheva *et al.*, 2003), we have begun to analyze the *Pisum sativum* CKX (PsCKX) gene family. Protein alignment and Southern blotting indicate that the PsCKX gene family is similar to those of *Arabidopsis* and maize in that it is a multi-gene family containing a conserved FAD-binding domain. The CKX expression profile throughout development is now being pursued to complement the biochemical data.

F3.3 A gain-of-function mutant of AtMKK9 affects multiple hormone signals in *Arabidopsis*

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Mitogen-activated kinases (MAPKs) are important signaling modules in all eukaryotes. The large number of MAPK cascade components identified in the *Arabidopsis* genome, relative to other organisms, suggests that these signaling modules have expanded roles in the Plant Kingdom. However, the interactions between MAPK modules, as well as the downstream targets of MAPK signaling cascades in plants are poorly understood. Amongst recent efforts to better characterize these pathways, it has been proposed that a MAPK cascade including AtMKK4, AtMKK5 and AtMPK6 participates in the regulation of ethylene production by stabilizing ACS6, a rate-limiting enzyme of the hormone biosynthesis pathway (Liu et al., 2004). In this study, we report that another MAPK kinase (MKK), AtMKK9, may also be associated with the ethylene pathway. AtMKK9 is a class D MKKs in *Arabidopsis*, a group for which no functions have been assigned to date. We have cloned AtMKK9 and modified the gene to encode a catalytically active (CA-MKK9) variant of the kinase. In order to investigate the downstream events regulated by the activity of AtMKK9, we have transformed *Arabidopsis* plants with CA-MKK9 under the control of a dexamethasone-inducible promoter. The induction of CA-MKK9 results in a rapid rise in ethylene production. Moreover, this ethylene burst is followed by the appearance of lesions that resemble accelerated senescence, a process known to be modulated by ethylene. In order to gain further insight into the cellular processes controlled by AtMKK9, we are investigating the early changes in transcriptional activity resulting from CAMKK9 induction, using full-transcriptome 70-mer oligo microarrays. The early activity of CA-MKK9 appears to result predominantly in repression of transcription. Amongst the down-regulated genes may be potential regulators of jasmonic acid biosynthesis. Overall these results point toward a role for AtMKK9 in modulating multiple hormone signals in *Arabidopsis*.

F3.4 Cytokinin-induced changes in gene expression and epigenetic inheritance in Arabidopsis

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Epigenetic studies seek to understand the regulation and inheritance of gene expression that control the phenotype. Exogenous cytokinin 6-benzylaminopurine (BAP) to populations of *Arabidopsis thaliana* Landsberg *erecta* induced aberrant floral phenotypes and changes in transcript levels of numerous

genes. Some of the altered transcript levels were heritable. Analysis of genetic responses using *Arabidopsis* Affymetrix GeneChips® indicated that BAP treatment significantly altered transcript levels of as many as 1765 genes (approximately 8% of the genome). Microarray analysis of transcript levels in the first non-treated generation (i.e. offspring of the BAP-treated plants) detected a significant change in 247 genes. This study identifies gene-families and pathways that may be responsive to cytokinins in a heritable fashion.

F3.5 Developmental regulation of the GA biosynthesis genes, *GA20ox*, *GA3ox*, and *GA2ox* during germination and young seedling growth of pea (*Pisum sativum*) L.

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To understand the role of gibberellins (GAs) during germination and early post-germinative stages of large-seeded dicotyledonous plants, we profiled the expression pattern of genes encoding three regulatory GA biosynthesis enzymes (*PsGA20ox1*, *PsGA3ox1* and *PsGA2ox1*) in pea (*Pisum sativum* L.) using real-time RT-PCR. To broaden our inferences on the role of GAs in these processes, we compared the GA biosynthesis gene expression patterns in two distinctly different genotypes of pea ('Alaska' a model cultivar for vining pea containing the wild-type internode length gene *LE* and 'Carneval' a model cultivar for semi-leafless field pea containing the *le-1* mutation producing shorter internodes), both of which germinate readily on imbibition under normal environmental conditions. Residual amounts of *PsGA20ox1*, *PsGA3ox1*, and *PsGA2ox1* transcripts were detected in the mature embryos (0 days after imbibition; DAI) of both genotypes. Transcription of *PsGA20ox1*, *PsGA3ox1*, and *PsGA2ox1* mRNAs occurred in all tissues examined (cotyledons, embryo axis, shoots and roots from 0.5 to 6 DAI) and was developmentally regulated within each tissue. Cotyledonary GA biosynthesis gene transcript patterns suggest that a signal from the axis triggers GA biosynthesis in the cotyledon. The high levels of *PsGA20ox1* and *PsGA3ox1* mRNA in the embryonic axis at 1 DAI suggests that the embryo axis is a major site for GA biosynthesis for stimulation of axis expansion. GA biosynthesis gene expression in 2 to 6 DAI shoots and roots (when their growth in fresh weight and length increased linearly) indicates a key role for *de novo* GA biosynthesis in early growth of seedlings. Supported in part by NSERC grant #138166.

F3.6 Functional analysis of GAMYB gene in rice.

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GAMYB is a MYB related transcriptional factor and activates transcription of hydrolysis enzyme gene during seed germination under controlling of gibberellins (gas).
We isolated lower GAMYB expression mutant and analyzed pleiotropic expression and biological function of GAMYB gene in rice. The mutant showed lower expression of osgamyb but did not defect of GA3 responsiveness. The mutant also showed pleiotropic expression on a wide range of plant developmental processes such as number of floret, primary rachis branches, tillers, flower initiation, internodes elongation, elongation of leaf and leaf sheath. We also analyzed osgamyb overexpressor and osgamyb rnai lines. The osgamyb overexpressor showed increased number of caryopsis, plant height, number of primary rachis branches, late flowering under long day condition than WT. The rnai lines showed decreased plant height, less number of primary branches and early flowering under long-day condition like as low expression of osgamyb mutant. The results showed that the osgamyb suppress the transition from vegetative to reproductive state under long-day condition. So the GAMYB also controls flowering in the short-day plant, rice.

We also investigated the effect of exogenous GA3 on flowering and internode

elongation. GA3 treated WT showed strong internode elongation whereas the GA3 treated MT showed early flowering and no strong elongation. The results indicated that the GAMYB suppress the flowering under controls of GA3.

F4. Natural Products I

F4.1 Functional analysis of *EgMyb1*, a *Eucalyptus* R2R3-Myb gene involved in the regulation of phenylpropanoid metabolism

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R2R3-Myb transcription factors play important roles in regulating plant specific biological processes such as phenylpropanoïd metabolism including lignification. Indeed, the promoters of several genes encoding phenylpropanoïd and lignin biosynthesis enzymes contain Myb consensus binding sites.

We have previously cloned an R2R3-Myb gene, *EgMyb1* from a *Eucalyptus gunnii* xylem cDNA library. EgMYB1 show strong sequence similarity to several R2R3 MYB thought to act as negative transcriptional regulators of phenylpropanoïd genes (AtMyb4, AmMyb308/330). Analyses of transcript abundance indicate that EgMyb1 is preferentially expressed in *Eucalyptus* secondary xylem, of both stems and roots, and moderately expressed in leaves. In order to gain insight on EgMyb1's function, it was ectopically over-expressed in poplar, a well-established model for wood formation studies. Phenotypic characterization using histological, biochemical and molecular analyses of several transgenic lines will be presented. In order to discover potential targets of EgMyb1, we conducted a microarray analysis comparing the global gene expression between control and three of the transgenic lines. Real time quantitative PCR was used to confirm these data.

Altogether, our results suggest that EgMyb1 is a potential regulator of the phenylpropanoïd pathway genes, altering lignin and anthocyanin biosynthesis.

F4.2 Use of an array to assay phenylpropanoid pathway expression in developing bean (*Phaseolus vulgaris* L.) seeds.

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A variety of phytochemicals in bean, derived from the phenylpropanoid pathway, have important effects on human health, plant disease resistance, seed coat colour and nodulation. Gene sequences available for phenylpropanoid pathway genes in other species were aligned and the conserved regions were used to design PCR primers that were used for RT-PCRs with bean seedling RNA. The sequences of the cloned cDNA segments were determined and confirmed by BLAST searches. Fragments of thirty four structural and regulatory phenylpropanoid pathway genes were identified and used to construct a bean phenylpropanoid pathway array on a glass slide. Fluorescently-labeled cDNA probes were synthesized from RNA isolated from developing seeds of 12 bean varieties and hybridized individually to the array with an equivalent amount of control cDNA consisting of an equimolar mixture of cDNA from all 12 bean lines labeled with a different fluorescent dye. Levels of red and green fluorescence associated with each spot on the array were measured with an array scanner. The results indicated that the array could be used to assay bean germplasm for the activities of many phenylpropanoid genes in developing seeds, simultaneously. The beans were also analysed for total phenolics, tartaric esters, flavonoids, and anthocyanins. Significant positive and negative correlations were found between microarray-determined phenylpropanoid gene activity levels in developing seeds and the levels of phenolics in the mature seeds. The results suggest that this methodology could be used to screen bean germplasm for the activities of genes coding

for proteins directing the synthesis of important secondary metabolites.

F4.3 Selective cytotoxicity of red grape wine polyphenols against MCF-7 breast cancer cells

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Food components influence the physiology by modulating gene expression and biochemical pathways within the human body. The disease-preventive roles of several fruit and vegetable components have been related to such properties. Polyphenolic components such as flavonoids are strong antioxidants and induce the expression of xenobiotic-detoxifying enzymes. The mechanism of selective cytotoxicity induced by red grape wine polyphenols against MCF-7 breast cancer cells was investigated in relation to their interference with calcium homeostasis. MCF-7 cells showed an increase in cytosolic calcium levels within 10 minutes of treatment with the polyphenols. Immunohistochemical localization of calmodulin showed similar levels of gold labelling in both MCF-7 and the spontaneously immortalized normal MCF-10A cells. Polyphenol-treated MCF-7 cells showed swelling of ER, dissolution of nucleus and plasma membrane, and reduced mitochondrial membrane potential. These cells were arrested at the G2/M interphase. MCF-10A cells did not show such changes after polyphenol treatment. The results suggest that polyphenol-induced calcium release may disrupt mitochondrial function and cause membrane damage causing selective cytoxicity in MCF-7 cells. This property could further be refined into a prevention strategy against breast cancer either independently or in conjunction with conventional prevention therapies where a positive drug-nutrient interaction can be demonstrated.

F4.4 Evaluation of antioxidant and anti-proliferative properties of polyphenols from novel grape lines

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Numerous epidemiological studies have shown an inverse relationship between the intake of fruits and vegetables and the incidence of certain cancers. Dietary polyphenolic compounds may act as anti-cancer agents by a variety of mechanisms, including antioxidant or free radical scavenging activity. Grapes contribute to the phenolic composition of wine, which is partly responsible for the protective effects against cardiovascular diseases and certain cancers. Polyphenol content of thirty unreleased lines of grapes developed at the Vineland Grape Research Station, University of Guelph, were evaluated and three high polyphenol lines (containing 600-900mg/100g fresh weight) were selected for further studies. Antioxidant activity as measured by superoxide scavenging capacity showed a dose-dependent increase with increasing polyphenol content whereas, hydroxyl radical scavenging capacity was relatively lower in all the three grape lines. Polyphenol fractions from these grape lines also showed selective cytotoxicity towards MCF-7 breast cancer cells. This shows that polyphenols from the unfermented grapes also possess selective anti-proliferative properties towards MCF-7 cells, just as those in wine. MALDI-TOF-MS analysis of anthocyanins from the selected grape lines showed variations in their composition that might be related to the unique differences in their antioxidant characteristics. Anthocyanins identified by MALDI-TOF-MS include glycosides (with acetyl and coumaroyl derivatives) of Malvidin, Petunidin, Cyanidin, Peonidin and Delphinidin.

F4.5 Plant ?-glutamyl hydrolases and folate polyglutamates: characterization, compartmentation, and co-occurrence in vacuoles.

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?-Glutamyl hydrolase (GGH, E.C. 3.4.19.9) catalyzes removal of the polylglutamyl tail from folyl and *p*-aminobenzoyl polyglutamates. Plants typically have one or a few GGH genes; *Arabidopsis* has three, tandemly arranged on chromosome 1, which encode proteins with predicted secretory pathway signal peptides. Two representative *Arabidopsis* GGH proteins, AtGGH1 and AtGGH2 (the At1g78660 and 78680 gene products, respectively) were expressed in truncated form in *Escherichia coli* and purified. The recombinant enzymes had low K_m values (0.5 – 2 µM) for folyl and *p*-aminobenzoyl polyglutamates but, despite 80% sequence identity, differed in mode of action. AtGGH1 cleaved pentaglutamates mainly to di- and triglutamates whereas AtGGH2 yielded mainly monoglutamates. Analysis of subcellular fractions of pea leaves and red beet roots established that GGH activity is confined to the vacuole and that this activity, if not so sequestered, would deglutamylate all cellular folylpolyglutamates within minutes. Purified pea leaf vacuoles contained an average of 20% of the total cellular folate, compared to ~50% and ~10%, respectively, in mitochondria and chloroplasts. The main vacuolar folate form was 5-methyltetrahydrofolate, of which 50% was polyglutamylated. In contrast, the principal mitochondrial and chloroplastic forms were 5-formyl- and 5,10-methenyltetrahydrofolate polyglutamates, respectively. In beet roots, 15 to 60% of the folate was vacuolar, and was again mainly 5-methyltetrahydrofolate, of which 75% was polyglutamylated. These data point to a hitherto unsuspected role for vacuoles in folate storage. Furthermore, the paradoxical co-occurrence of GGH and folyl polyglutamates in vacuoles implies that the polyglutamates are somehow protected from GGH attack or, less plausibly, that GGH and folyl polyglutamates are segregated into distinct vacuole subpopulations.

F5. Soil fertility and forage management

F5.1 Spatial and temporal variability of crop yield and soil fertility in relation to landscape

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A field study was conducted on hummocky landscape at the Manitoba Zero Tillage Association Research Farm near Brandon Manitoba to determine the spatial and temporal variability of crop yield as related to soil fertility and landscape position. This information was used to develop a method for delineation of management units related to precision farming. Variable-rate fertiliser management systems can improve efficiency of fertilizer use and environmental sustainability. Adoption of this technology has been hampered due to the difficulty of classifying fields into management units, the high cost of sampling soils on a grid basis, and the variability of soil and plant properties in the landscape.

Soil fertility, surface curvature, crop yield, weed populations and plant diseases varied considerably across the landscape in this study. Yield zones were identified based on cluster analysis of 4 years of normalized crop yield data. Although distinct yield zones were identified, soil nitrate-nitrogen and soil phosphorus were highly variable within these zones. Furthermore soil fertility and crop yield varied considerably between years within yield zones. This variability was attributed to excess soil moisture and flooding in the landscape. These sources of spatial and temporal variability of crop yield and soil fertility will be considered in the development of management strategies for variable application of nitrogen fertilizer.

F5.2 The effect of nitrogen source, rate and season of application on forage yield, protein content and nitrogen uptake

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A project was initiated in 2002 at two locations in Northern Alberta (Peoria and Rycroft) to assess the effect of ammonium nitrate, urea, polymer coated urea and NBPT-treated urea on forage yield and protein content. Nitrogen in these sources was applied

at four rates (0, 60, 80 and 120 kg N ha^{-1}) both in the fall and in the spring of each year (starting in the fall of 2002). Fall fertilizer application was carried out in late fall (October 30, 2002 and November 3, 2003) with a Hege applicator, both years on snow (approximately 2.5 and 20 cm in 2002 and 2003, respectively). Spring fertilizer applications were carried out on May 1, 2003 and April 6, 2004 on partly frozen soils. Peoria had been seeded to meadow bromegrass, whereas Rycroft to a 50-50 mixture of smooth bromegrass and alfalfa. Yields were higher with spring fertilizer application in three of the four site years. Overall, all fertilizer sources, except the polymer coated urea, provided the same yield responses both with fall and spring N application. Higher N uptake in the forage was obtained with application of NBPT-treated urea. Maximum yield, protein and N uptake was obtained with the application of 80, 80 and 100 kg N ha^{-1}, respectively.

F5.3 Influence of forage management and species on soil mineral nitrogen supply rates and seasonal dynamics

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Soil mineral-N (MN) supplies have ecological, agricultural and environmental implications. Grasslands are noted for low MN supplies, but little is known about MN dynamics under short term forage stands and impacts of management. Species were 30-yr-old grass (OG), 3-5 yr.-old alfalfa (A) (*Medicago sativa* L.), meadow bromegrass (MB) (*Bromus riparius* Rhem.) and annual-cereal (ANN). The OG was a mixture of smooth bromegrass (*Bromus inermis* Leyss.), quackgrass (*Elytrigia repens* (L.) Nevski) and Kentucky bluegrass (*Poa pratensis* L.); the ANN was a mixture of winter triticale (X *Triticosecale* Wittmack) and oat (*Avena sativa* L.). Management was either hay or pasture. Pasture was a grazed, 1.2 ha-paddock, while hay was an exclosure within each paddock, all replicated three times for 3 yr. Each year OG, MB and ANN received 100 kg N ha^{-1} of fertilizer-N; all species received 30 kg P_2O_5 and K_2O. MN supply rate was determined as a biweekly flux using paired cation and anion probes (PRS ^{TM}) inserted and removed from identical slots in three locations within the paddock and exclosure from May to October. Nitrate-N (NN) and MN supply rates were 9 to 10 times higher in May compared to Oct. Initially MN and NN supply rates for hay > pasture, but by mid summer pasture > hay and by fall hay = pasture. Averaged over season and years NN supply rate for ANN was 2 to 5 times more than OG; A and MB were intermediate and similar. Spring MN and NN supply rates for ANN were higher than others; by July OG was very low and remained constant until Oct. MN and NN supply rates of others declined more slowly from spring to fall, but ANN, MB and OG were similar by Oct.; A maintained higher MN and NN supply rates into fall despite receiving no fertilizer-N. In spring ammonium-N (AN) supply rate for OG was 3 times other species, then, decreased to the same level as others by July. The NN: AN ratio peaked for hay in June, then decreased until a low level by fall; the ratio for pasture rose above hay to peak in July after one grazing, then decreased. The NN:AN ratio for OG was 5 to 20 times lower than other species from May to August. The NN:AN ratio of ANN was 3 to 4 times greater than MB and A from May to July. MN supply rates of short term species were larger, had higher NN content and were more dynamic throughout the season than OG. Management effects were more subtle than species effects on MN supply.

F5.4 Forage yield and persistence of three short-lived perennial grasses in monoculture and mixture with alfalfa at a semiarid location in southern Saskatchewan.

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Crop rotations in semiarid regions of the Canadian prairies typically do not include perennial forage crops. Three short-lived perennial grass species, Dahurian wildrye grass (*Elymus dahuricus* Turcz. Ex Griseb), intermediate wheatgrass (*Elytrigia intermedia* (Host) Nevski) and slender wheatgrass (*Elymus trachycaulus* (Link) Gould ex Shinners), were compared for their forage productivity and persistence as short duration (3 years) forage stands, either in a mixture with alfalfa (*Medicago sativa* L.) or in monoculture,

in three trials at Swift Current, Saskatchewan. Intermediate wheatgrass produced more forage and was more persistent than Dahurian wildrye or slender wheatgrass when grown in monoculture in all three trials. In alfalfa mixtures, however, the grasses produced similar forage yield in 2 of the 3 trials. Yield compensation by alfalfa grown in mixture with Dahurian wildrye and slender wheatgrass appeared to maintain total forage yield as these grass stands thinned during the third year of the trials. Dahurian wildrye appeared to have the best potential as a short-lived grass species for short-rotation forages in annual cropping systems of this region.

F5.6 Evaluation of the Illinois Soil Nitrogen Test for Estimating Potentially Mineralizable Soil Nitrogen

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Nitrogen is a major constituent of all plants, and thus is considered one of the most important nutrients in crop production. As a result, crops are usually supplied with large amounts of nitrogen from synthetic fertilizer or manure. However, it must also be considered that 1 to 4% of soil organic nitrogen from soil organic matter (SOM) may be mineralized to inorganic nitrogen during a single growing season. Therefore, an understanding of the behaviour of nitrogen in the soil-plant system is essential for maximizing agricultural productivity and profitability while reducing the impacts of nitrogen fertilization on the environment. In a fully phased rotational study, potentially mineralizable N was estimated in the wheat phase of a field pea-wheat-sweet clover-oat rotation using the Illinois Soil Nitrogen Test for amino sugar-N.

F6. Carbon flow in the environment.

F6.1 For peat's sake: The role of fungi in peatland carbon dynamics

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Peatlands are a dominant landform in the northern hemisphere, covering about 12% of Canada's landscape and storing between 180 and 277 Gt carbon (C). Fungi are important decomposer organisms (saprobes); however, relatively little is known about fungi in peatlands. To date, anamorphic ascomycetes are most frequently isolated fungi from peatlands, representing nearly 63% of all taxa. Zygomycetes are the second-largest group of fungi and represent 10% of all taxa. Chytridiomycetes, teleomorphic ascomycetes, and basidiomycetes are less frequently isolated from peat and together comprise 11% of all isolates. The remaining 16% of isolates were not identified or were sterile taxa. From a functional perspective, purely saprobic and mycorrhizal fungi significantly influence C dynamics, with the former group being the primary decomposer organisms. Each of these groups is characterized by a suite of extracellular enzymes, which facilitates the degradation of specific plant storage and structural C polymers. Organic matter decay follows a sequence whereby simple molecules are generally degraded prior to more complex polymers, often polyphenolic in nature. Litter quality variables, such as N and P concentrations, figure prominently in the succession of fungi as organic matter decomposes, while peat and water chemistry variables appear to be of less importance to saprobes. Hence, litter comprised of substantial quantities of complex polymers, such as lignin and related polyphenolic complexes, decomposes very slowly. Surprisingly, recalcitrant polymer degraders, mostly basidiomycetes and select groups of ascomycetes, have rarely been isolated from peat, which may explain the accumulation of these polymers in peat. Mycorrhizal fungi are widespread in peatlands and shuttle nutrients and water from the soil solution to their host plants. While their enzymatic profiles may be more limited compared to saprobes, many of these fungi have the ability to degrade simple and complex polymers as well and hence may also be significant decomposers of organic matter. How disturbances, such as climate change and wildfire, affect peatland microbial communities and their roles is virtually unknown. These aspects of peatland microbial ecology require immediate attention.

F6.2 The Sensitivity of Douglas-fir (*Pseudotsuga menziesii* Mirb.) Root

Respiration to Seasonal Temperature Change in Three Age Classes of Coastal Forest

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Root respiration contributes 33-60% of total CO_2 efflux from the soil and its rate is significantly affected by temperature. The sensitivity of roots to temperature change is routinely estimated using a Q_{10} (i.e. the proportional increase in respiration for a 10°C increase in temperature). If acclimation in either respiratory capacity or temperature sensitivity occurs, then Q_{10} will be an inadequate predictor of respiratory response. We investigated seasonal changes in biomass-specific respiration rate and Q_{10} in the roots of Douglas-fir from three forest sites on Vancouver Island. These sites were all disturbed at one point and were at the time of measurement 55 (DF49), 16 (HDF88) and 4 (HDF00) years of age. Respiration measurements were carried out from May-Aug of 2004. Sites were visited biweekly and fine white roots were excised and immediately used to obtain respiration rates at a number of different temperatures (Spring - 8, 15, 22 and 29°C; Summer - 10, 16, 22 and 28°C). Rates were based on how quickly root biomass consumed O_2 in an aqueous buffer, as determined by a Hansetech oxygen electrode. Rates of root respiration at constant temperature were much higher in the latter part of the season, showing no within-site acclimation response to warmer soil temperatures. There was a general trend for the HDF00 site to level its respiratory response in late summer whereas results from the DF49 and HDF88 sites did not show this tendency. During summer, respiratory capacity of roots from DF49 was significantly higher than in roots from HDF88. The temperature sensitivity (Q_{10}) of respiration increased consistently throughout the season at all sites, from values near 1.3 in late May-early June to ~2.4 in late July and August.

F6.3 The role of common mycorrhizal networks in belowground carbon flow between plant neighbours

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Ectomycorrhizal fungi can readily form belowground hyphal connections between root systems, providing a pathway for direct interplant carbon and nutrient flow. Transfer through common mycorrhizal networks (CMNs) has been suggested as one potential factor regulating composition and structure of plant communities. We examined ectomycorrhizae and belowground carbon transfer between paper birch (*Betula papyrifera* Marsh.) and Douglas fir (*Pseudotsuga menziesii* (Mirb.) Franco) using pulse labelling in the laboratory and field to determine: 1) effects of transfer pathways (CMN versus soil) on the magnitude of carbon transfer, and 2) effects of host plant phenology on the direction and magnitude of carbon transfer. Results from the laboratory showed that significantly more net carbon transfer occurred when hyphal connections were intact than severed. In the field, carbon transfer was examined between paper birch and Douglas fir in spring, summer, and fall using dual ^{13}C-^{14}C pulse labelling. At the spring labelling, birch were just leafing out while fir was pre-bud burst, whereas in summer, birch was fully leafed out and fir was actively growing. In the fall, birch was starting to senesce and fir had set bud for the winter. This was reflected in different photosynthetic rates of the two species. Significantly more carbon was transferred from paper birch to Douglas fir in summer, and from fir to birch in the spring and fall. Our laboratory and field findings extend previous studies, providing evidence that ectomycorrhizal fungi facilitate belowground carbon transfer between paper birch and Douglas fir. Further studies are required to determine if transfer amounts are of sufficient magnitude to influence plant productivity and diversity.

F6.4 Uptake of inorganic carbon by the acidophilic green alga *Chlamydomonas acidophila*.

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The acidophilic alga, *Chlamydomonas acidophila,* will grow in media ranging in pH from 2.5 to 7.0, but displays the highest growth rate at pH 7.0 and the photosynthetic rate at pH 7.0 is twice that at pH 2.5. The alga expresses an external carbonic anhydrase (CA) when grown in media above pH 5.5 and

CA activity increases as the pH of the medium is raised. Attempts to inhibit external CA with acetazolamide, usually thought to be a weakly-permeable inhibitor of CA, was found to inhibit photosynthesis, indicating that it permeates these cells and inhibits internal CA.

In order to investigate the mode of inorganic carbon (Ci) uptake, external CA was removed by brief digestion with trypsin. The affinity of trypsin-treated cells for Ci was lower than that of control cells indicating that external CA is required for effective photosynthesis at neutral pH. Comparisons of the rate of photosynthesis at pH 7.0 with the calculated rate of CO_2 supply demonstrated a lack of active bicarbonate uptake and mass spectrometric monitoring of CO_2 and O_2 fluxes by the cells gave no evidence of active carbon dioxide uptake. Determinations of the overall internal pH of the cells, using ^{14}C-benzoic acid or [2-^{14}C]-5,5-dimethyloxazolidine-2,4-dione, showed that the cells maintain a neutral internal pH over an external pH range of 3.0 to 7.0. These data indicate that the ΔpH between the cell interior and the external medium was great enough to allow the accumulation of inorganic carbon by the diffusive uptake of CO_2.

F6.5 Non-photosynthetic carbon metabolic flux in *Chlamydomonas reinhardtii*: ^{13}C-isotopomer distribution ratios of proteinogenic amino acids

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The alga *Chlamydomonas reinhardtii* can grow either as a photosynthetic autotroph using CO_2 as carbon source or as a heterotroph using the two-carbon compound acetate, which is converted to succinate and glyoxylate via the glyoxylate pathway. The objective of our research program is to develop flux maps of the metabolic pathways affected by this change in carbon source. *Chlamydomonas* (wild type strain CC-125 mt$^+$) were grown under continuous light (500 µmol/m^2/sec PAR) in two identical mini-chemostats (100 mL). Nutrient media (minimal growth medium with 10 mM acetate) was pumped into each chemostat at a rate of 83.5 µL/min. 24 Hours prior to the addition of ^{13}C-acetate, one chemostat was placed in complete darkness while the other was kept in the light. ^{13}C-labeled acetate (1-^{13}C) was supplied to both chemostats by replacing 25% of the unlabeled acetate with ^{13}C-labeled acetate. The algae in the chemostat overflows were collected for 1-hr periods at 24 and 48 hours after the addition of ^{13}C-labeled acetate. The algal proteins were extracted and hydrolyzed, and the amino acids derivatized with methyl chloroformate (MCF), and separated by GC-MS. The isotopomer distribution ratios of the derivatized amino acid mass spectrometry fragments were calculated on the basis of the distribution of their individual ion counts, correcting for the natural occurrence of ^{13}C atoms. The isotopomer distribution ratios of these proteinogenic amino acids showed that both in the light and the dark (i.) a steady state of carbon metabolism was obtained within 24 hours after the start of the addition of ^{13}C-labeled acetate and that (ii.) this steady state continued until the end of the experiment (50 hours). Furthermore, there was a significant difference between the isotopomer distribution ratios of the proteinogenic amino acids isolated from dark- and light-grown algae. For example, the ratios between the fractions containing 0, 1, or 2 ^{13}C atoms of the mass 144 fragment of MCF-derivatized leucine was 0.752 : 0.171 : 0.077 in the light-grown, compared to 0.884 : 0.053 : 0.063 in the dark-grown algae. These isotopomer distribution ratios of the proteinogenic amino acid mass fragments are used to calculate a metabolic flux map of the pathways involved in the metabolism of acetate by *Chlamydomonas* under varying nutritional and environmental conditions.

F6.6 Grazing and topographic effect on short-term litter decomposition in a natural rangeland.

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Plant litter decomposition is fundamental to maintain the natural fertility and productivity of natural ecosystems. Evidence shows that by consuming the more nutritive foliage, grazing animals negatively affect litter quality. Therefore, does grazing decrease litter decay rate? Topography also influences soil characteristics and plant communities. Higher soil fertility at the bottom slopes will produce

more NPP and litter. Does this litter have different decay rate than litter produced in other topographic levels? If so, how will grazing animals interact with topography and modify the litter decay rate?, will this affect nutrient cycling and ecosystem sustainability?. Two experiments conducted on Black Chernozemic sandy loam at Kinsella Ranch, Central Alberta, will answer these questions. For the topographic effect, litter was mixed and placed at upper (U), middle (M), and lower (L) topographic levels in grazed (G) and ungrazed (UG) plots in a random block design. For the grazing effect, litter collected in U, M, and L levels from G and UG plots was placed in the same conditions in a nested design. To determine the mass loss (decomposition) is utilized the litterbag technique. There are six incubation times, and five repetitions. As litter chemical composition is related to decomposition, non-polar (NPC) and water-soluble carbon (WSC), holocellulose, lignin, and ashes were determined. The following results refer to the first incubation time (66 days). The mass loss was faster ($P<0.05$) in L (14.9%) than in M or U levels (12.8% and 11.4%). This occurred because the water filled porosity was higher in L (0.6±0.1) than in M or U (0.3±0.1) levels. Graze negatively effected ($P<0.0001$) litter decay. M and L had faster decomposition in UG (19.2% and 27.5%) than in G (11.5% and 22.1%). G and UG had similar values (13.2%, 15.7%) in U location. Differences in litter quality, as a result of grazing, explain the decomposition rate. G litter had lower ($P<0.05$) proportion of NPC than UG litter (38.7 vs. 54.6 mg g^{-1}) and higher values ($P<0.05$) of holocellulose (633.4 vs. 610.9 mg g^{-1}) and lignin (139.6 vs.127.9 mg g^{-1}) in M location. A similar trend was detected in L level. There was a clear micro-topographic effect on litter decomposition, and decay was negatively affected by grazing. Implications for the ecosystem are slow nutrient cycling, and less available nutrients for plant uptake. This condition will decrease NPP and eventually a shift of species; over time less C will be allocated to the soil.

F7. Tree physiology

F7.1 Are hybrid poplar clones commonly grown in the Canadian Prairies adaptable to soil moisture deficit?

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As a result of predicted climate change, many plant species may be faced with new environmental conditions to which they are not well adapted. Woody plants species such as poplars (*Populus* spp.) may be particularly vulnerable unless they are sufficiently adaptable to the new environment. Poplars are native to moist areas, but hybrid clones are being grown in many areas of the Prairies where moisture may be limiting. As a result, there is increasing concern whether currently used poplar clones will be able to withstand the increasingly severe and more frequent drought periods that are predicted. This project, examines the responses of several common Prairie clones of hybrid poplar ('Brooks 6', 'Hill', 'Northwest', 'Tristis', 'Walker' and 'WP69') to soil moisture deficit. The treatment consisted of several dry-down cycles during which the soil water potential reached about –1.3 MPa, followed by re-watering. As expected, plants growing under a soil moisture deficit had lower total biomass and leaf area than well-watered plants. There was however, no significant difference in root biomass between treatments, due to differences in root growth response between clones. As a result, there were significant clone x treatment interactions in leaf area:root wt ratio. Also as expected, clones growing under soil moisture deficit had higher water-use efficiency (WUE) as determined by their $\Delta^{13}C$ values. Results from this study indicate that differences in WUE and growth, and possibly drought tolerance, exist among the hybrid poplar clones studied here, which will allow for selection of superior hybrid poplar clones that will be best adapted to local growing conditions accounting for future climate change.

F7.2 Phytohormonal regulation of transpiration (E) and stomatal conductance (g_s) among canopy layers of a mature Sugar Maple (*Acer saccharum*) stand.

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Despite vastly different microclimates and functional roles that exist among mature hardwood canopy layers, there has been little or no study of corresponding hormonal physiology. Cytokinins (CK) have been associated with the regulation of stomata and gas exchange in several plant species (eg. *Zea mays, Commelina communalis*); however results seem to be species specific. This study addresses the potential role of CKs in regulating leaf gas exchange in mature trees. A canopy walkway at the James McLean Oliver Ecology Centre (JMOEC) was used to study a stand of ~80 year old sugar maples (*Acer saccharum*). Leaf samples were collected from the upper and lower canopies of 10 trees over the month of August 2003. CKs within leaves were identified and quantified using (LC –(+) ESI-MS/MS). The outer region of the upper canopy had the highest CK levels. All leaves contained higher levels of ribosides, in particular trans-Zeatin riboside and Dihydrozeatin riboside (active forms), than nucleotides or free base forms of CK. The upper canopy had higher transpiration (E) and stomatal conductance (g_s) coinciding with higher PAR, UV-B and leaf temperature. Total CK concentrations had significant positive linear relationships with E. *In situ* applications of low concentrations of the synthetic CK, Benzyl aminopurine and high concentrations of Abscisic acid (ABA) both reduced E and g_s in the upper canopy. Results suggest that CKs tend to accumulate in canopy leaves in response to severe microclimate conditions and play a role in controlling water loss. Study has been continued through 2004 to further illuminate the roles of CKs and ABA and any potential interaction of the two phytohormones. Leaf gas exchange parameters and microclimate were monitored at 3 positions on 6 trees from June-July 2004. Leaf samples were collected weekly for quantification of CKs and ABA. This should help clarify the apparent redundant function of these two phythormones within a forest canopy.

F7.3 Growth and water transport in mycorrhizal seedlings of *Pinus banksiana* and *Picea glauca* treated with NaCl

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Salinity is a major environmental factor that adversely affects plant growth and survival. Salinization of the soil is a world-wide growing problem which presently affects about 10% of the world's land. Although various salts may contribute to the salinity problems, NaCl is the most common and highly phytotoxic form of salt NaCl inhibits aquaporin-mediated root water transport and upsets osmotic, nutrient and water balance in plants.

Our earlier studies demonstrated that ectomycorrhizal associations of *Hebeloma crustuliniforme* with *Picea glauca* reduced root and shoot uptake of Na^+ and increased stomatal conductance in salt-treated plants. We obtained similar results with several different species of ectomycorrhizal fungi in association with various conifer and angiosperm tree species. In the present study, we inoculated six-month-old *Pinus banksiana* and *Picea glauca* seedlings with three different strains of ectendomycorrhizal fungus *Wilcoxina mikolae* v. *mikolae*, *W. remhii* and *W.* sp. that were collected from the boreal forest in northeastern Alberta. The non-inoculated (mycorrhizal control) and inoculated seedlings were treated with 0 (NaCl control) and 45 mM NaCl. Although the effects varied between the studied strains, the results suggest that *Wilcoxina* offered little protection to both species of conifer seedlings against salt stress. The results will be discussed in the context of seedling responses to salt.

F7.4 Carbon Isotope Discrimination provides a powerful option for the identification of high Water Use Efficient Cashew (*Anacardium occidentale* L.) clones for crop improvement

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Cashew is an important commercial crop cultivated in dry and marginal lands in peninsular India. Water is the most predominant limitation to achieving the yield potentials of this crop. As per the Passioura's growth and yield model, water use associated with root systems and water use efficiency are the important determinants of total biomass. We examined this model using selected constrating Cashew clones raised in containers. The total water used and biomass accumulated over a period of 150 days was determined using one-year-old seedlings by adopting gravimetric approach. Water Use

Efficiency, the amount of biomass produced per unit water transpired showed (WUE) showed a significant positive relationship with total biomass indicating that total biomass among Cashew clones can be enhanced through improvement in WUE. Further, since the propagation of Cashew is through vegetative means, identification of high WUE types as scion material for grafting on a know rootstock can be exploited to enhance yield potentials. We noticed a strong inverse relationship between Carbon Isotope Discrimination ($\Delta^{13}C$) and WUE among the Cashew clones. We report here for the first time that the Carbon Isotope Discrimination approach can be used to identify the desirable clones for scion material. To examine this aspect, the leaf samples from the filed established trees corresponding to the clones examined in the container experiments were collected and their $\Delta^{13}C$ was determined. A significant positive regression of $\Delta^{13}C$ values between the established trees and container grown seedlings confirms that carbon isotope ratios can be adopted as an accurate tool to identify the trees from which scion material can be obtained and grafts thus developed is expected to increase drought tolerance as well as Cashew productivity under dry environments.

F7.6 Functional analyses exploring the impact of high nitrogen availabity on wood formation in *Populus*

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Poplar species and their hybrids are being used in intensive forestry because of their fast growth rate. In order to sustain the growing demand for wood products, foresters are using silvicultural practices, like nitrogen fertilization, to increase biomass. However, surprisingly little is known about how nitrogen nutrition affects wood formation. In this project, we are investigating the effects of nitrogen availability on wood formation and wood properties in *Populus trichocarpa X deltoides*. Histochemical analyses of greenhouse-grown sapling supplied with 0 mM, 1mM or 10mM ammonium nitrate for up to 28 days showed that xylem tissues from high N-treated trees displayed a decreased lignin deposition relative to adequate or low N-treated trees. This observation was confirmed by chemical analyses using pyrolysis molecular beam mass spectroscopy and Klason lignin determination. Microscopic and fibre quality analyses indicated that at least part of this difference in lignin deposition is due to an increased duration of xylem development. Analysis of gene expression profiles by microarrays revealed a suite of genes that show differential transcript abundance under high versus low nitrogen availabilit. A number of genes associated with wood formation were found to be diffentially expressed according to nitrogen availability. Certain genes involved in lignin biosynthesis show decreased levels of transcripts. In addition, specific genes encoding putative components of signal transduction pathways are upregulated in response to elevated nitrogen. Our results suggest that nitrogen availability has a dual effect on wood formation: i) by increasing the duration of xylem maturation and ii) by altering resource partitioning via particular metabolic pathways which are in part mediated by changes in gene expression.

G1. Resistance to diseases: Unraveling the plant responses (part 2)

G1.1 An alternative agriculture approach opens a new door for enabling sustainable disease tolerance in crops

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Crop losses due to diseases and the cost of controlling them through off-farm chemicals amount to tens of billions of dollars. Pre-harvest and post-harvest losses occur maximally in rice, wheat and corn, with soybeans, cotton, coffee and barley being close. Approaches used for disease resistance

include: breeding and selection of resistant/tolerant lines, spraying off-farm chemicals, and, in recent years, enhancing host defense via targeted genetic engineering. Extensive chemical use – nitrogen fertilizer, pesticides and plastic mulch, for instance, to combat pathogens in conventional vegetable production potentially contributes to environmental pollution. In recent years, alternative agriculture practices have tested cover crops like hairy vetch (*Vicia villosa*) as a source of on-farm biological inputs to reduce erosion and the use of agrochemicals without impacting the yield or quality of the produce. Interestingly, interactions between the cover crop mulch and the tomato plant in the field plots were found to result in delayed leaf senescence and increased disease tolerance as compared to the plants cultivated in plastic mulch. To understand the molecular basis of these beneficial attributes, we tested whether increased longevity and disease tolerance of vetch grown tomato plants are linked to increased expression of specific defense genes and subsequent accumulation of their products. Our laboratory has identified several key genes that have been implicated in senescence or disease tolerance, one group of which is also involved in carbon and nitrogen metabolism while the second constitutes key hormone receptor kinase(s) and hormone biosynthesis genes. A model, built from these data, is being tested to understand how these interactions empower disease tolerance and increased longevity of tomatoes.

G1.4 High frequency of rearrangements in R-gene loci of the progeny of tobacco plants exposed to compatible virus

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Previous work in our laboratory has shown that a compatible interaction between the pathogen *Tobacco mosaic virus* (TMV) and the plant *Nicotiana tabacum* (tobacco), results in the production of a systemic signal that led to changes in plant genome stability. This locally generated signal named systemic recombination signal (SRS), traveled faster than a virus and was able to trigger a systemic somatic recombination increase in non-infected plant tissue. Moreover, the rearrangements in the transgene of infected plants were transmitted to the next generation. This phenomenon if directed on the right gene (read R-gene) may constitute as an adaptive measure to viral infection. While analyzing the progeny of plants that received SRS we observed an 8.2-fold increase in the instability of N-gene-like, R gene loci. In contrast, the actin-like loci showed no sign of instability. Such loci-specific differences in gene rearrangements could possibly be explained by epigenetic control over gene reshuffling. Indeed, changes in R-gene stability were paralleled by global genome, by actin-like loci-specific hypermethylation and R-gene loci-specific hypomethylation. Genome hypermethylation of the progeny is believed to be part of general protection mechanism against stress, whereas the locus-specific hypomethylation is associated with the higher frequency of rearrangements. Such a specific methylation pattern, paralleled by increase in recombination events, triggered by a pathogen attack, could be a sign of adaptive changes in plants.

Our data suggests the existence of a specific epigenetically controlled mechanism that promotes rearrangements in R-gene loci upon contact with a compatible pathogen. Future studies are clearly needed to understand the signal specificity and the mechanism underlying the methylation changes.

G1.5 Methyl-jasmonate and salacylic acid regulate expression of defence-related genes and reduce infection by common bunt in wheat (*Triticum aestivum* L.)

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In most incompatible plant-pathogen interactions, defence responses are triggered following activation of signalling pathways involving salicylic acid (SA) and jasmonic acid (JA). The effects of SA and JA applied to wheat seedlings after 1-, 2-, or 3-wks following emergence, on infection levels and transcript levels of pathogenesis-related (PR) genes following inoculation with *Tilletia laevis* (Kühn in Rabenh.) were investigated. SA, Actigard7 (formulation of SA, Syngenta), and methyl-jasmonate (MeJA), applied to the highly susceptible wheat variety >Laura= reduced infection levels as much as 100%, 100%, and 82%, respectively, compared to the untreated, inoculated controls. Using Real-Time PCR (RT-PCR) to study the regulation of gene

expression for several classes of PR-genes, three patterns among the treatments were observed: 1) Genes that were upregulated in response to MeJA treatment compared to the corresponding untreated checks. Their expression was potentiated by infection by *T. laevis* and the highest transcript levels were associated with the 3-wk seedlings. The seven genes were *Chitinase1 (chit1), chit3, chit4, PR-1.1, PR-1.2, ß-1,3-glucanase 1*, and a lipase; 2) Genes that were upregulated in response to both SA and MeJA, not potentiated by infection, and the highest transcript levels were associated with the 1-wk seedlings. These included the non-soluble lipid transfer proteins ns-LTP-1 and ns-LTP-2, and *ß-1,4-glucanase 3.* 3) One gene, a *ß-1,3;1,4-glucanase 2* responded exclusively to SA, was potentiated by infection, and the highest expression with associated with the 3-wk seedlings. These results demonstrated that exogenous applications of SA and MeJA applied to *T. laevis* infected wheat differentially activated different groups of defence-related proteins and reduced infection.

G1.6 Bacterial Cyclic b-(1, 2) Glucan acts in systemic suppression of Plant Defense Responses

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Plants are able to defend themselves against attack by a variety of potential pathogens through the deployment of both constitutive and induced defenses. To cause disease, a successful pathogen must counter or evade pre-formed defenses and suppress or fail to elicit induced defences. We show here a systemic suppression of plant immune responses by the extracellular cyclic β-(1,2)-glucan of the phytopathogen *Xanthomonas campestris* pv. *campestris* that is associated with the transport of the molecule throughout the plant. This is, to our knowledge, the first report showing that a phytopathogen suppressor is systemically transported. Systemic suppression is a novel counter-defensive strategy that may facilitate pathogen spread in plants, and may have important implications for the understanding of plant-pathogen co-evolution and for the development of phytoprotection measures.

G2. Arctic and alpine plants (part 2)

G2.1 Evolution of arctic species of bluegrass (*Poa*)

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Poa is a worldwide primarily cool temperate genus comprising over 500 species, and is one of the more speciose genera in the arctic. The evolution of arctic species is examined first in the context of a worldwide phyogeny of the genus based on trnT-trnF and ITS sequence data. The nine Canadian arctic species belong to at least five distinct lineages in four of the five major clades of the genus. Each species has a unique evolutionary history, with its closest relatives outside of the arctic mostly in alpine areas. Biogeographic and phylogenetic evidence support a Eurasian origin for four species, a North American origin for two species, while the origin of three species remains unclear. Confict between and within the two data sets plus high ploidy levels suggest the presence of both ancient and recent hybridization in the evolution of arctic

species. Four species were further examined for infraspecific variation within North America using restriction site and/or sequence data. *Poa pratensis* s.l. and *P. arctica* s.l. were found to have a complex phylogeographic history, and apart from *Poa pratensis* subsp. *alpigena*, which may be distiguished based on a large cpDNA deletion, there was little support for infraspecific taxa. A preliminary analysis of *P. glauca* suggests higher diversity in the western low arctic combined with hybridization with closely related species in the southernmost part of its range. In the case of *P. hartzii* infraspecific cpDNA varation detected in high arctic populations is considered to be the result of *P. glauca* chloroplast capture via hybridization and introgression.

G2.3 Migration and Speciation in Moonwort Ferns (*Botrychium* subgenus *Botrychium*) in North America.

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Moonwort ferns, *Botrychium* subgenus *Botrychium*, are a common but often overlooked component of arctic, alpine, and boreal habitats of North America. Three new species and numerous new species disjunctions have been recorded in Alaska and Canada since 2000. This diversity results from at least two waves of dispersal and diversification. The first established a set of North American endemic and near-endemic diploid and allotetraploid species including a distinct North American genotype of *B. lunaria*. This probably occurred during and as a result of Pleistocene glaciation. A more recent migration of European *B. lunaria* into northern North America has produced a second set of allotetraploid species.

The presence of two different genotypes of *B. lunaria* has resulted in the peculiar phenomenon of morphologically and genetically distinct tetraploid species having been produced from different combinations of the same diploid progenitors. Increased awareness of *Botrychium* among arctic and alpine botanists will likely result in discovery of additional species occurrences that will further clarify the history and diversification of these ancient ferns.

G2.5 Haplotype diversity patterns among arctic and alpine *Packera* species: species level promiscuity and the heartbreak of hybridization.

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Phylogeographic analyses of arctic and alpine plants have provided insights into the origin and evolution of these floras, especially in Europe. Similar studies in North America are less common. Our phylogeographic studies of *Packera* (Asteraceae) have included species from both arctic and alpine regions and have highlighted the importance of hybridization, migration and genetic isolation in the evolution of these species. Arctic species contain cpDNA haplotypes from both coastal and interior regions to the south, suggesting the importance of two different migration routes in their evolution. Alpine species also contain haplotypes from these two regions and vary in the level of intrapopulational haplotype diversity they contain. Breeding system also plays a role in determining the amount of intrapopulational haplotype diversity present. Overall, the haplotype variation patterns highlight and emphasize the importance of hybridization in the evolution of *Packera* species in both arctic and alpine regions and reveal a surprising level of genetic diversity within populations.

G3. Photosynthesis and light responses

G3.1 A comparison of methods to measure photosynthesis in field grown soybean (*Glycine max* L. Merr)

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Research has found that the relationship between photosynthetic rate and yield is variable and dependent on crop, environment and method of determination. Future modification to the photosynthetic capacity of a plant and the incorporation of these characteristics into varieties with plant breeding will require instruments that measure photosynthesis rapidly, reliably and are easy to operate. A field comparison was made between the Minolta Spad 502 Chlorophyll Meter, the Opti-Sciences OS-30 Chlorophyll

Fluorometer and the LiCor 6200 Portable Photosynthetic System in two soybean trials. One comparison was done in varieties with known differences in photosynthetic rate, while the other was done in a population of F5 sister lines selected from a cross made between varieties differing in photosynthetic rate and maximum leaf area index. During the growing season concurrent measurements were made with the three instruments. Data will be reported on the relationship among these instruments and the advantages and disadvantage of each instrument will be discussed.

G3.2 Deactivation of excess excitation energy in plants exposed to low light and chilling stress

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In order to cope up with a sudden change in the environment and consequent stress induced damages, plants develop several protective mechanisms. The major one is the prevention of the damages to photosynthetic electron transport caused by reductants and ATP formed in excess of the demand for CO_2 assimilation. Plants exposed to moderate light during cold mornings are vulnerable to photo-oxidative damages as carbon fixation is limited. In a group of chilling-sensitive plants (*Cucumis sativus* L., *Lycopersicum esculentum* L., and *Zea mays* L.) and chilling–tolerant *Hordeum vulgare* L., exposed to moderate light at low temperature, photosynthetic electron transport is disrupted at multiple sites and the quantum yield of PSII declines. This process and the severity of damages to PSII is distinct from the one caused by excessive light at room temperature. In the latter, quenching of the excitation energy by non-photochemical pathways (NPQ) in the time-scale of several seconds to minutes confers protection for the functional components of the photosynthetic apparatus. However, it is unclear yet how absorbed light energy is dissipated in plants exposed to moderate light and chilling stress. In the present study, we evaluated the contribution of the mechanism(s) by which the excess energy is quenched in such conditions. For this purpose, the dark relaxation of the fluorescence rise kinetics (from induction curves) was analysed in samples exposed to a combined stress and then allowed to recover at room temperature. The recovery experiments reveal that: (i) the fast relaxing components of the NPQ (energy dependent quenching of fluorescence and quenching related to state-transitions) does not participate in the dissipation of excess energy during chilling-induced photoinhibition; ^{2} the recovery process is much slower as the quenching due to photoinhibition increased; and ^{3} either oxidized P680 or the inactivated PSII centers may act as an efficient quencher of excitation energy. These aspects will be discussed in detail.

G3.3 Reversal in foliar orientation from normal by reverse application of phototropin (s) /(resultants) in *Epipremnum aureum* or sakina akhter effect in *Epipremnum aureum.*

S.M.N.BABAR

Reorientation of leaves in *e.aureum* is an advance phylogenetic character and the process starts as soon as the leaves are disturbed from their normal position.a major role played, other than phototropin (s) / hormone (s). ® is the directional flow of the resultants_ from lamina to petiole resulting in spiral stress in petiole or reorientation i.e. Adaxial surface which earlier moved away from proper light moves toward appropiate light. (normal behaviour) on the other hand if under normal conditions “resultants” are made to flow in reverse order the adaxial surface becomes abaxial deviation from the normal behaviour “sakina akhter effect”. Hence the directional flow of resultants is responsible for the orientation and reorientation of leaves in *E.aureum.*

G3.4 The role of ethylene in phytochrome – cryptochrome cross-talk in Arabidopsis seedlings grown under low and high R/FR ratios with equal blue light irradiance

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Low red to far red (R/FR) ratio is the major component of the shade avoidance syndrome (SAS) in plants. The reduction in total red light (relative to far red light) caused by shading from canopy or neighbouring vegetation is detected by a red/far red photoreceptor, phytochrome. In Arabidopsis, several reports have indicated interaction of phytochrome with cryptochrome (blue light photoreceptor) on a

molecular level. In shade, there is a reduction not only in total red light, but also in total blue light. Photoreceptors have been shown to work in concert with phytohormones and thus determine plant form. Ethylene, a gaseous plant hormone, has been suggested to play a major role in establishing the etiolated phenotype associated with plant growth in shade. We have set up an experimental system, where both low and high R/FR ratios are coupled with the same blue light and total light irradiance. Two Arabidopsis cryptochrome mutants: *cry2-1* (contains a large deletion of the CRY2 gene, background – *Col-4*) and *cry1* (lacks blue light-dependent inhibition of hypocotyl elongation, background – *Ler-0*) were selected to investigate whether the absence of phytochrome-cryptochrome cross-talk will alter the morphology and ethylene production of Arabidopsis seedlings grown under low R/FR ratio conditions. Here, we will present our data and discuss the role of blue light in SAS and its interaction with ethylene.

G3.5 Endogenous gibberellin (GA) levels in response to varied light irradiance in *Stellaria longipes* alpine (sun) and prairie (shade) genotypes

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Plants grown under low light irradiances (low PAR [photosynthetically active radiation]) show a shoot elongation growth (etiolation) similar to that observed under enriched far red light (FR). Individuals of the *Stellaria longipes* alpine population (1D non-shade [sun] genotype) grow on an open alpine plateau at 2,453 m in the southern Alberta Front Ranges. They have very short internodes and small, but wide leaves. In contrast, plants in a lower elevation population (7B [shade] genotype) grow in an open "prairie-type" grassland in the lower foothills. They have long internodes and long and narrow leaves. We used 1D and 7B as model sun and shade plant systems to study the role of GAs in the etiolation associated with low PAR. Both genotypes responded to low PAR treatment in a similar manner with regard to increased stem growth. However, reduced PAR modified the endogenous GA_1 levels of the two genotypes quite differently. The 7B "shade" genotype showed increased GA_1 (the "growth effector" GA) levels over time under low PAR, while the 1D genotype showed just the opposite trend. Exogenous application of growth-active GA_3 and GA_4 significantly promoted etiolation of 7B genotype, even when grown under low PAR, but had no significant effect on the 1D "sun" genotype under low PAR. Thus, while the growth responses to reduced PAR are similar (in direction, but not magnitude) for these two genotypes, which have very different phenotype characters, their endogenous phytohormone (GAs) responses are quite different. It appears that the phenotype plasticity associated with the "shade" 7B genotype is directly related to its ability to synthesize GA_1 and respond to applied GAs. In contrast, the "sun" 1D genotype, with a very reduced stature, appears to have muted its response to relatively high endogenous GA_1 levels, and also to applied GAs, e.g. some other factor is controlling the magnitude of its shoot growth response.

G4. Natural Products II

G4.1 Bioprospecting the wild blueberry: impact of nutrient management practices and biproduct utilization

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With increasing awareness of the antioxidant properties of the wild blueberry (*Vaccinium angustifolium* Ait.), more emphasis is being placed on developing cultural management practices to enhance fruit polyphenolic levels and assessing various processing biproducts as potential polyphenolic sources for the nutraceutical market. The effect of nitrogen (N), phosphorous (P), and potassium (K) fertilizers on the polyphenolic content was examined in 2003 and 2004 using Folin-Ciocalteu, pH differential and HPLC procedures. Results indicated that neither soil applied N, P or K, or foliar applied N treatments significantly affected total anthocyanin or phenolic content of ripe wild blueberry fruit. Conversely, upon examining the polyphenolic content of various plant components, significant differences in polyphenolic levels were present with the red leaves, green leaves, stems and immature berries having 1,147, 626, 511, and 141%

greater total phenolic levels respectively, than ripe blueberry fruit. Therefore, results from this preliminary investigation indicate that nutrient management practices have minimal effect on polyphenolic content of mature blueberry fruit. In addition, these studies indicate that the leaf and stem debris may provide a superior polyphenolic source for fractions destined for the nutraceutical market.

G4.2 Genetic transformation of Bee Balm (*Monarda didyma*)

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We are developing *Monarda didyma* (Bee Balm) as a model system to study regulation of isoprenoid (terpenoid) metabolism in plants. Bee Balm is a member of the mint family (*Lamiaceae*) grown throughout Canada, from British Columbia to Manitoba and Quebec. Several species are used for ornamental and medicinal purpose (e.g., for the treatment of stomachaches, headaches, and fevers), and its monoterpene-rich essential oil has extensive applications in aromatherapy and in the manufacturing of soaps and perfumes. Current work aims to establish a transformation procedure for this plant. An *in vitro* regeneration procedure (including callus, shoot, and root formation) has been developed. Work is underway to optimize the process using various auxins and cytokinins. Preliminary data shows that this plant grows well on Murashige and Skoog medium, and roots efficiently in response to Naphthalene Acetic Acid (NAA). In addition, cultured leaf discs produce callus and subsequently regenerate shoots in response to various cytokinins. This work has been supported by NSERC and internal OUC grants to SS Mahmoud.

G4.3 Modification of the carotenoid pathway in carrot roots to produce novel ketocarotenoids

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Carotenoids are the most abundant pigments in nature, and play an important physiological role in many organisms. Carrot roots contain both α and β carotenes but not the ketocarotenoids. Ketocarotenoids, such as canthaxanthin and astaxanthin, are found only in some algae; they are commercially important compounds for the aquaculture and pharmaceutical industries. For biosynthesis of astaxanthin, a β-carotene ketolase gene (from the alga *Haematococcus pluvialis*) and a β-carotene hydroxylase gene (from *Arabidopsis thaliana*) were cloned. Bacterial expression vectors containing the above genes were mobilized into β-carotene or zeaxanthin-producing recombinant strains of *Escherichia coli*. Our experiments demonstrated the efficient *in vitro* synthesis of astaxanthin/ zeaxanthin in recombinant *E. coli* that contained β carotene ketolase/β carotene hydroxylase genes, respectively. In order to alter the carotenoid pathway in carrot plants for astaxanthin synthesis, β-carotene ketolase gene and β-carotene hydroxylase gene were co-mobilised into carrot plants via *Agrobacterium*-mediated transformation employing the phosphinothricin acetyl transferase gene (*bar*) as a selectable marker. Several gene constructs were made using different plant promoters (CaMV-35S, CaMV-35S-2X and Arabidopsis Ubiquitin) as well as promoters from *Agrobacterium rhizogenes* (RolD, Ags, Mas and Ocs+Mas) in the binary vector pCAMBIA 1300. The pea Rubisco small sub-unit transit peptide sequence was used for targeting the novel enzymes into chromoplasts. We are currently regenerating carrot plants from tissue culture, which are putatively transgenic for β-carotene ketolase gene and/or β-carotene hydroxylase gene that are capable of producing novel ketocarotenoids.

G4.4 The relationship among morphological and anatomical features and technical ethnobotanical uses of the common cattail (*Typha latifolia* L.).

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A unique fusion of Western scientific investigation and ethnobotanical approaches was undertaken to establish an understanding

of morphological and anatomical features and their relationship to First Nations uses of the common cattail, *Typha latifolia* L. This species has been an important plant for a large variety of uses including storage, transportation and shelter. Morphological characteristics included leaf shape and size and stem length and diameter. At the anatomical level in both leaves and stems, in addition to gross structure, fibre location and abundance was found and proportion of aerenchyma tissue was calculated. It was determined that morphology and anatomy could be connected to specific First Nations technical uses. The general goals of this research are to build linkages between Western science and Traditional Ecological Knowledge (TEK) and to document and protect the ethnobotanical and environmental importance of *T. latifolia*.

G4.5 Phytochemical and agronomic variations in *Echinacea pallida* var. *angustifolia* plants from cultivated populations in British Columbia and Washington grown under common greenhouse conditions.

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The cultivation of E. *pallida* var. *angustifolia* (EAN), relative to that of other crop species, is in its infancy. This is reflected by observably high phenotypic variation within cultivated populations. The EAN market still relies on wildcrafting due to the misperception of greater potency in wild sourced material compared to cultivated material. As a first step in the establishment of an EAN breeding program focused on the development of phytochemically rich elite cultivars with superior agronomic qualities, we evaluated root phytochemical content and agronomic trait variations in greenhouse-grown plants harvested at 6 months of age. The plants were grown from seeds randomly selected from 4 cultivated populations in B.C., 1 in Washington and 2 wild populations. Significant variations in agronomic traits such as leaf trichome density were observed between cultivated populations. Correlations between phytochemical contents of shoots and roots were also measured to evaluate the possible effectiveness of a non-destructive genotype selection method based on shoot phytochemical content. The findings from these experiments have potentially important implications in up-coming genotype selection work.

G4.6 Incorporation of molecular and chemical methodologies as a reliable approach for Echinacea species identification

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Due to market demand, *Echinacea angustifolia* is most commonly grown as a medicinal herb in Alberta. There is confusion between this species and other relatively similar morphologically species called *E. pallida*. Authenticity of this species remains a problematic issue for the Echinacea growers to sell their products. This can be achieved through incorporation of molecular testing and chemical analyses of active ingredients. Seeds of the above two species were obtained from different sources in western Canada (Alberta, British Columbia, Manitoba and Saskatchewan) as well as *E. purpurea* originally obtained from Ontario. A set of twenty arbitrary oligonucleotide primers (Operons) was used to develop discriminating markers among *E. angustifolia*, *E. pallida* and *E. purpurea*. Using RAPD methodology, five primers (OPA 02, OPA 08, OPA 12, OPA 16 and OPA 17) were found capable to produce clear polymorphism among these species. Three of these primers (OPA 2, OPA 16 and OPA 17) in addition to another 3 reported primers (OPC 02, OPH 13 and OPM 02) were used in testing 43 samples provided by growers. These primer markers distinguished *E. angustifolia* from the other 4 Echinacea species used as controls (i.e. *E. pallida* and *E. purpurea, E. tennisie*, *E. paradoxa, E. atroruben*). Chemical testing on 37 root samples by HPLC showed that cichoric acid was less than 0.3 mg/gm and Echinacoside content ranged between 0.01-0.28. Developing a specific primer showed that simple sequence markers can be used to discriminate among the *Echinacea* species. Eleven potential

marker sites were identified, nine primer sets were designed and five of these markers show variation among the species.

G5. Breeding

G5.1 Characterization in Hard Spring wheat and phenotypic analysis in DH population RL4452/AC Domain for seed dormancy and pre-harvest sprouting resistance

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Pre-harvest sprouting (PHS) is a persistent threat to spring wheat production in the high rainfall area of western Canada. Sprouting damage in the harvested crop can reduce the value of the grain by up to 40%, and badly affects the milling and end-use quality. Red and dormant seeded genotypes are likely to be more PHS resistant than white and non-dormant seeded genotypes. The purposes of this study were: to characterize the PHS resistance of red and white seeded spring wheat germplasm, and to determine PHS of lines in a red seeded doubled haploid (DH) population derived from the cross RL4452 (low level of resistance) X AC Domain (high level of resistance).

Twenty five different wheat genotypes of various sources were evaluated for seed dormancy (SD) and PHS resistance in Glenlea, Manitoba and Lincoln, New Zealand in 2003. Red seeded genotypes had the highest levels of SD and PHS resistance. The red seeded genotype, AC Domain, was a good source of resistance for PHS. Good PHS resistance was observed in the red seeded genotypes AC Majestic and Red RL4137. The white seeded genotypes Kanata, White RL4137 and AUS1408 performed well only in Manitoba, but might be good sources of PHS resistance. Dormant seeded genotypes tended to have high levels of PHS resistance in both Manitoba and New Zealand.

In 2003 and 2004, 185 lines of DH population RL4452/AC Domain were also phenotyped for SD and PHS. Highly significant differences among lines and nearly normal distributions for all variables suggest that genetic variation for SD and PHS exists within the population. Transgressive segregates were also detected outside the dormancy and resistance levels of the parents in this population. This suggests that not only AC Domain alleles, but also RL4452 alleles contributed to SD and PHS resistance in the population. The phenotypic values collected on the RL4452/AC Domain population will be merged with a previously developed genetic map to identify QTLs that control PHS and SD.

G5.2 Introgression between *Sinapis alba* (Yellow mustard) and *Brassica napus* (Oil-seed rape) as a potential source for genetic enhancement and crop improvement

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The tribe *Brassiceae* comprises of economically important genera including edible seed oil species *Brassica napus* and *Sinapis alba* which offer a range of agronomically important complementary characteristics. *Sinapis alba* (yellow mustard; SalSal, 2n = 24) is grown in Canada as a condiment crop and provides important reservoirs of useful genes for resistance to major diseases and insect pests and tolerance to abiotic stresses. *Brassica napus* (oil-seed rape; AACC, 2n = 38) is an important potential source of high oil content and high seed yield of canola oil (low glucosinolate and low erucic acid). Canola-quality *S. alba* lines have been developed at the AAFC, Saskatoon Research Centre. To widen the Brassica gene pool and for transfer of useful alien genes, we have produced intergeneric F1 hybrids of *S. alba x B. napus*. Intergeneric reciprocal crosses were made, using ovule and embryo culture techniques. *S. alba* & x *B. napus* % crosses resulted in 4.3% intergeneric F1 hybrids, whereas reciprocal crosses of *B. napus* & x *S. alba* % yielded only 0.35% intergeneric F1 hybrids. The hybrid plants were intermediate between the two parents and were confirmed by flow cytometric analyses and morphological characterization. The F1 hybrids were male sterile and were

confirmed by cytological examination of pollen mother cells. None of the F1 hybrids set any seeds after self-pollination or cross pollination of the F1 hybrids, however backcrossed plants were obtained through embryo rescue. Intergeneric hybridization between the two genera (*B. napus* and *S. alba*) is expected to combine the desirable characteristics of *B. napus* (high oil content and high yield) with *S. alba* (drought tolerance and disease resistance) in oil seed breeding. The aim of current research is to create and utilize the genetic diversity between *Sinapis* and Brassica species and transfer valuable traits from *S. alba* to *B. napus* and vice versa. Furthermore, the F1 hybrids represent a population with DNA from both species and combining traits for biotic and abiotic tolerances along with agronomic traits is an important objective in canola breeding. Likewise, increasing oil content and seed yield for yellow mustard is essential for the development of yellow mustard as an oil seed crop. These F1 hybrids also offer an opportunity for genetic and molecular studies. The prospects of wide hybridization which offers a promising approach to enrich the current gene pool and to improve the oil seed rape including yellow mustard as a potential oil crop are discussed.

G5.3 Optimum Growth Conditions for the Selection of dual-purpose flax in the CDC Breeding Program.

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In Canada, flax (*Linum usitatissimum* L.) is grown for its seed oil. However, a major disadvantage associated with growing oilseed flax is the straw. Flax straw is difficult to incorporate into the soil after harvest. Instead, the majority of flax straw is burned in the field. This increases the workload for farmers, as well as creating air pollution. Agronomic concerns are also associated with burning, since it leaves fields vulnerable to wind and water erosion. A small market does exist for Canadian farmers to sell their flax straw for making high quality paper products and some plastic composites. However, fibre-based and fibre-using industries are growing world wide, and flax straw fibre is becoming an important product. Flax straw fibre concentration fluctuates among varieties and environments. High, consistent fibre concentrations are essential if the fibre in oilseed flax is to become an important product to Canadian farmers. This study will assemble the agronomic information necessary to increase the concentration of straw fibre in the Crop Development Centre (CDC) Flax Breeding Program. Three experiments were conducted: how seeding rate and row spacing affect straw fibre concentration, the effects of nitrogen fertilizer rates on straw fibre concentration, and also how seeding date affects fibre concentration. CDC Mons, Vimy, and CDC Gold were the oilseed varieties used, along with the fibre flax variety, Hermes. In all experiments, seed yield, protein and oil content were measured in order to understand the relationship of stem fibre concentration with these characters.

G5.4 QTL mapping of coleoptile length, root length, and seed vigor index in wheat under normal and osmotic stress conditions

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Wheat (*Triticum aestivum L.*) genotypes with longer coleoptiles and greater seed vigor index are preferred in arid regions, as these traits are considered to confer some form of drought tolerance This study was designed to localize quantitative trait loci (QTLs) controlling root length, coleoptile length, and seed vigor index under normal and osmotic stress conditions. A set of 108 recombinant inbred lines (RILs) of the 'International Triticeae Mapping Initiative' mapping population (ITMI), along with the parental lines 'Opata' and 'M6', were germinated under controlled conditions. Two separate randomized complete block designs each with two replicates; including one under normal conditions (0 bar) using distilled water and the other under simulated osmotic stress (-8 bar) imposed by poly-ethylene glycol (PEG-6000) were conducted on germinating seeds. Germination percentage, root length, and coleoptile length were recorded and seed vigor index was calculated. Using a composite interval mapping approach, one putative QTL was identified for coleoptile length (chromosome 6B), two for root length (chromosomes 5B and 7D), and two for seed vigor index (chromosomes 6B and 7D) under osmotic stress condition. These QTL were all different from those detected under unstressed osmotic conditions.

G6. Phytoremediation and xenobiotics

G6.1 Identification of plant species suitable for phytoremediation of hydrocarbon/metal contaminated soils

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Numerous plants have natural ability to remediate organic and inorganic pollutants. However, some species or ecotypes, and varieties within species demonstrate extraordinary phytoremedation properties. The challenge is to identify those species or ecotypes that would perform best under the soil and climatic conditions (moisture, temperature) of a given contaminated site. Four plant species, reed canary grass (*Phalaris arudinacea*), slender wheat grass (*Elymus trachycaulus*) and hay mix consisting of brome grass (*Bromus spp*) and alfalfa (*Medicago sativa*) were tested in soil beds in a greenhouse facility for their ability to reduce concentration of hydrocarbons and metals in landfarm soil amended with industrial sludge. Sludge was applied 11 times at three rates (0, 1 or 2 times the current landfarm rate). Toxicity of sludge application to plant growth was tested using weighed biomass, while soil microbial diversity was evaluated using BIOLOG® Ecoplates with multiple sole carbon substrates. No natural attenuation of total extractable hydrocarbons (TEH) in the fallowed soil was detected. Three-month-long application of sludge at the current and doubled industrial rates did not affect growth of the aboveground biomass of reed canary grass, while almost totally exterminated slender wheat grass. Establishment of strong and bulky root system prior to sludge application was a prerequisite for plant survival and thus for successful phytoremediation. All tested species contributed to reduction of TEH in soil amended with doubled industrial rate of sludge. Reed canary grass was the most efficient species and it was able to lessen TEH in soil to the control level. All tested crops treated with sludge increased BIOLOG® substrate utilization. However, root-promoted enhancement of microbial diversity that assisted in the loss of hydrocarbons in soil was the most pronounced in reed canary grass. Although all species were able to remove metals from sludge-amended soil, reed canary grass exhibited the highest phytoextraction potential. At the doubled application rate, this species extracted 59% of the elements while slender wheat grass and hay mix removed only 30% and 40% of the metals, respectively. Of the four species tested, reed canary grass is the most suited to be used for phytoremediation of landfarm soil.

G6.2 Effects of paper mill sludge on growth of plants in mine tailings

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Many areas in North America are contaminated with metal laden mine tailings that can move off site and contaminate adjacent sites and groundwater posing an ecological and environmental threat. To decrease the movement of metals off site, mine tailings could be biologically stabilized by establishing a vegetative cover. Amendments can be added to alleviate the physical and chemical properties of tailings which limit revegetation of the tailings. The objective of this study was to determine the effect of paper mill sludge and fertilizer on the emergence, survival and growth of alfalfa (*Medicago sativa*), red fescue (*Festuca rubra*), and slender wheatgrass (*Agropyron tachycaulum*) grown in mine tailings. The paper mill sludge increased emergence for alfalfa and slender wheatgrass, and had no effect on the emergence of red fescue. Survival of all three species was increased by paper mill sludge. Adding paper mill sludge resulted in growth of the three species that was similar to that of fertilized plants. Paper mill sludge and fertilizer together resulted in the greatest plant growth for alfalfa, red fescue and slender wheatgrass. Alfalfa grew the most out of any species, followed by red fescue. These two species seem especially well suited for biological stabilization, because they both developed large rooting systems. The results of this study suggest that paper mill sludge alone or in combination with fertilizer would be an effective way to increase plant growth in mine tailings.

G6.3 Does addition of crude oil to sub-boreal forest mini-ecosystems alter established ectomycorrhizal communities?

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Boreal and sub-boreal forest soils are occasionally subjected to localized surface contamination with oil spilled during petroleum extraction, refinement or transport activities. The extraradical mycelia of ectomycorrhizal (ECM) fungi account for most microbial biomass in forest floor soils and provide habitat (C sources and colonization surfaces) for heterotrophic bacterial communities. The impacts of oil on ECM communities (fungal and bacterial) are unknown. To determine if varying amounts of crude oil altered ECM communities in sub-boreal forest soils, surface-sterilized lodgepole pine or paper birch seeds were planted in approximately 10 g of either mor humus (FH) or coarse woody debris (CWD) layers (sources of microbial propagules) overlying sandy mineral (Bf) soil in Cone-tainer™ pots. Seedlings grew under greenhouse conditions for four months; "BC Light" crude oil (0, 0.073, 0.146, 0.219 g/ cm^2) was then applied to the organic soil surface around the seedling stem. Mini-ecosystems were destructively sampled at three-week intervals, starting one week after treatment. ECMs were described morphologically and then fingerprinted using amplicon length heterogeneity (LH-PCR) of fungal rDNA. Bacteria from mycorrhizosphere soils were assessed by LH-PCR of amplified rDNA, as well as by C source utilization profiles (Biolog EcoPlates™). At 10 weeks, most roots were colonized by several (1-7) ECM fungi. Some types appeared to be associated with specific plant or soil treatments, with a trend towards lower species richness with increased time since higher oil treatments. The density of culturable bacteria was $>10^4$ CFUs /g in organic and mineral soils after 1 week, regardless of plant, soil or oil treatment. At 7 weeks, C source utilization profiles did not vary significantly with oil treatment and most substrates were used by bacterial communities. LH-PCR analyses should provide increased resolution for trends seen in ECM communities following oil treatment.

G6.4 Phytoremediation of nickel contaminated soils using sunflower (*Helianthus annuus* L.) colonized by arbuscular mycorrhizal fungi

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Strategies of heavy metal (HM) removal from contaminated soils include the use of HM tolerant plant species, such as sunflower. Little is still known on the physiological impact of HM toxicity for plant survival, especially on nitrogen (N) and secondary plant metabolism. Current phytoremediation strategies focus on ameliorating plant tolerance and increasing HM uptake. The use of mycorrhizal fungi has been proposed since their association with plants has been shown to increase HM uptake and alleviate HM plant stresses. This study aimed to determine the impact of arbuscular mycorrhizal (AM) fungal colonization on the potential of sunflower, *Helianthus annuus* L. cv. Lemon Queen, for phytoremediation purposes, specifically of nickel (Ni). Sunflower plants were grown under greenhouse conditions in a factorial design, with or without AM inoculation by *Glomus intraradices* Schenck & Smith. After 8 weeks of growth, plants were subjected during 2 further weeks to four Ni concentrations (0, 100, 200 and 400 mg kg^{-1}), and then harvested. Ni accumulation was greater in roots > leaves > flowers for both AM and non-AM plants. Nickel contamination significantly decreased total plant dry mass (DM). Percent AM root colonization tended to decrease from 78% to 64% with the increasing Ni concentration input. The AM colonization did not significantly affect the DM or shoot height of Ni-contaminated plants. The protein concentration and activities of key N-assimilating enzymes (NR, GS and GDH) as markers of the primary metabolism, and the concentration in sesquiterpene lactones (characteristic of the secondary metabolism in the Asteraceae family) have been determined. Further investigation on the contribution of AM fungi to sunflower HM tolerance is still ongoing.

G6.5 Phytoremediation: Effects of AM colonization in 'wild' Tobacco plants grown in Zn-contaminated soils

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This greenhouse study aimed to determine the effect of colonization by the arbuscular mycorrhizal (AM) fungus, *Glomus intraradices* Schenck & Smith, in the plants of 'wild' tobacco (*Nicotiana rustica* L. Var. Azteca), under Zn-nutrient stress. Plants of *N. Rustica* were grown from seeds in AM or non-AM inoculated substrate and subjected to four soil-Zn concentrations (0, 50, 100, and 250 mg Zn kg^{-1} dry soil; from a $znso_4^-$ solution). Contrary to our previous stated hypotheses, tolerance to increasing soil-[Zn] was not enhanced in AM than non-AM plants; there were also no significant differences among the plant physiological parameters (e.g. Root or shoot mass, shoot height, or leaf chlorophyll and protein concentrations). In addition, it was found that the AM root colonization increases from 14% to 81% with increasing soil-[Zn]. Whereas no differences were found for Zn content in shoots, the AM roots subjected to the highest soil-[Zn] had significantly and almost half time lower Zn content than in non-AM roots. These results suggest that as the soil Zn toxicity threshold was likely not reached, the effect of soil Zn contamination in combination to AM colonization should be further experienced under more stressful conditions in *N. Rustica*. More importantly, it has been shown that Zn may enhance or be involved in AM root colonization. Accordingly, AM fungi could play an important role in soil-contaminate immobilization processes and may therefore hold an integral part in the phytoremediation process of heavy metal contaminated soils.

G6.6 The role of sclerids in the Cyperaceae: Relation to plant habit

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A recent study has determined the unusual presence of highly lignified sclerenchyma cells, or stone cells, within *Eriophorum vaginatum* L., a tussock forming sedge. This study suggested the survival of the species in nutrient challenging environments might be tied to the presence of stone cells. That is, stone cell function may be to facilitate internal nutrient recycling within the corm of this species. Stone cells were not found in the closely related, but rhizomatous (non-tussock forming), *E. schucherzeri* Hoppe. The mechanism of *E. vaginatum*'s survival and the role of stone cells have not been determined. We are investigating the presence of stone cells in relation to plant growth form (habit) in E. *vaginatum* and closely related species of the Cyperaceae. We are currently examining tussock forming and rhizomatous species, e.g. *E. vaginatum*, *E. angustifolium* Honck., *Scirpus cyperinus* L., *S. caespitosus* L., and *Carex* spp., to determine if the stone cells are a plant habit phenomenon. This research will contribute to the understanding of stone cells in their possible role in plant growth form, within the Cyperaceae. Findings to-date will be presented.

G7. Cell Biology

G7.1 Molecular control of nuclear and subnuclear targeting of the plant CDK inhibitor ICK1

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The precise control of the cell cycle relies on a complex gene network in which cyclin-dependent kinases (CDKs) play a central role. ICK1 is the first plant CDK inhibitor that has been shown to inhibit the activity of plant CDK and cell division, and affect plant growth and development profoundly [1,2,3]. ICK1 has distinct domains for its functions in inhibition of CDK activity, *in vivo* stability and nuclear localization [4]. In this study we investigated the sequences and mechanisms for regulating nuclear targeting of ICK1 using a large set of deletion and site-specific mutants fused to the green fluorescent protein (GFP) in transgenic *Arabidopsis* plants and tobacco cells. The central region of ICK1 contains a nuclear localization signal (NLS) (residues 78-86) that was able to target GFP fusion proteins to nuclei. Surprisingly, the N-terminal region could also confer nuclear localization independently, leading to the identification of a novel signal. Furthermore, the C-terminal region could also affect the nuclear localization of ICK1. In addition to nuclear localization, ICK1 showed discrete distribution in subnuclear domains. Results in identifying the sequences regulating the subnuclear localization will be presented. These results

indicate that the cellular distribution and nuclear localization of ICK1 are controlled delicately by independent mechanisms.

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[3] Zhou Y, Wang H, Gilmer S, Whitwill S, Keller W, Fowke LC (2002) *Planta* 215: 248-257.
[4] Zhou Y, Li G, Brandizzi F, Fowke LC, Wang H (2003) *Plant J* 35: 476-489

G7.2 Looking beyond the cytosol: Housekeeping enzymes targeted to multiple subcellular compartments in *Arabidopsis thaliana*

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Methylation occurs in every compartment of the plant cell, performing a wide variety of tasks ranging from viral resistance to chromatin modification. Yet two key enzymes involved in the methylation process, ADK and SAHH, are thought to be only localized within the cytosol. To investigate how the methyl cycle can occur in multiple subcellular compartments, we examined the localization of ADK and SAHH isoforms in *Arabidopsis thaliana*. Immunolocalization studies utilized 3-week-old meristem and leaf sections bound to antibodies specific for ADK or SAHH. The enzymes were then recognized using secondary antibodies conjugated to either 7 or 15 nm gold beads and detected using TEM. Complementary studies followed CaMV35S promoter driven ADK and SAHH cDNAs fused to GUS reporter genes throughout the cell. Both methods detected ADK and SAHH within the nucleus and chloroplast as well as the cytosol. In addition, this pattern of multiple targeting sites was not limited to the type of tissue or isoform being observed. These findings show great promise in expanding our knowledge of the methylation process. Future study will be dedicated to examining the transport of these enzymes into their respective cellular compartments.

G7.3 Determination of *cis*-acting regulatory elements of Arabidopsis ribosomal protein genes *ATRPL23A-1* and *-2*.

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Plant survival requires protein synthesis by ribosomes comprised of a 1:1 complex of large and small subunits that assemble on mRNA. The large (60S) subunit of the plant cytoplasmic 80S ribosome is comprised of three ribosomal RNAs (25S, 5.8S & 5S rRNAs), ~50 ribosomal proteins, and contains the peptidyl transferase center. In *Arabidopsis thaliana*, the large subunit proteins ATRPL23A-1 and -2 (L23A-1 & -2), are members of the conserved L23/L25 family of primary rRNA binding proteins that must be present for subsequent ribosome assembly. While Arabidopsis *L23A* gene products share 94% amino acid sequence identity, their 5' regulatory regions show less than 50% nucleotide sequence identity. To identify *cis*-acting elements necessary for *in planta* expression of *L23A-1* and *-2*, up to ~900 bp of their 5' untranslated regions (UTRs) were truncated from 5'- and 3'-ends, and fused upstream of *uidA*, encoding the ß-glucuronidase (GUS) reporter gene. Constructs were used to infiltrate Arabidopsis and expression was analyzed via GUS assays. Greatest expression of full length UTRs was observed in mitotically active tissues. Deletions that affected splicing of a UTR intron present in *L23A-1* and *-2* diminished or abolished expression. Putative *cis*-acting regulatory motifs differed between genes, but both shared interstitial telomeric motifs (*telo*-boxes) that appeared important for activity. Our findings suggest a link between the cell-cycle and activation of *L23A-1* and *-2*, and also indicate that this gene family may contribute to ribosome heterogeneity.

G7.4 Compartmentation of *de novo* NADP(H) biosynthesis: Subcellular and Tissue Distribution of NAD(H) Kinases *in Arabidopsis thaliana*

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In the model higher plant *Arabidopsis thaliana*, the *de novo* biosynthesis of the vital triphosphopyridine nucleotide NADP is catalyzed solely by two NAD^+ kinases (NADKs) and one NADH kinase (NADHK) that use Mg^{2+}-ATP to phosphorylate either oxidized or

reduced NAD respectively. Calmodulin, a ubiquitous eukaryotic Ca^{2+}-binding protein, responds to intracellular $[Ca^{2+}]_{cytosolic}$ fluxes or "signals" by activating numerous target enzymes. Interestingly, plants possess a Ca2+ and calmodulin (CaM)-dependent NADK that has been implicated in stress response and pathogen defence.

Since the chloroplast and peroxisome are considered to be impermeable to pyridine nucleotides produced in the cytosol, NADP(H) *de novo* synthesis by NAD(H)K isoforms would appear to be necessary for the supply of NADP. Unlike other eukaryotic mitochondria, plant mitochondria uniquely possess a $NAD(P)^+$-transporter that allows passive diffusion of $NAD(P)^+$ between the cytosol and mitochondrial matrix. The presence of this mitochondrial $NAD(P)^+$-transporter would appear to negate the requirement for a NADP-synthesizing enzyme that is present in, for example, yeast mitochondria.

Determining the compartmentation of each NAD(H)K is key to elucidating the *in vivo* role each NAD(H)K isoform may play in the cell. We have sought to determine the subcellular localization of each NAD(H)K isoform by a combination of transient *in vivo* expression of NAD(H)K-GFP fusion protein and immunological analysis of the subcellular fractions.

However, variances in NAD(H)K isoform transcription may determine the *in vivo* importance and role of each isoform during development and in different tissues. We have sought to explore the tissue distribution of the NAD(H)Ks through the use of NAD(H)K-promoter:: ß-glucuronidase (GUS) fusions in order to gain insights into the function of each NAD(H)K isoform in different tissues.

Preliminary findings and possible roles of NAD(H)Ks in higher plants are presented.

G7.5 Characterization of peroxisomal malate dehydrogenase; a microtubule binding protein.

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The cytoskeleton of plant cells acts as a basic framework for cell morphology and plays a prominent role in many cellular functions. This structure forms a dynamic network of highways for cellular trafficking of organelles, proteins and RNA. A tubulin-binding protein fraction was isolated from *Arabidopsis* suspension cell culture using tubulin chromatography. This protein fraction consisted of well known microtubule associated proteins as well as proteins involved in RNA binding, translation, signaling and various metabolic pathways. Malate dehydrogenase (MDH) was one of the many peroxisomal matrix proteins found in the tubulin binding fraction. To determine if MDH's association with tubulin conferred microtubule binding, a GFP-MDH fusion construct was bombarded into fava bean epidermal cells. Cells expressing the fusion construct labeled peroxisomes as well as filamentous structures in the cell cortex. The filamentous structures were identified as cortical microtubules through drug treatment studies. Further immunofluorescence studies showed that MDH did not label the other cellular microtubule arrays. These results suggest that MDH binds to microtubules prior to import into the peroxisomes. The role of MDH's association with microtubules is currently being investigated.

G7.6 An efficient, new selectable marker for the selection of transgenic plants

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Selectable markers play a pivotal role in genetic engineering by facilitating the selection of genetically transformed organisms out of a mixed population of transformed and untransformed individuals. Only a handful of approximately 50 marker genes that are currently available are in wide use. The most common among these is the gene conferring resistance to the antibiotic kanamycin. However, antibiotic-resistant genes have attracted much criticism because of concerns about spreading antibiotic resistance to human beings and animals. We have developed an efficient new marker based on a thiol methyltransferase (TMT) gene that circumvents these concerns. The enzyme encoded by this gene detoxifies thiocyanate and iodide ions by methylating them to volatile products. The presence of this gene thus allows transformants to grow in the medium containing above ions, which are highly inhibitory to untransformed individuals at low

concentrations. The efficiency of this selectable marker was tested in several species by using a convenient root culture system. Transgenic individuals could be visually selected in all species with high efficiency (75 – 100 %) within one week of transformation.

H1. Horticulture and greenhouse crops

H1.1 An overview of Northern Vigour® research in horticultural crops

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The production advantages of vegetatively propagated crops such as potatoes grown in northern latitudes compared to southern sources had long been known by potato growers in Europe and North America. Studies at the Department Plant Sciences of the University of Saskatchewan had previously demonstrated that the relative vigour and yield potential of seed potatoes produced in Saskatchewan were superior to those from the USA. This phenomenon has been termed "Northern Vigour®". The present projects examined the potential "Northern Vigour" response in strawberries (*Fragaria* x *ananassa* Duch.), garlic (*Allium sativum* L.), medicinal crops including narrow-leaved purple coneflower *(Echinacea angustifolia),* valerian (*Valeriana officinalis)*, burdock (*Arctium lappa*), floral crops calendula (*Calendula officinalis),* and aromatic crop catnip (*Nepeta cataria)*. The primary objective is to develop Saskatchewan as a supplier of high quality, superior-yielding planting materials, or sources of phytomedicinals for national and international markets. Studies in the U.S. were conducted in collaboration with UC Davis, Oregon State University and commercial growers. Several greenhouse and field studies were performed over a period of 10 years to examine the consistency of this environmental response and to develop more specific cultural practices to enhance certain responses. With some variations in response, in general, there is a significant and positive influence of northern locations on increasing plant quality.

H1.2 Aquaponics: A novel greenhouse production system and its challenges.

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Aquaponics is a combination of aquaculture and hydroponic greenhouse crop production systems. The two most important challenges for optimal plant growth in an aquaponic system are nutrient balance and pest management. Presence of fish and nitrifying bacteria as an integral part of this production system preclude the use of chemical pesticides for pest management, and is therefore limited to the use of biological control agents (BCA). Although a number of BCA are available for regular greenhouse production systems, little is known about their effectiveness in aquaponic systems. To test BCA, we have developed a mini-aquaponic system, which consists of a 125 cm diameter x 50 cm deep aquaculture tank, filtration system, and a plant tray with a surface area of 115 x115 cm^2 and 15 cm deep. The water flows from the aquaculture tank into the filtration unit and then into the plant tray with siphons, from where it is pumped into the aquaculture tank using a submersible pump. With this system, we are testing the effectiveness of *Paenibacillus polymyxa* strain PKPB1 for the control of root rot of cucumber cv. Spacemaster (*Cucumis sativus* L.) caused by *Pythium aphanidermatum*, and any potential harmful effects of *P. polymyxa* on fresh water fish from genus *Tilapia*.

H1.3 Modeling CO_2 emission scenarios in greenhouse tomato production

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Carbon dioxide enrichment of the atmosphere is considered to be essential for profitable greenhouse tomato production in British Columbia. Carbon dioxide trapping by tomato plants is beneficial in that it partly consumes the CO_2 produced with greenhouse heating. However, it is now customary for growers to apply additional CO_2, supplementing the amount co-generated during greenhouse heating, to enhance yields and profits. This will result in additional CO_2 escape to the earth's atmosphere, particularly when vents are open. Carbon dioxide escape is undesirable in terms of sustainability of the industry.

This work examines current CO_2 management practices in BC tomato greenhouses. The aims are: to quantify total carbon use and costs associated to achieve present yields, and to examine different CO_2 enrichment scenarios and their effect on yield and costs of production. The ultimate goal is to provide the industry with information to help improve their decisions on CO_2 enrichment, in terms of cost effectiveness and sustainability, and to provide society with an accurate representation of carbon usage under greenhouse conditions.

Two years of environment, yield, natural gas and liquid CO_2 gas data were collected from a local tomato greenhouse. Using the Acock (1978) model for photosynthesis, modified by Nederhoff (1994) and Chalabi (2002) canopy photosynthesis rate was determined from the environment data. Weekly fruit yield was determined using the method of Aikman (1996) and Chalabi (2002); a method where modelled photosynthesis rates are converted to grams of fruit produced and summed for a week for a given season.

The models used for estimating fruit production, fit reasonably well with the actual data; and were deemed useful for estimating production under different CO_2 scenarios. Using the Acock-Nederhoff-Aikman-Chalabi model yield responses to differing CO_2 concentrations will be calculated.

H1.4 N fertilization and rib discoloration incidence in crisphead lettuce

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Rib discoloration (RD) in crisphead lettuce has been associated with high temperature. However, the effects regarding other field factors such as N fertilization to the expression of the physiological disorder is unknown. A two-year experiment including two plantings per year was conducted to study the effect of N fertilization (0, 40, 80, 120 kg/ha) on the incidence of RD in two lettuce varieties grown on organic soils.

The 2001 season was particularly warm and dry in July and August and led to the production of small lettuce heads with a high expression of RD symptoms. The heads of the Ithaca variety were larger, less dense (planting 1 and 2) and heavier (planting 1) with a longer core (planting 1 and 2) than those of the Summertime variety. In planting 1, the Ithaca variety showed a high incidence of RD (85%) and no response of N levels while the Summertime variety had a lower incidence of RD without N application (58%) than with 40 kg/ha N application (85.4%). In planting 2, the Ithaca variety had a greater incidence of RD (66%) than the Summertime variety (47%), but there was no effect of N fertilization.

In 2002, lettuce heads were heavier (740 g) than in 2001 (546 g). In the first planting, the two varieties had the same head weight, size and incidence of RD, but the Summertime variety had a longer stem and a denser head than the Ithaca variety. N fertilization positively affected head weight, size and stem length, but not head density. Lettuce plants grown with 80 and 120 kg/ha of N showed a higher RD incidence (42% and 49%, respectively) than those grown with 0 (18%) and 40 kg/ha (24%). In the second planting, the Summertime variety had heavier, larger and denser heads, due to a late harvest expressed by longer cores (111 mm versus 69 mm for the Ithaca variety) and a higher incidence of RD (96%) compared to the Ithaca variety (85%). No effect of N fertilization on RD was observed.

Overall, this study showed that under highly conducive conditions, the N level had no effect on RD incidence. Under moderate disease expression (<40% incidence), applying 40 kg/ha compared to 80 kg/ha or 120 kg/ha reduced RD incidence by half, with no reduction in head size and density, and a limited reduction in head weight. Variety effect

was more important than N fertilization effect, suggesting that breeding for RD resistance is a more efficient strategy to reduce RD than adjusting N fertilization.

H1.5 Breeding Strawberries Rich in Antioxidant Phenolic Compounds

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Eighteen strawberry advanced lines and cultivars were evaluated for their phenolic content and antioxidant capacity using several methods. High antioxidant capacity was found for 'Harmonie', 'Saint-Jean d'Orléans' and 'Saint-Laurent d'Orléans', which reported to have better shelf-life than 'Kent'. However, the correlation between antioxidant capacity and post-harvest quality preservation was not always straight forward. Cultivar 'Saint Pierre', characterized by very good shelf life displayed similar level of antioxidants to other cultivars with shorter shelf life. 'Harmonie' 'Saint-Jean d'Orléans', 'Orléans', and some advanced selections had higher hydroxycinnamic acids, benzoic acids, and flavonols than 'Kent'. The significant variation in antioxidant capacity and total phenolic compounds in this study clearly shows the potential value of certain new cultivars and advanced lines to be considered as parents in a breeding program. The future plan is to examine individual antioxidant and their role in disease resistance and extended shelf life and to use selected genotypes as parents to developed new lines. The development of new strawberry fruit with higher antioxidant capacity not only may enhance resistance to diseases and increase shelf life but also stimulate greater interest in the nutraceutical aspects to strawberry consumption and potentially help to reduce risks of cancers and heart diseases.

H1.6 Automated monitoring of greenhouse plants

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A simple, non-invasive technology to automatically record the growth and water use of plants has been developed for use in research and commercial greenhouses. With indeterminant plants such as tomato, two pairs of load cells are used to measure up to 11 physiological and irrigation parameters simultaneously. One set of load cells (lower) measures the mass of the growing media sitting in a leachate collection trough, thereby enabling measurement of crop water use and 5 irrigation parameters. The other set (upper) is suspended from above to measure the mass of the plant canopy, and from that, 24-hour growth as well as the weight of harvested fruit. In the case of short-stature determinant plants, only the lower load cells are used, and somewhat fewer parameters are measured. Data from the load cells are collected on a data logger, subjected to preliminary on-board processing, and recorded every five minutes. The information is then relayed to a PC for presentation. Calculations are based on short-term changes in mass. Water use is expressed either as a rate per minute or as a cumulative total over 24 hours; growth is expressed as a 24 hour total. The extent of hydration (water content) of the plant canopy during the course of the day can also be inferred from the data. Additional information on percent media moisture and leachate percentage is collected and displayed automatically. The benefits for the grower include [1] better information about the current status of the crop [2] early warning about sudden changes in crop growth and water use, or the onset stress [3] easy comparison of different zones, seasons and years, and [4] relatively inexpensive equipment which can be easily expanded. For the researcher, information derived from the system may be used in conjunction with climate and plant physiological and biochemical data to develop models of crop function or performance.

H2. Cold stress

H2.1 Transcript Profiling in *Arabidopsis thaliana* using Serial Analysis of Gene Expression (SAGE).

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Many plants acquire an increase in freezing tolerance through cold acclimation, an exposure to low non-freezing temperatures. Complex physiological changes are associated

with the cold acclimation process, many of which are preceded by alterations in gene expression. The advent of high throughput genomic technologies combined with the completion of the Arabidopsis genome sequence allows changes in gene expression to be monitored on a global scale. We have used the Serial Analysis of Gene Expression (SAGE) to produce a quantitative analysis of gene expression, to identify differences between Arabidopsis plants growing under normal (22ºC;125µE) and low temperature conditions (3ºC;125µE) for varying amounts of time.

A total of five SAGE libraries were generated from plants exposed to normal temperature and four low temperature treated conditions, where the length of exposure was 30 minutes, 2 hours, 2 days and 1 week. These data are presently being analyzed in order to identify novel low temperature induced transcripts.

H2.2 Differential enzyme activities in *Brassica napus* cell suspension cultures exposed to low temprature and osmotic stresses.

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Analysis of gene products in plant cultures exposed to abiotic stress may reveal factors which may prove useful in imparting stress tolerance to crops. Previous work in our lab, using microspore-derived cell suspension cultures of *Brassica napus* cv. Jet Neuf as a model system, used comparative 2D PAGE to identify microsomal proteins that were differentially expressed in response to low temperature or osmotic stress. Assays were developed for several of the enzymes among these identified proteins, and the cells and stress treatments re-visited, in an attempt to confirm that differences in gel spot intensities correlated with differences in actual enzyme activities. Cells, typically grown at ambient room temperature (~21^{o} C), were either grown at 7^{o} C (cold stress) or in the presence of 20% (w/v) polyethylene glycol 4000 (osmotic stress) for 12h and 72h. Cells were harvested and microsomal fractions prepared from the challenged cells as well as control cells. Activity of ß–glucosidase (EC 3.2.1.21) increased up to 65% in response to 12h cold stress. Conversely, the activities of succinate dehydrogenase (EC 1.3.99.1), and acetolactate synthase (EC. 4.1.3.18) decreased, indicating that the activities of these two enzymes were down-regulated in response to low temperature. In the osmotic stress study, ß–glucosidase activity increased in a manner similar to the low temperature challenge, implying that this enzyme may play an important role in the adaptation of plants to environmental stresses. These studies may contribute to a better understanding of the adaptation mechanisms of plants to abiotic stresses, allowing us to exploit these mechanisms in a manner that may ultimately lead to hardier crops.

H2.3 Changes in gene expression in microspore-derived cell suspension cultures of *Brassica napus* exposed to low temperature or osmotic stress.

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Analysis of differential gene expression in plant cultures exposed to abiotic stress may reveal genes that may be useful in understanding stress tolerance in crops. Suppression subtractive hybridization (SSH) was used to examine gene expression in oil-forming microspore-derived cell suspension cultures of *Brassica napus* L. cv Jet Neuf subjected to low temperature or osmotic stress. In the low temperature studies, the cells were acclimated at 5° C for 12h and 72h prior to RNA extraction and subsequent cDNA library construction. Polyethylene glycol (PEG)-4000 (20% w/v) was used to apply osmotic stress for the same time periods prior to RNA extraction. SSH cDNA libraries for 12h and 72h low temperature (up-regulation), 12h osmotic stress (down-regulation) and 72h osmotic stress (up-regulation) were constructed and approximately 5000 colonies were isolated for each library. Random

sequencing of DNA inserts from 96 colonies from the 12h low temperature forward library (i.e., up-regulated in response to low temperature stress) identified 36 sequences with a relatively low level of redundancy (21%) among the independently picked clones. Twenty-two of these genes were also identified in the low-temperature response of winter wheat (*Triticum aestivum*) determined through a separate study. cDNA sequences encoding proteins involved in signal transduction, carbohydrate metabolism, cell cycle regulation, regulation of gene expression and protein synthesis were among the up-regulated transcripts represented in the low temperature acclimation SSH cDNA library. These results help identify the numerous metabolic pathways in the cell that are either affected by, or responsive to, low temperature, and may provide several starting points to further investigate and exploit these pathways in *B. napus* in the future. Sequencing results from the other SSH libraries are currently being analyzed and will also be reported.

H2.4 Characterization of low temperature regulated genes using microarray analysis and real-time PCR in winter wheat (*Triticum aestivum*).

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Winter survival is a key limitation to winter wheat production in western Canada. The most important factors reducing winter wheat survival are low temperatures and snow mould injury. Resistance to these stresses are induced by low temperatures that occur naturally during the fall and early winter. Approximately 9,200 distinct amplicons derived from cDNA libraries made using different low-temperature hardened wheat and rye genotypes were printed on the ECORC microarray slides. Among these elements are a selection of 450 putative transcription and translation factors, and other DNA binding elements. These arrays were probed with short-term cold hardened RNA from three winter wheat cultivars PI178383 and CI14106 (snow mould resistant, freezing susceptible), and DH+ (snow mould resistant, freezing tolerant). Additionally, expression of low temperature-regulated candidate genes, including a glycine-rich sequence and different sugar-transporter sequences, were evaluated using real-time PCR. Differential regulation was observed among the different sugar-transporter contigs evaluated. Results of expression profiling will be discussed in relation to resistance to snow moulds and freezing temperatures among the different wheat lines employed in the study.

H2.5 Relationship between genetic polymorphism and cold tolerance in alfalfa

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Recurrent cycles of selection were performed within the alfalfa (*Medicago sativa* L.) cultivar Apica to generate populations potentially more tolerant to freezing (TF). A bulk segregant analysis (BSA) approach was subsequently used to identify genetic polymorphisms associated to cold adaptation. Pooled DNA extracts from ~45 genotypes from each of the original population (TF0) and two derived populations (TF2 and TF5) were hybridized to candidate genes known to be associated with cold adaptation. Two striking polymorphisms that showed opposite responses to selection pressure were uncovered for genes encoding putatives dehydrin (intensification) and lipid transfer protein (disappearance). This result indicates that improvement of freezing tolerance does not solely rely on the increase of favorable genes but also requires the elimination of unfavorable ones. Genetic variability for freezing tolerance was determined within the cultivar Apica (TF0) and the two TF-derived populations following their acclimation to natural winter conditions. Freezing tolerance of the above mentioned 45 genotypes was determined as the ratio of average regrowth of six clonal propagules exposed to a freezing stress (-12^{o}C) over the average regrowth of six unstressed propagules (regrowth ratio from 0 to 1). The frequency of genotypes that maintained superior regrowth after freezing (ratio > 0.75) markedly increased from TF0 to

TF5. Each genotype was concomittantly scored for the presence or absence of candidate genes that are polymorphic among TF populations in order to establish their relationship with freezing tolerance. This analysis revealed a two-fold increase in the frequency of the polymorphic dehydrin homolog in cold tolerant (24%) than in the cold sensitive (12%) genotypes. Our results illustrate that BSA in combination with unique genetic material can efficiently uncover genetic polymorphisms associated with cold tolerance. The observations of polymorphic changes with genes of unknown function or not typically related to stress tolerance can provide valuable evidence to unravel new gene functions.

H2.6 Winter Rye Glucanases Play a Dual Role in the Cold

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Extracellular protein complexes containing chitinases and glucanases are expressed at cold temperatures in winter rye (*Secale cereale* L.) and display antifreeze activity (AFA). The AFA catalyzes the modification of ice crystals growth in the apoplast at subzero temperatures. As a first step toward the determination of the influence of each individual protein complex component on the AFA, we cloned, characterized and expressed the cold-induced glucanases from winter rye. Eight different cDNAs were isolated from a cold-acclimated winter rye cDNA library by probing with a barley ß-1,3-glucanase cDNA. Seven of the cDNAs, including M1GLU and M2GLU, had high amino acid identities to basic ß-1,3-glucanases of other cereals, whereas the sequence of A1GLU was similar to acidic ß-1,3-glucanase and ß-1,3-1,4-glucanases isolated from barley, rice, and wheat. Demonstrating a potential multifunctional activity, the glucanases were specifically expressed under cold, drought, and ethylene treatments. When they were expressed in *E. coli* and purified, M1GLU and M2GLU both catalysed the hydrolysis of laminarin and modified the growth of the ice. A1GLU exhibited not only ß-1,3-glucanase and ß-1,3-1,4-glucanase activities when laminarin and ß-D-glucan were used as substrates, but also a low level of AFA. Hypothetical modeling of M2GLU binding to the surface of ice showed a potential ice-binding domain on the surface of M2GLU. The recombinant ß-1,3-glucanases remained hydrolytically active at subzero temperatures. We conclude that glucanases alone are able to modify the ice crystals independently from the other protein components found in the apoplast. Thus, winter rye glucanases provide plants with a dual protective mechanism against pathogenic organisms such as snow molds, and also prevent the formation of large ice crystals in the apolpast, which would be lethal.

H3. Mycology

H3.1 Characterization of fungal root endophytes allied to the ericoid mycobiont *Rhizoscyphus ericae* (= *Hymenoscyphus ericae*) and to the *Piceirhiza bicolorata* ectomycorrhizal morphotype

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Sterile fungi isolated from surface-sterilized roots of the Ericaceae were hypothesized to be conspecific based primarily on restriction fragment length polymorphisms and provisionally named as Variable White Taxon (VWT). Additional isolates of culturally similar fungi obtained from roots of the *Orchidaceae*, *Pinaceae, Betulaceae* and *Salicaceae*, and given informal names such as Sterile white 1 (SW1), were suspected to represent the same taxon. To evaluate conspecificity and infer phylogenetic affinities, partial nuclear ribosomal DNA sequences were determined. Parsimony analyses supported a species level distinction for VWT/SW1 and indicated that the fungus is placed within the *"Hymenoscyphus ericae* aggregate," a species complex including the well-known ericoid mycobiont *H. ericae* (*Leotiomycetes*), recently renamed as *Rhizoscyphus ericae*, several unnamed taxa and *Cadophora finlandica.* We describe a new genus and species for isolates of VWT/SW1. This root-associated fungus can be identified by morphological characters in conjunction with ITS sequence data. Its mycorrhizal status is not yet clear. ITS analyses indicate that the "*H. ericae* aggregate" includes several other well-supported clades. Two additional species

are described for isolates that include ectomycorrhizal mycobionts of the "*Piceirhiza bicolorata*" morphotype.

H3.2 Arthropod dispersal of cycloheximide resistant fungi in central Alberta.

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Keratin-rich debris supports a wide range of saprophytic fungi and some species, such as those in the Onygenales, have a special predilection for these substrates. In contrast to many saprophytes, the Onygenales are not major components of the airborne mycota and ostensibly rely on arthropods to locate new substrate and to disperse their propagules. We modified techniques designed to assess air borne fungi to survey the fungi being carried by arthropods in a boreal forest ecosystem, and to look for evidence of arthropod dispersal of the Onygenales. Traps baited with substrates rich in either keratin (coyote dung) or cellulose (moose dung, brown rotted wood, and white rotted wood) were used to capture arthropods from a field site in a wooded area approximately 100 km west of Edmonton. Fungi were isolated from these carriers using agar media amended with cycloheximide, an antibiotic that selectively inhibits a wide range of fast growing fungi but allows the growth of the Onygenales and several other taxa. 1696 arthropods representing 13 orders were collected and fungi representing 53 species in 9 orders were recovered. The Hypocreales were the most abundant (627 isolates and 11 species). The Mycosphaerellales (538 isolates in four species) and Eurotiales (341 isolates in seven species) were also well represented. The Onygenales were represented by 57 isolates, but accounted for 11 species. Statistical analysis of the data showed that the keratin-rich substrates attracted significantly more arthropods than did the cellulose-rich substrates, and that dung attracted arthropods with a different suite of fungi compared to the arthropods attracted to plant-based substrates.

H3.3 First report of the teleomorph of *Colletotrichum truncatum*.

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Anthracnose of lentil, caused by *Colletotrichum truncatum* ((Schw.) Andrus and Moore), is a serious threat to lentil (*Lens culinaris* Medik.) grown in western Canada. The teleomorph stage of this pathogen was induced to form under laboratory conditions. Seven single-conidium isolates were incubated alone and in nine pair-wise combinations. Three of the pairings produced fertile perithecia, but no perithecia formed when the isolates were incubated separately suggesting that they may be self-sterile. Perithecia were brown-black, erumpent, solitary or in small groups, obpyriform to ampulliform, 180-(250)-350 µm x 90-(150)-210 µm. Asci were cylindrical, narrowing slightly at the apex, unitunicate, evanescent, 58-(75)-93 µm x 5-7 µm, and contain 8 ascospores. Ascospores are hyaline, aseptate, oblong, 8-(10)-15 µm x 3-[4]-5 µm. The ascospores were smaller than many other *Glomerella* species described in the literature.

All three fertile pairings consisted of isolates of the two physiological races that occur in western Canada, designated CT0 and CT1. The possibility of sexual recombination between these two races has profound implications for disease resistance breeding in lentil. The occurrence of the teleomorph under field conditions, however, still needs to be determined. We hope that the discovery of the teleomorph of *C. truncatum* from lentil can be used to resolve questions about the relatedness of this pathogen to *C. truncatum* from other hosts as well as to facilitate genetic studies of virulence in lentil isolates.

H4. Conifer genomics and herbivory

H4.1 The Arborea Project: A functional genomics approach to identifying regulators of wood formation in conifers

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Wood formation is a dynamic process that responds to numerous developmental and environmental cues. These cues, in turn, modulate both the quantity and properties of the wood produced during a growing season. As part of the Genome Canada-funded project *Arborea*, we are using a functional genomics approach to identify and characterize regulators that govern morphological and biochemical properties of secondary xylem, i.e. wood, in white spruce (*Picea glauca* [Moench] Voss). The cornerstone of this approach is a suite of candidate genes that we hypothesize encode regulators of processes important to wood formation such as carbon partitioning and meristem function, based on the known functions of their predicted homologues in herbaceous plants. Most of these candidate genes belong to multigene families, including the MYB, LIM, HD-ZIPIII and KNOX families of transcription factors. cDNAs encoding putative members of these families have been cloned from white spruce, and phylogenetic and gene expression approaches are being used to elucidate the function of the candidate genes.

To complement these studies of the endogenous white spruce genes, transgenic spruce trees have been generated that misexpress the putative loblolly pine (*Pinus taeda* L.) homologue of each of the candidate genes. To date, over 10000 transgenic spruce plantlets have been transferred to the greenhouse, and analyses of these trees are underway. Histochemical analyses of several of these transgenic lines have revealed marked phenotypic effects, particularly on tissue organization and cell wall structure. To complement the macrophenotypic analyses, transcript profiling of these transgenic lines using an *Arborea* custom-produced 9K cDNA microarray is in progress. The molecular phenotype revealed by these microarray analyses will play an integral role in delineating the function of these candidate genes in wood formation.

H4.2 Investigation of the role of genes encoding LIM-only proteins in poplar wood formation

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With the aim of investigation of genetic determinants related to wood formation, we isolated three putative LIM domain transcription factors from poplar xylem cDNA libraries. These genes encode LIM-only proteins (LMO) and show high levels of similarities with Ntlim1, a transcriptional regulator of some lignin biosynthetic genes in tobacco. These proteins are characterized by the duplication of LIM domains defined by conserved patterns of cysteine and histidine and consisting of two specialized zinc fingers. We will present results from investigation of the potential role of these LMOs during wood formation in poplar using two approaches:
1- Expression patterns analysis of LMO genes in different tissues and conditions culture using real-time RT-PCR and cDNA microarrays. First studies revealed a reduction in the expression level of these candidate genes in tension wood, an "abnormal" tissue formed in leaning angiosperms trees associated with stem eccentricity and tree axis reorientation. Tension wood is characterized by distinctive fibres that have a particular poorly lignified secondary cell wall layer that is rich in crystalline cellulose and is referred to as G-layer. Surprisingly, plants fertilized with a high quantity of nitrogen seem to produce xylem with G-fibres similar to leaning trees. To lead to a better understanding of the implication of LMO genes in those two biological processes, we conducted an experiment combining tension wood formation and nitrogen hyper-fertilization. We are using this study to simultaneously identify the transcript abundance for 3,400 *Populus* genes between tension wood, fertilized wood and normal wood using spotted cDNA microarrays.
2- Localization of the LMO proteins in tobacco cells using chimeric genes constructed between the LMO proteins and Green Fluorescent Protein (GFP) under the control of the 35S promoter. The aim of these experiments is to establish whether the three poplar LMO proteins that we have isolated are nuclear.

H4.3 Genomics discovery of lodgepole pine defense mechanisms against the mountain pine beetle-vectored fungal pathogen (*Ophiostoma clavigerum)* and evaluation of

a new conifer microarray chip for gene expression profiling across the pine family.

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The current mountain pine beetle (*Dendroctonus ponderosa*) epidemic is the largest outbreak of a bark beetle in recorded history affecting millions of hectares of lodgepole pine (*Pinus contorta*) in British Columbia and Alberta. In addition to the bark beetle itself, the beetle-vectored blue stain fungi (e.g. *Ophiostoma clavigerum*) contribute much to tree mortality. Both bark beetles and fungi trigger a number of tree defense responses. The tree's defense mechanisms include biosynthesis of terpenoids and phenolics, some of which can be deterrent or toxic to the bark beetle and fungus. A number of genes involved in terpenoid and phenolic defenses have been identified in other conifer species and some of these genes have been shown to be inducible by insect attack or treatment of trees with the plant defense elicitor methyl jasmonate (MeJA). However, to our knowledge, no lodgepole pine defense-related genes and their involvement in response to MPB/fungus attack have been characterized. In order to advance our knowledge of lodgepole pine defense responses we are using a recently developed 16.7k cDNA microarray for large-scale gene expression profiling in lodgepole pine trees inoculated with *Ophiostoma clavigerum* or treated with MeJA. Initially, we demonstrated that a 16.7k spruce cDNA array chip can be used successfully for gene expression profiling across closely and distantly related members of the pine family (Pinaceae), including species of spruce, pine and firs. Microarray gene expression profiling in lodgepole pine revealed a large number of differentially expressed genes in response to fungal inoculation and in response to MeJA treatment. Differentially expressed genes include genes involved in phenolic and terpenoid biosynthesis, as well as genes involved in defense signaling. These microarray results provided a set of target genes and pathways for further characterization of their role in lodgepole pine defense mechanisms against blue stain fungal pathogen.

H4.4 Exploring large-scale transcriptional responses in hybrid poplar to herbivory by forest tent caterpillars: Genome organization, cDNA cloning and gene expression of a Kunitz trypsin inhibitor family

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Plants respond to insect herbivory using a combination of direct (e.g. protease inhibitors, condensed tannins, alkaloids etc.) and indirect mechanisms (e.g. phenolic and terpenoid volatiles). To understand the inventory of defense mechanisms in poplar and how these are regulated, we utilized a microarray containing ~15,000 unique cDNAs to examine the transcript profiles of leaves from *Populus trichocarpa x deltoides* trees exposed to continuous feeding by the defoliating insect *Malacosoma disstria* (forest tent caterpillar), compared to leaves treated with mechanical wounding, *M. disstria* regurgitant, or methyl jasmonate, as well as untreated control leaves. Among the set of genes responsive to these stress treatments, we identified 29 array elements with high similarity to the Kunitz family of protease inhibitors (PIs), the majority of which were rapidly and strongly induced. PIs accumulate to high levels in many plants in response to wounding and are posited to inhibit digestive enzymes in the insect's gut, thereby decreasing the nutritional content of the plant tissue. Real-time PCR was applied to 12 of these transcripts, confirming induction of up to 100-fold in response to stress, and also providing constitutive patterns of expression across tissue types. A search of 86,000 poplar ESTs derived from 15 cDNA libraries, combined with targeted sequencing of selected clones identified more than 60 full-length cDNAs from four *Populus* genotypes. Using these full-length cDNAs to search the poplar genome sequence indicates the Kunitz PI family consists of at least 29 unique loci, the majority of which are organized into five gene clusters within the genome. The characteristics of a large gene family, organized within the genome as gene clusters, that is also rapidly inducible in response to insect herbivory may offer adaptive advantages for poplar trees in the constant evolutionary "arms race" between plants and insect pests.

H4.5 Traumatic resinosis in Sitka spruce – a resistance mechanism against attack by the white pine weevil?

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The white pine weevil (*Pissodes strobi*, Peck) is a serious pest of Sitka and interior spruce in British Columbia, Canada. This pest damages the terminal leader causing growth loss and stem deformities that severely impact tree value. Thus far there are no effective methods to control the white pine weevil. As a result, selection and breeding for weevil resistant genotypes holds promise to minimize weevil damage associated with reforestation programs.

In spruce, weevil attack induces a traumatic resinosis response that is characterized by the *de novo* synthesis of oleoresin that is sequestered in newly formed resin ducts. Oleoresin is a complex mixture of monoterpenes, sesquiterpenes, and diterpene resin acids that provide chemical and physical protection against herbivores and pathogens. To determine whether traumatic resinosis is an important determinant of resistance we have examined this response in Sitka spruce genotypes that are resistant or susceptible to weevil attack. To achieve this we have undertaken molecular, histological and chemical approaches to examine 1) the expression of genes that encode enzymes responsible for terpenoid formation (terpene synthases TPS), 2) the formation of traumatic resin ducts (TRD) in the xylem, and 3) changes in resin profiles. We found that mechanical wounding and weevil attack increased TPS expression coincident with the formation of TRD. The level of altered TPS expression was greater in resistance genotypes than in susceptible ones for some TPS genes indicating the induced resinosis response is important for resistance to weevil attack.

H5. Pathogen resistance mechanisms

H5.1 A proteomic analysis of resistance to *Erysiphe pisi* in mutants of *Pisum sativum* × *P. fulvum*

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Genetic resistance to *Erysiphe pisi* is believed to be a one- or two-gene-controlled recessive trait. However, proteomic information about regulation and expression of resistance in *E. pisi* is not available. In our studies, several mutant plants susceptible to *E. pisi* were found in an F1 population developed from a cross of two resistant genotypes, "Tara", a commercial cultivar of *Pisum sativum*, and a wild line "JI 595935" (*Pisum fulvum*), where all progeny were expected to be resistant. This mutation provides a tool for the study of resistance mechanisms. HPLC, electrophoresis and LC-MAMA were used to detect the resistance-related proteins in leaf extracts. The protein profiles of F1 leaf tissues from normal resistant plants and from susceptible mutants were compared using one-dimensional and two-dimensional SDS-PAGE. In the 1-D gel profile, a significant additional band at 51 kDa was identified as a ribulose-bisphosphate carboxylase large chain precursor. Approximately 200 individual protein spots were detected on the 2-D gel profiles, and at least 11 of these were different in the proteome of resistant plants compared to susceptible mutants. Some of these proteins were identified by mass spectrometry and database searching as ß-1,3-glucanase, cysteine proteinase, ABA-responsive protein, profucosidase, and endochitinase. Qualitative differences were also found between the leaf extracts of mutant and wild individuals. Results are discussed in terms of the possible mechanisms of expression and regulation of the gene(s) for resistance to *Erysiphe pisi* in pea plant defense systems.

H5.2 Altered expression of proteins caused by Fusarium head blight infection in spikes of wheat cultivars with various levels of resistance

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Fusarium head blight (FHB) of wheat causes severe economic losses in many of the wheat-growing regions of the world. Although wheat resistance against FHB has been identified and used in breeding FHB resistant cultivars, limited knowledge is available about the mechanism of fungal infection, colonization, and spread within spikes of wheat. A proteomic approach was used to investigate the expression of fungal proteins and the alteration of protein expression in the FHB infected spikes of resistant cultivar, Wangshuibai, and a susceptible cultivar, Crystal, respectively. Flowering spikelets of Wangshuibai and Crystal were inoculated with conidiospores of *Fusarium graminearum* and H_2O. Proteins were isolated from spikelets harvested 24, 48, and 72 h after inoculation. Two-dimensional gel electrophoresis and a fluorescent staining were used to display, quantitatively measure, and compare expression levels of proteins in spikes inoculated with *F. graminearum* and H_2O, respectively. Although most protein maintains same expression level between FHB and H_2O inoculated spikelets 24, 48, and 72 h post inoculation, there were proteins detected to be down-regulated, up-regulated, and newly induced by FHB infection. All these proteins were excised for identification using LC-MS/MS analysis. Proteins originating from the wheat and Fusarium genomes were identified. A function could be assigned to most polypeptides of wheat and about half of the Fusarium ones. Among the wheat ones, kinases, germin, serpin, translationally controlled tumor protein, peroxiredoxin, invertase and nucleic acid- binding proteins were observed. Superoxide dismutase, mannitol dehydrogenase and glyceraldehydes were some of the identified proteins with known functions from Fusarium. Significant differences were detected between the expression of the identified plant and fungal proteins between FHB susceptible and resistant cultivars. Discussions, based on the potential functions of these proteins, about their possible roles in the process of FHB initial infection and resistance will be presented.

H5.3 Characterization of phenolic compounds in two pine species resistant to the European race of *Gremmeniella abietina*, the causal agent of scleroderris canker

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Recently, large amounts of suberin and lignin were revealed within anatomical barriers in *Pinus contorta* and *P. banksiana*, two species resistant to the European race of *Gremmeniella abietina*. In the present study, cytochemical tests were carried out to characterize other phenols involved in resistance. Examination of tissues after toluidine blue staining or under blue excitation revealed an increase in polyphenols during the infection process. In the transition zone, phenolic compounds were present in numerous globules within phloem cells while in the healthy zone they appeared in large vacuoles filling these cells. In control shoots, only the periplasmic area of the parenchyma cells contained phenols. Among the phenols that increased in transition or healthy zones of infected shoots, condensed tannins and proanthocyanidins detected by the DMCA (dimethylaminocinnamaldehyde) test, and catechins and condensed tannins observed with the vanillin method were noticeable. When observed under UV light, tissue stained with the Neu's reagent revealed flavonoids as a bright orange fluorescence, particularly in the cortex region very close to the periderm or following the ligno-suberized barrier that was formed in the transition zone. In the non-infected tissues, much less flavonoids were detected. The intense whitish green fluorescence observed in infected tissues with the Neu's reagent indicated that caffeic acid could play a role in resistance while the deep blue fluorescence revealing the presence of gallic acid and gallocatechin derivatives in both infected and non-infected tissues suggested that the role of these compounds in defense is more difficult to determine. This study revealed that multiple phenolic constituents produced in specific areas in *P. banksiana* and *P. contorta* probably are important defense mechanisms after challenge by the European race of *G. abietina*.

H5.4 High throughput analysis of gene expression in snow mold resistant winter wheat using microarray analysis.

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Resistance to snow moulds and low temperatures reduce winter survival and yield of winter wheat (*Triticum aestivum* L.) and many other crops in western Canada. Disease resistance and freezing tolerance both increase when winter wheat is exposed to low-temperature hardening conditions that occur during the fall and early winter. The oligo microarray developed by the Functional Genomics of Abiotic Stress Project contains over 18,000 wheat sequences spotted in duplicate and was used as a high through-put technique to identify low temperature-responsive genes that are involved in the over-wintering process in winter wheat. The 'spotted' probes were targeted with mRNA from PI181268 (snow mould resistant, freezing susceptible) or Norstar (snow mould susceptible, freezing tolerant) winter wheat sampled prior to hardening and hardened at 2°C for various periods of time (6h, 12h, 24h, 7d, 21d). The expression profiles of the 'spotted' wheat sequences at different stages of cold hardening at 2ºC and between PI181268 and Norstar, were compared using the varying intensities of the different dye labelled targets. Information gained from these microarray experiments will be used to identify candidate genes specifically involved in low temperature-induced resistance to diseases and freezing temperatures. These results will permit a better understanding on cold hardening-induced disease resistance and its relationship to freezing tolerance in winter wheat.

H6. Crop management and disease

H6.1 Control of Sclerotinia Head Rot in Sunflower.

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Head rot caused by *Sclerotinia sclerotiorum* (Lib.) de Bary is widely spread in sunflower (*Helianthus annuus* L.) growing areas in North America. This disease devastated the Manitoba sunflower crop in 2004 with up to 80% incidence resulting in heavy yield losses. In the absence of genetic resistance in commercial sunflower hybrids, nine fungicides were applied to the heads at early flowering and/or late flowering in a randomized complete block field trial with four replicates, to study the effects of fungicides in reducing the head rot severity, and avoiding the heavy losses in yield and quality of seed. The sunflower heads were artificially inoculated with *S. sclerotiorum* 24 hr after each fungicide application, and the trial was misted for 5 mn every 30 mn for two weeks after inoculation. Disease incidence and severity were assessed 10-day intervals starting after the first application until harvest. The cool and wet conditions throughout the 2004 growing season were ideal for disease development resulting in 77% disease incidence and a disease index of 7.3 in the control treatment. The fungicides JAU6476™, Benlate®, and Lance™, significantly (P = 0.05) reduced disease incidence to 52-57%, and diseases index to 4.5-6.1. Ronilan™ and Quadris™ reduced disease incidence to 60-63% and disease index to 5.8-6.9. Fluazinam™, Maxim™ and Topsin™ reduced disease incidence to 60-70% and disease index to 5.4 -7. All fungicides resulted in significant (P = 0.05) yield improvement and reduction in the percentage of sclerotia in harvested seed.

H6.2 Impact of four types of lime on potato scab incidence in eight potato soils.

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We reported previously that addition of lime (98% $CaCO_3$; 4.6 t/ha) can cause a dramatic increase in potato scab [*Streptomyces scabiei*] in site GM soil where the pH was increased by 2 units. Here, we report on the impact of four types of lime (calcitic (37% calcium), dolomitic (22% calcium and 11.5% magnesium), HaveLock Mag (31% calcium and 5% magnesium), and Hydra-Lime+ (10% hydrated lime, 10% gypsum, and 80% magnesium limestone) from Graymont, Havelock, NB) on scab in eight potato soils, including soil from site GM. The soils (pH 5.4 to 7.1) were collected from commercial potato fields in Ontario, Prince Edward Island, and New Brunswick, lime added at 2.3 t/ha in all soils and at 7 t/ha in some soils, placed in micro-plots, potato cultivar Snowden planted, and the resulting scab severity determined. None of the lime treatments caused a significant increase

in scab severity in any of the soils compared to the control treatments. In four soils (including GM), all lime treatments caused a slight increase in scab disease. Hydra-Lime+ consistently caused the least amount of increase compared to the other limes, and in one soil caused a significant decrease in scab severity. Doubling the amount of lime added did not increase scab severity more that the 2.3 t/ha rate. None of the lime treatments had any significant effect on tuber yield except for the high rate of calcitic lime in one soil which reduced yield by half. Addition of lime initially increased soil pH between 0.2 and 0.8 units which remained more or less the same for the whole growing season. Thus, addition of these limes to the soils tested here at 2.3 and 7 t/ha rates appears to be safe with respect to not significantly increasing scab. One reason why these limes did not significantly increase scab may be because they did not cause a large increase in soil pH like $CaCO_3$ can. These results do not mean that an increase in soil pH directly causes an increase in scab but just that it is correlated.

H6.3 Effect of crop rotation on severity of red root rot and grain yield in corn

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Red root rot (RRR) is a disease of corn (*Zea mays*) newly reported from Québec and Ontario. This disease causes a dark discoloration of roots, ranging from red to dark brown, and a reduction in total root mass. Symptoms can be detected as early as 5 weeks after sowing and progress until the end of the growing season. RRR is caused by a complex of soil pathogens including *Phoma terrestris* as the main component, *Pythium* spp., and *Fusarium* spp. The impact of various crop rotations, including soybean, wheat, and corn, on RRR development and on grain yield of corn was investigated under conventional and reduced tillage on a loamy soil at Saint-Hyacinthe, Québec. Disease severity of RRR on roots was assessed visually (0-9 scale) early and late in the growing season in each of five years. For each year of observation, two or more consecutive years of corn increased RRR symptoms compared to other crop rotations. One break year of soybean or wheat between two years of corn was enough to reduce severity of RRR. Symptoms were higher under reduced tillage than under conventional tillage. Corn yield was correlated with symptoms. Grain yield of corn, when following two break years of either wheat and/or soybean, was 8% higher than yield of any back-to-back corn. Such yield increase ranged from 4 to 15% with years and tillage systems. The occurrence of RRR in all plots suggests that actual yield losses in growers' fields are unsuspected. Results also suggest that growing corn back-to-back is unsuitable whereas RRR causes more yield losses under such condition. More research is required on this damaging disease since it could become an important concern in corn production in Canada as is the case in some areas of USA.

H6.4 Effect of fish emulsion pre-plant soil amendment on common scab of potato and tuber yield in eight commercial potato field soils

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Fish emulsion or liquid fish concentrate is mainly used as foliar fertilizer for crop production. However, we recently demonstrated that it is also effective for control of damping-off diseases. Here, we report on the efficacy of fish emulsion as a pre-plant soil amendment to control common scab [*Streptomyces scabiei*] of potato in 2004 microplot and field studies. Potato soils of different soil pH (5.2-6.9) and characteristics from eight commercial fields in Ontario, New Brunswick, and Prince Edward Island with a scab history were used. Fish emulsion (1% vol/mass soil or 20,000 L/ha) was incorporated into naturally-infested soils and Snowden tubers planted into them 2 weeks later. Fish emulsion treatments received no additional nitrogen or potassium fertilizers in microplots but received the usual fertilizer regime used by the growers in each field location. In microplots, pre-plant soil application of fish emulsion significantly reduced scab severity by 57%-90% in six of eight soils and increased total tuber yield by 53%-145% in four soils compared to the untreated controls. In one of two field locations, fish emulsion pre-plant soil amendment significantly reduced scab severity and increased the percentage of disease-free tubers. Total tuber yield was not affected at

either field location possibly because the added nitrogen from fish emulsion was not adjusted for. Although effective broadcast rates of fish emulsion may not be economical, it is possible that banding or furrow applications can lower costs without compromising efficacy. This study clearly demonstrated that fish emulsion as a pre-plant soil amendment can reduce potato scab severity in a wide range of soils.

J1. Agronomy

J1.1 Forage yield and quality of traditional forage legumes under acid soil conditions

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Vast tracts of land in the foothills area of the Eastern Slopes of the Canadian Rockies and the Prince George area of BC have acid soils. Economic viability of ranches located in these areas depends on choice of appropriate forage legume crops and cultivars. Two long term studies planted in the foothills area of South West Alberta indicated that alfalfa has better ability than other forages to with stand low soil pH (5.5 - 6.0) and other adverse conditions of this area. Among the alfalfa cultivars, AC Blue J, Algonquin, Apica, Nordica and Spredor III produced high forage yield and quality. Red clover and bird's foot trefoil produced high quality forage but produced low yield and only survived for two years. The two non-bloat causing forages, sainfoin and cicer milkvetch, produced lower yield than alfalfa. Forage quality of sainfoin was lower than alfalfa but cicer milkvetch produced very high quality forage. Forages harvested from the plots were fed to livestock without any apparent adverse effect. This long term study under acid soil conditions allowed us to select acid tolerant germplasm for the forage legumes which will be used for development of acid tolerant cultivars.

J1.2 Measuring phyllochrons in barley to use for seeding date recommendations.

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In earlier work on seeding dates of barley in central Alberta, we found that those cultivars with short phyllochrons, independent of maturity, had the least yield reduction under late seeding conditions (early June versus early May). As a follow-up to that work, phyllochrons of seven barley cultivars, recently developed and released by the Field Crop Development Centre, were assessed under controlled environmental conditions (16/8 h $20/15^{o}$C at approximately 450 µ-mol m^{-2} sec^{-1}). Leaf counts of the main culm were made three times a week from coleoptile emergence to flag leaf fully emerged. Phyllochrons were determined as the inverse of the regression of leaf number against accumulated growing degree days (GDD; 0^{o}C basis). Linear regressions were used to fit the data and daylength was not considered to be an interacting factor (days were not lengthening or shortening). Excellent fits were found for all cultivars with r-square values of 0.98 or 0.99. Phyllochrons ranging from short to long were: 72 GDD $leaf^{-1}$ for Tyto; 74 GDD $leaf^{-1}$ for Trochu; 77 GDD $leaf^{-1}$ for Kasota; 81 GDD $leaf^{-1}$ for Vivar; 86 GDD $leaf^{-1}$ for Niobe; 87 GDD $leaf^{-1}$ for Manny; and 92 GDD $leaf^{-1}$ for Ponoka. Final leaf numbers of the main culm were: 8 leaves for Manny; 8.8 leaves for Kasota; 9.0 leaves for Vivar; 9.5 leaves for Ponoka; 9.8 leaves for Niobe; 9.9 leaves for Trochu; and 11 leaves for Tyto. Final leaf numbers and phyllochrons can be used to predict relative rates of development. Based on this work, development reflected known maturity differences for Kasota, Ponoka, Trochu, and Tyto. However this work resulted in an underestimation of maturity for Manny and Vivar and overestimation of that for Niobe; and this discrepancy may reflect an inherent problem in using linear regression to estimate phyllochrons for all cultivars. More work needs to be done to determine if the rapid leaf development of Tyto may be linked to competitive ability, especially with weeds. This work does provide us with the basis for a recommendation that under late seeding conditions, Tyto, Trochu and Kasota should be the cultivars of choice to minimize yield loss.

J1.3 Role of crop residue in mitigating frost damage in early sown canola

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Stubble impacts crop microclimate in ways that have not been well-investigated, particularly in the context of crop establishment, growth and yield. In this study, the effects of different stubble heights in mitigating frost damage of early-sown canola seedlings was examined in the black soil , sub-humid climate of southwest Manitoba. Canola was sown into wheat stubble 5, 15, or 45 cm in height in the Fall, early and late Spring. Plots were instrumented to monitor soil and canopy air temperature. Frost events (ambient air temperature at 100 cm < -5^{o} C) were recorded at different times during the spring after seedling emergence but before appearance of the 2nd true leaf. In some cases during these frost events the temperature at the soil surface fell below -5^{o} C in the very short stubble for up to four hours whereas in the tall stubble, surface soil temperature was rarely less than 1^{o} C. Plant counts on emerged seedlings were performed at three dates at 6 day intervals. Severity of frost damage was assessed based on a 5-point scale within 6 days of incidence of frost. Some level of frost damage occurred in all stubble treatments. Frost damage was uniform throughout all short stubble (5 cm) plots The highest occurrence of undamaged seedlings was found amongst tall stubble treatments where stubble was upright and uniform. Discussion focusses on the potential benefits of cereal residue in reducing the incidence of frost damage on young canola seedlings.

J1.4 Economic Impacts of Tillage, Phosphorus, and Preceding Crops on Flax

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Conservation farming is a system approach that considers all factors that affect production. Reduced tillage is a method of conservation farming, which impacts nutrient availability and fertilizer/chemical management decisions. Phosphorus (P) supply, and its availability in early growth stages, is critical for optimum crop yield. Producers frequently avoid P application in flax and increase the P supply in the preceding crops due to flax sensitivity to seed-placed monoammonium phosphate. The preceding crop supplies residual P and, depending upon it association with mycorrhizae, may also impact yield and performance of the highly mycorrhizal flax. By more clearly defining the P requirements of flax, canola and wheat grown under different management systems, it is possible to reduce inputs while maintaining or improving crop yield and quality. While many research studies have evaluated the economic impact of tillage systems on N fertility requirements, there has been very limited information available on the economic impact of tillage management and P phytoavailability or on the impact of the tillage system and past phosphorus fertilizer management on phosphorus response of crops. The objective of this study is to evaluate the economic impact of flax on tillage system, P fertilizer application, preceding crop, and level of P fertilizer applied in preceding crop. The study included canola and wheat as preceding crops grown under zero tillage (ZT) and conventional tillage (CT) with three levels of P fertilizer at two locations. Results indicated that net income was affected by preceding crops but not by P application and ZT system generated higher net income compare to CT system. Generally, there was lower net income variability associated with treatments under ZT and when wheat was preceding crop. Net income of treatments was higher when flax was grown after wheat than after canola, which may be explained by greater mycorrhizal formation after wheat than canola.

J1.5 Spatial Variations of Wild Blueberry Leaf Chlorophyll and Nitrogen Levels Using Hyperspectral Techniques

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Inconsistencies in wild blueberries growth and yield have led to increasingly intensive nutrient management practices that are based on time consuming, destructive, multi-procedurial and costly plant tissue analyses. Hyperspectral techniques may be an alternative to these procedures and provide additional inter-field nutrient and vegetative cover variability. A preliminary field experiment examining the feasibility of using hyperspectral techniques at plant, plot and small field scales for estimating temporal and geospatial

changes in N content was established at a commercial field in the sprout year at Mount Thom, NS in 2004. A three factor, central composite rotatable design was used with soil-applied nitrogen (0 to 80 kg N·ha-1), phosphorous (0 to 220 kg P_2O_5·ha^{-1}), and potassium (0 to 80 kg K_2O·ha^{-1}) applications consisting of ammonium sulfate, triple superphosphate, and muriate of potash respectfully. Leaf tissue nitrogen levels were measured using a LECO CNS-1000 elemental analyzer. The hyperspectral visible/near-infrared technique used in this experiment included a *compact airborne spectrographic imager* (CASI) at resolutions of 0.5 m, 0.8 m (5-10 m above ground) and 1.2 m (2800 ft above ground). Preliminary results indicate that the CASI was effective in detecting chlorophyll levels variations. CASI was also effective in estimating foliar N levels. Further investigations are needed to assess within field variability of total foliar P, K, Ca and Mg.

J2. Physiology and environment

J2.1 Fluorodetection of copper toxicity in sunflower plants

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UV-induced chlorophyll (ChlF) and blue-green (BGF) fluorescences emitted by leaves were proposed as useful indicators of the physiological state of plants and of their responses to environmental stress such as mineral deficiencies, pathogens, prolonged exposure to UV, etc. Considering the increasing occurrence of copper in ecosystems, it was important to also evaluate the potential of UV-induced fluorescences to detect the toxic effect of this heavy metal on plant growth. Sunflower plants were cultivated during a 11 day-period in a growth chamber in presence of normal (2 ?M) and toxic (75 ?M) concentrations of Cu^{2+} in the nutrient solution (Hoagland). During the experiment, biomass accumulation, CO_2 assimilation rates, chlorophyll and soluble polyphenols concentrations, and ChlF and BGF intensities were regularly measured. On the 2nd day of the experiment, lower biomass accumulation and lower root/shoot ratio were already apparent in plants treated with 75 ?M Cu^{2+} compared to control plants. On the 4th day and thereafter, Cu-inhibited plants showed lower CO_2 assimilation rates, lower chlorophyll concentration but higher concentration of soluble polyphenols compared to control plants. Concerning fluorescence intensities, it is noteworthy that only one day after the start of the experiment, it was possible to detect a decrease of UV-induced ChlF in Cu-inhibited plants. However, no difference in BGF from control and Cu-inhibited plants were observed throughout the experiment. Therefore, these results indicated that the more pronounced inhibitory effects of Cu^{2+} on plant growth relative to photosynthesis may increase carbon allocation toward polyphenol synthesis. The accumulation of these UV-absorbing compounds in the cell vacuoles of leaf epidermis will then decrease UV-excitation of chlorophylls in leaf mesophyll.

J2.2 Combined effects of ultraviolet-B radiation and carbon dioxide on *Brassica napus*

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Few studies have investigated the interaction of UV-B radiation and CO_2 concentration on plants. We studied the combined effects of UV-B radiation and CO_2 concentration on canola (*Brassica napus* cv. 46A65) under four growth conditions – elevated CO_2 with UV-B, ambient CO_2 with UV-B, elevated CO_2 without UV-B, and ambient CO_2 without UV-B (control) – to determine if the adverse effects of UV-B are mitigated by elevated CO_2. Elevated CO_2 significantly increased plant height and seed yield, whereas UV-B decreased them. Elevated CO_2 ameliorated the adverse effects of UV-B in plant height. UV-B did not affect the physical characteristics of leaf but CO_2 did. Certain flower and fruit characteristics were affected negatively by UV-B and positively by CO_2. UV-B did not affect net photosynthesis, transpiration and stomatal conductance but decreased water use efficiency. However, elevated CO_2 significantly increased net photosynthesis and water use efficiency and decreased stomatal conductance. Neither UV-B nor CO_2 affected chlorophyll fluorescence. UV-B significantly decreased chlorophyll *b* and increased the ratio of chlorophyll *a*/*b*. However, elevated CO_2 decreased only the ratio of chlorophyll *a*/*b*. UV-B significantly increased UV-absorbing compounds while CO_2 had no

effect on them. Both UV-B and CO_2 significantly increased epicuticular wax content. Many significant relationships were found between morphological, chemical and physiological parameters. This study showed that elevated CO_2 can ameliorate the adverse effects of UV-B radiation in *B. napus*.

J2.3 Carbon isotope discrimination as a selection criterion for improved water use efficiency and productivity of barley on the prairies

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This study was done to evaluate the application of carbon isotope (^{13}C) discrimination (?) as a selection criterion for improving water use efficiency (WUE) and productivity of barley on the Canadian prairies. Five genotypes (first experiment) and 10 genotypes (second experiment) were subjected to drought at the jointing stage to study the relationship between ?, WUE and barley productivity. Drought caused considerable reductions in aerial biomass and grain yield of all the genotypes examined in both experiments. Significant genotypic variation was found in WUE of the genotypes. Significant correlations were found between ?, and WUE as well as ? and aerial biomass and grain productivity, which highlight the potential of ? (leaves or seeds) as a rapid and reliable method for evaluating WUE and productivity of barley. Genotypes (Manny, Trochu and Seebe) with the highest WUE (low ?) under drought conditions showed performance comparable to the genotypic average under well-watered conditions. This suggests the potential for improving WUE under drought conditions without yield penalties when conditions are optimum. More research is needed to test this technique under field conditions and to establish a standard protocol that can be used to develop new, improved, water efficient barley varieties.

J2.4 Physiological responses and dry mass partitioning in three ecotypes of *Stellaria longipes* grown at elevated CO_2.

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Genets from three ecotypes of *Stellaria longipes*, originally collected from alpine tundra, prairie and sand dune habitats in Western Canada, were grown at ambient (365 ppm) and elevated (1000 ppm) atmospheric carbon dioxide (CO_2) for 21 days in solution culture. Photosynthesis (A) and transpiration (E) rates were measured in newly developed leaves. Water use efficiency (WUE) was calculated from the values of A and E. Dry mass partitioning was measured by determining leaf mass area (LMA), specific leaf area (SLA), leaf mass ratio (LMR) and leaf area ratio (LAR). Relative water content (RWC) of shoot was also measured. Elevated CO_2 significantly increased A and WUE but significantly decreased E. There were no significant differences between genets in A, E or WUE. Dry mass attributes were significantly different between genets within CO_2 concentrations, but elevated CO_2 did not affect LMA, SLA, LMR or LAR. Relative water content was also significantly different between genets within CO_2 concentrations but was not affected by elevated CO_2. In conclusion, physiological parameters were significantly affected by elevated CO_2 while dry mass attributes were significantly affected by genet.

J2.5 Study of cold acclimation and freezing tolerance in different species of Brassicaceae.

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Many temperate plants acquire freezing tolerance through an adaptive process called cold acclimation, in response to a period of low, non-freezing temperatures. Cold acclimation is associated with numerous physiological, biochemical and molecular changes. *Thlaspi arvense* exhibits a higher level of freezing tolerance than either of its close relatives, *Arabidopsis* or cultivated *Brassica napus*, making it a good model for studying the mechanisms of cold tolerance in crucifers. The aim of this study is to uncover the mechanisms conferring freezing tolerance in *T. arvense,* and to compare the low

temperature responses of *T. arvense* with those of *Arabidopsis* and *B. napus*.

The freezing response (LT_{50}) of excised leaves and the levels of leaf soluble sugars were examined during cold acclimation of *T. arvense* (3 ^{o}C day/night, 12h photoperiod, 150µE light). Freezing tolerance of *T. arvense* increased from -5^{o}C to -18.5^{o}C after four weeks of cold acclimation and the total soluble sugar level also increased during cold acclimation. Microarray analysis was performed to identify novel cold responsive genes and uncover signaling pathways active during cold acclimation of *T. arvense*. Approximately 400 cold responsive genes with greater than two fold changes over the non-acclimated control were identified. Bioinformatics has determined the putative function of these genes, which is allowing insights into the expression pathways which are being activated in the wild crucifer compared to the model plant. A few novel candidate genes have been selected for further characterization. The expression pattern of candidate genes during cold acclimation is being compared in *T. arvense, B. napus* and *Arabidopsis* using northern hybridization and RT-PCR. *Arabidopsis* lines with mutations in each of the candidate genes have been identified and over-expression lines are being generated to determine their role in the cold acclimation response.

J2.6 Comparative gene expression analysis during cold acclimation in winter and spring wheat.

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The very wide range of genetic variability for freezing tolerance among wheat cultivars make it an excellent species to characterize the genetic basis of freezing tolerance. Freezing tolerance develops during acclimation at prolonged above-freezing temperatures, thus it is a trait that is ideally suited to gene expression analysis.

In order to characterize the regulation of freezing tolerance in wheat a targeted cDNA amplicon microarray enriched for regulatory genes was used to compare changes in gene expression profiles during cold acclimation in winter and spring wheat cultivars during 14 days of growth at 4°C. The Genome Canada program, Functional Genomics of Abiotic Stress (FGAS), used virtual subtraction to enrich for cDNAs from genes with low abundance mRNA and thus enrich the EST set for regulatory genes. The 80,000 sequences EST data set was mined for clones with high similarity to protein kinases, protein phosphatases, RNA binding proteins, E3 ubiquitin ligases, GTP and calcium binding proteins, and 11 families of transcription factors. Approximately 1700 cDNAs candidate regulatory genes were selected and supplemented with 4000 random clones from FGAS and NSF wheat EST collections to create a 6500 cDNA-amplicon microarray. The comparison of gene expression profiles in the winter cultivar Claire and the spring cultivar Quantum showed a large number that the initial complex changes in gene expression were very similar between the two cultivars, but they differed markedly at later periods of cold acclimation. Over 300 signal transduction candidate genes were found to be significantly induced or repressed in response to cold treatment and 85 of these had different levels of induction or repression in winter and spring wheat.

J3. Tissue Culture

J3.1 Isolated microspore culture of Canadian 6x triticale cultivars.

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Production of haploid and double haploid plants is useful for not only breeding purposes but is also a potential tool for various biotechnological applications such as genetic engineering or *in vitro* selection for desirable traits. Many factors including cultivar, growth conditions and pre-treatment of donor material, and media composition can influence the frequency of embryogenesis in isolated microspore culture and subsequent production of double haploids. Two induction media developed for wheat; NPB99 and CHB3 with or without ficoll (100gl^{-1}) were tested on isolated microspores derived from 9 Canadian 6x triticale cultivars. Significant interactions were

observed for the number of embryoids and calluses induced, green and albino plantlets regenerated and fertility of green plants. Ficoll was beneficial in both media to increase numbers of embryoids and green plants. Overall, medium NPB99 supplemented with ficoll provided the most suitable condition for these cultivars, except AC Alta, which performed slightly better on CHB3 supplemented with ficoll. Only *cv.* Wapiti was not amenable to androgenesis. Cultivars AC Certa, AC Copia, AC Alta, Sandro, Ultima, Frank, Pronghorn and Banjo produced respectively 10.0, 9.3, 6.3, 5.2, 4.3, 2.7, 2.6 and 1.3 green plants per Petri dish (35,000 microspores). Twenty two percent of total lines produced were fertile, and considered doubled haploids. This study shows that isolated microspore culture of Canadian hexaploid triticale can be attractive tool for breeders and molecular biologists.

J3.2 Isolation, Culture and Genetic Engineering of Triticale Protoplasts

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The combination of characteristics inherited by Triticale from its parents wheat and rye make it an attractive target for a variety of biotechnological studies. Also, plant protoplasts are an interesting system for genetic engineering. Keeping this in view present study has been initiated with an ultimate goal of introducing various genes of interest in Triticale protoplasts. We have successfully isolated mesophyll protoplasts from seven day old *in vitro* grown plants of Triticale cultivar AC Alta. The leaves are incubated in an enzyme solution (combination of macerozyme and cellulase) for at least 4 hrs in dark at 28^0C. The isolated protoplasts are then purified on 17% sucrose gradient. Very high yield of protoplasts is obtained with 70-80% viability. The purified protoplasts are used further, either for culturing to obtain multicellular stage or for DNA uptake studies. The protoplast culture is carried out using agarose bead method supplemented with MS medium as a nutrient source. Also, we are investigating the capacity of Triticale protoplast for foreign DNA uptake with or without carrier molecule(s).Simultaneously, role of various cell membrane permeabilizing agents (such as saponin and toluene) on protoplasts and their aid in DNA uptake by the protoplasts is under investigation. The fluorescent DNA selection marker GFP has been employed as the gene of interest for the primary studies of DNA uptake by the protoplasts.

J3.3 Overcoming TDZ-induced inhibition of adventitious shoot elongation and developing a one-step *in vitro* cloning procedure using zeatin in strawberry.

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In an attempt to improve the shoot regeneration and elongation protocol for 'Bounty' strawberry (*Fragaria ananassa* Duch.), leaf, sepal and petiole explants were compared on two different media supplemented with varying levels of thidiazuron (TDZ) and N^6-benzyladenine (BA). Sepals proved more effective than the leaf discs and petiole segments for regenerating shoots. TDZ supported adventitious shoot regeneration at moderate concentrations (1 to 5 µM) but inhibited shoot elongation. However, usable shoots were obtained within 4 weeks by transferring shoot cluster to medium containing 2 to 4 µM zeatin. An *in vitro* cloning protocol that enables shoot proliferation and rooting of TDZ-induced adventitious shoots in the same medium was developed. Shoots proliferated and roots developed best when adventitious shoots were cultured in the medium supplemented with 2 to 4 µM zeatin. Subculturing improved the number of shoots and roots per responding explant, shoot height, root length, and root vigor. *In vitro*-derived plantlets were acclimatized and eventually established in the greenhouse.

J3.4 Genes involved in microspore embryogenesis in *Brassica napus* microspore culture identified by differential display using an *Arabidopsis* microarray.

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The *Brassica napus* microspore culture model is a convenient, well-characterized system for studying microspore embryogenesis in plants. Several recent studies using this system have implicated the involvement of a diversity of genes in early embryogenesis however an examination of the whole transcriptome of embryogenic microspores has yet to be reported. Taking advantage of the

genotypic similarity between *Arabidopsis thaliana* and *B. napus* spotted cDNA microarrays containing 11,960 *Arabidopsis* cDNA clones representing 6424 genes from HHMI at the Yale University School of Medicine were used with two cDNA populations derived from embryogenic microspores and pollen-like microspores from 3-day old cultures. Differentially expressed genes between the two populations were identified, the corresponding *B. napus* cDNAs cloned, and further characterized in whole culture and flow-sorted microspores using real-time PCR. Two candidates (DWF1 and vacuolar ATPase) were significantly ($p>0.05$) over-expressed in embryogenic cells. DWF1 is a gene involved in the conversion of 2,4 methylenecholesterol to campestrol in brassinosteriod synthesis. Its expression dropped markedly in the day 4 control cultures with respect to same period induced cultures and was 6-fold higher in 3-day sorted embryogenic cells compared to pollen-like cells. The vacuolar ATPase is a complex multi-subunit proton pump involved in the movement of protons into cellular vacuoles. Its expression was initially higher in control cultures declining across both conditions but reversed after 3-days where its expression was 2-fold greater in sorted embryogenic cells with respect to pollen-like cells. The DWF1 expression pattern suggests that the production of brassinosteriods is important for induction and/or maintenance of embryogenic identity where the expression of vacuolar ATPase suggests that alkalinization of the cytoplasm is important for induction of embryogenesis 3 days after culture.

J3.5 Expression of the plant CDK inhibitor ICK1 leads to cell death in *Arabidopsis* microspores and pollen

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The cell cycle in eukaryotic organisms is controlled by cyclin-dependent kinases (CDKs). ICK1 is the first plant CDK inhibitor that has been shown to inhibit plant CDK activities and cell divisions, and affect plant growth and development profoundly when overexpressed [1,2]. ICK1 possesses different functional domains for CDK inhibition, protein stability and nuclear localization [3]. Previously, it was observed that targeted tissue-specific expression of *ICK1* in transgenic *Brassica napus* plants resulted in pollen with decreased viability and often abnormal nuclear numbers [4]. To further understand the mechanism for the effects of *ICK1* expression on pollen development, *ICK1* was placed under the control of the tobacco *NTM19* promoter that can activate gene expression at early stages of pollen development. In transgenic *NTM19-ICK1 Arabidopsis* plants, the number of pollen per anther was reduced. Light microscopy revealed that in addition to a reduced number, microspores and pollen of the *NTM19-ICK1* plants were more variable in size, and often had a reduced number of nuclei. They appeared less dense in cellular contents, and many of them had large vacuoles and little cytoplasm. Further studies showed that compared to the control plants, a much higher proportion of the pollen nuclei from *NTM19-ICK1* plants were positive by TUNEL staining. These results demonstrate that targeted expression of *ICK1* resulted in the degeneration and cell death of microspores and pollen in *Arabidopsis* plants.

1 Wang H, Fowke LC, Crosby WL (1997) *Nature* 386: 451-452.

2 Wang H, Zhou Y, Gilmer S, Whitwill S, Fowke LC (2000) *Plant J* 24: 613-623

3 Zhou Y, Li G, Brandizzi F, Fowke LC, Wang H (2003) *Plant J* 35: 476-489

4 Zhou Y, Wang H, Gilmer S, Whitwill S, Keller W, Fowke LC (2002) *Planta* 215: 248-257.

J3.6 Alternation of Morphology and Cation Contents in Bamboo Xylem Mutant, *vse*, Derived from Somaclonal Variation

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A xylem mutant (*vse*) was isolated from a bamboo plantlet after it was subcultured for seven consecutive years. This mutant was proliferated in 0.1 mg/l thidiazuron (TDZ) and its roots induced with 5 mg/l NAA. Results indicated that the growth of mutant was retarded in greenhouse and several morphological abnormalities were observed. As compared with the wild type, the mutant had thinner stems with less trichromes on its surface. The microscopic image showed that the xylems were smaller in diameter and with crystal-like structures in the pith. The leaves were shorter and narrower with sharp leaf blade angle. The roots were thinner and contained fewer numbers of xylems. The cation concentrations of both mutant and wild type were similar *in vitro* analysis except for iron and potassium, which were lower in mutant leaves in vivo. Iron chlorosis was noted on young and potassium deficiency on old leaves. All of the wild type plants survived but the mutant plants had only 27 percent (16/60) survival rate after one year in the greenhouse.

J4. Seeds

J4.1 An ABA receptor and its docking protein: Potential involvement in ABA signal transduction related to seed dormancy.

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Despite the progress that has been made on abcisic acid (ABA) action and biosynthesis, our understanding of its perception is still obscure. We have shown earlier that barley ABAP1 protein binds ABA in a receptor like manner. ABAP1 possesses a WW protein interaction domain that is likely necessary for its function. Currently, we are characterizing an ABA-responsive protein (ABA45) that interacts with ABAP1, as determined by GST Pull-Down assays, via a consensus sequences known to interact with the WW domain. ABA45 is present in barley leaves, aleurones but it is more abundant in embryos. It is up-regulated by ABA and down-regulated by GA in barley aleurone tissues. We are studying the expression profile of the two proteins as a function of dormancy status and under different stress conditions. The level of ABAP1 transcripts in embryos is higher in dormant lines of barley .We propose that ABA45 is a membrane platform protein that interacts with the ABA receptor ABAP1 and in so doing initiates the activation of downstream components of the ABA signal transduction pathway.

J4.2 A model for seed dormancy regulation mediated by the ethylene receptors in *Arabidopsis thaliana*.

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Seed dormancy is a plant adaptive strategy for the conservation of future generations. It is defined as the inability of a viable seed to germinate under favourable conditions. The transition from dormancy to germination is therefor an extremely regulated metabolic process. Many factors have been implicated in the regulation of this process, perhaps the most extensively studied being the phytohormones abscisic acid and gibberellic acid. Ethylene has been implicated in seed dormancy and germination as various insensitive mutants show altered dormancy and germination patterns, however the exact mechanism of this involvement remains unclear. We have used such mutants for an evaluation of seed germination throughout seed development. Based on these results, we have constructed a hypothetical model for the mechanism of ethylene participation in this developmental process.

J4.3 Abscisic acid signaling networks in seed storage product accumulation in microspore-derived embryos of *Brassica napus* using a targeted cDNA microarray.

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We have designed and developed a custom cDNA microarray consisting of 696 *Arabidopsis thaliana* and *Brassica napus* clones as a diagnostic tool for studies of plant hormone

signaling and development. The cDNAs and genomic amplicons selected for this array are of sufficient size to make this a useful platform for cross-species hybridizations within closely related species of the *Brassicaceae*. In selecting cDNAs for this array we have targeted genes expressed in developing seeds, as well as genes involved in the synthesis, catabolism and signaling-response networks for each of the major plant hormones. This array has been used for 1) studies of abscisic acid (ABA) signaling networks involved in lipid and storage protein accumulation in seeds, 2) analysis of the efficacy of various ABA metabolites as substitutes for abscisic acid in seed-specific processes, and 3) investigation of early embryogenesis in *Brassica napus*.

Brassica napus microspore-derived embryos were treated with (+)-ABA for periods from 2 to 24 h, or deuterated ABA and ABA metabolites for 8h to study ABA signaling networks. The ABA time course microarray expression data revealed 191 significantly expressed genes by SAM (Significance Analysis of Microarrays). Differences in gene expression among ABA and ABA metabolites are being used to characterize the biological activities of these compounds.

J4.4 Proteomics of embryo and endosperm during seed germination in tomato

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The embryo and endosperm proteome analysis was conducted using one- and two-dimensional gel electrophoresis (1-DE & 2-DE) and mass spectrometry of germinating tomato seeds. The 1-DE profile was similar in both tissues but mobilization of the most abundant proteins occurred first in the endosperm. The 2-DE gels also showed similar protein spot pattern both in embryo and endosperm except with few quantitative and qualitative differences. Seventy five major spots of Coomassie blue stained 2-DE gels were selected, excised, in-gel digested with trypsin, and analyzed by matrix-assisted laser desorption/ionization-time of flight mass spectrometry (MALDI-TOF MS) and/or liquid chromatography-electrospray ionizationquadrupole/time of flight tandem mass spectrometry (LC-ESI- Q/TOF-MS/MS). Peptide MS and MS/MS data were searched against publicly available protein and expressed sequence tag (EST) databases, and 47 protein spots identified. Embryo-specific proteins included a BAC19.13 homologue whereas four spots specific to the endosperm were tomato mosaic virus coat proteins. The most abundant proteins both in the embryo and endosperm were seed storage proteins i.e., legumins (11spots), vicilins (11 spots), albumin (2 spots). Defense-related protein kinases, glycine-rich RNA binding proteins, actin binding profilin, , non-specific lipid transfer protein, and proteins involved in general metabolism were also identified both in embryo and endosperm. The role of some of the identified proteins in relation to seed germination in tomato is discussed.

J4.5 A soybean seed protein with carboxylate-binding activity

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The seed coat serves as a multifunctional organ with a role in protections as well as the supply of nutrients to the embryo sac during development. The composition of the legume seed coat differs from other seed tissues in many ways including its protein composition. We have purified and identified an abundant 24 kD protein (SC24) from soybean (*Glycine max* [L.] Merr) seed hulls. The corresponding cDNA and genomic DNA clones for SC24 were isolated and characterised, and expression patterns were determined under normal developmental condition as well as under carious stresses. The deduced protein sequence of 219 amino acids included an N-terminal signal peptide. Transcripts encoding SC24 were present in the seed coat from 30 days after pollination (DAP) until maturity, but the protein was not detected until the final stages of seed maturation. In mature seeds, most of the SC24 protein was localized to the parenchyma and aleurone layers of the seed coat and not in palisade and hour glass cells. The expression of *SC24* was also induced by wounding and pathogen infection. The SC24 protein bound to an affinity column containing an isophthalic acid ligand, and was eluted with 7 mM citrate. Polyclonal antibodies raised against recombinant SC24 cross-reacted with

the seed coat peroxidase enzyme, suggesting that these two proteins may share an antigenic determinant. To summarise, our results indicate that SC24 belongs to a novel class of plant defence proteins with carboxylate-binding activity.

J4.6 Isolation and Characterization of the Arginase Promoters in Loblolly pine (*Pinus taeda L.*) and *Arabidopsis*

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In conifers, the majority of the storage reserves that supply the developing seedling are located in the megagametophyte, a maternally derived haploid tissue that encloses the embryo. This tissue provides the seedling with the majority of the resources necessary for growth and development until it is photosynthetic. After the completion of germination, these reserves, composed of storage proteins and triacylglycerols, are hydrolyzed and rapidly transported to the cotyledons of the seedling where they are further metabolized.

One of the major amino acids released from the hydrolysis reactions is arginine. Arginine composes approximately 46% of the total stored nitrogen in storage proteins, and as such is believed to play a major role in providing the seedling with nitrogen. Once in the seedling, the arginine is hydrolyzed by the enzyme arginase to release ornithine and urea which are then utilized.

In order to achieve a better understanding of how this important developmentally controlled gene is regulated we have isolated the arginase promoter(s). Arginase was previously reported to be a single copy gene in Loblolly pine; however we now believe that a second arginase is present. Deletion series of both promoters have been constructed and transiently expressed in Loblolly protoplasts.

Arginase in *Arabidopsis* was also previously reported to be a single copy gene; however, a second arginase gene has recently been isolated. Knock-out mutants have been generated for each and enzyme activity assays show a low level constitutive arginase and a highly induced arginase that may be developmentally regulated. Again, in order to understand the mechanism of regulation of these two homologs we have completed deletion analysis of each of the promoters.

Poster Titles

P101. Transformation of *Brassica napus* with a cDNA encoding diacylglycerol acyltransferase-1 results in increased seed oil accumulation.

P102.The Toc159 family of chloroplast protein import receptors in *Arabidopsis thaliana*

P103. The Role of DET1 and Damaged DNA Binding Proteins (DDB1 and DDB2) in Arabidopsis DNA Repair and Light Signaling

P104. Sulfide-Resistant Respiration by Alternative Oxidase and the Potential for Sulfide Oxidation by Sulfide Quinone Oxidoreductase in Plant and Animal Mitochondria

P105. Mitochondrial Alternative Oxidase, Reactive Oxygen Species and Cell Death in Tobacco Leaves

P106. Root exudate proteomes of Arabidopsis thaliana and *Brassica napus*: An analysis of extracellular proteins by MudPIT and LC-MS/MS.

P107. Revealing the secrets of antifreeze protein promoters

P108. Plant responses to petroleum coke

P109. A Functional Genomics Analysis of the Phenylpropanoid Pathway in Arabidopsis

P110. Membrane-Based Heat Signaling And Hsf Activation In Tobacco Cells

P111. Inorganic carbon acquisition by the chrysophyte alga *Mallomonas papillosa*.

P112. Immunohistochemical localization of phospholipase D in strawberry (*Fragaria ananassa*

P113. Expression of heterologous *PREPHENATE DEHYDRATASE-LIKE* (*PDL*) of Arabidopsis for biochemical analysis.

P114. Expression and characterization of 7 abiotic stress inducible genes in Arabidopsis

P115. Engineering enhanced nitrogen use efficiency in rice (*Oryza sativa* L.)

P116. Developmental regulation of the GA biosynthesis genes, GA20ox, GA3ox, and GA2ox during germination and young seedling growth of pea (*Pisum sativum*) L.

P117. Programmed cell death and leaf morphogenesis in *Monstera obliqua* (Araceae).

P118. HBK2: A homeobox gene with potential for producing high quality somatic embryos in spruce.

P119. AUTOBAHN: a gene that has a role in auxin influx in Arabidopsis leaves

P120. Phenotypic characterization of a cambiumless mutant in *Arabidopsis thaliana*.

P121. Investigating phloem development and differentiation using Microarrays and In situ hybridization.

P122. ABI3/VP1 and the regulation of Arabidopsis and tomato (*Lycopersicon esculentum*) seed germination

P123. Microarray analysis of bast fibre gene expression in hemp

P124. Transcriptional responses to the xenobiotics 2,4-dinitrotoluene , 2,6-dinitrotoluene, and 1,3-dinitrobenzene

P125. Glutathione improves shoot apical meristem identity during white spruce (*Picea glauca*) somatic embryogenesis

P140. Impact of isolation on pollination of Tall-Grass Prairie plants.

P141. The effect of temperature and photoperiod on photosynthesis and the development of dormancy in contrasting ecotypes of dogwood (*Cornus sericea* L.)

P142. The study of Alpine plant Ferulago angulata (schlecht.)Boiss essential oil constituents extracted from seeds of two different habitats, Nevakoh and shahoo, Zagross mountain ,West of Iran.

P143. The Cypress Hills as habitat for *Crataegus*

P144. Study of Genetic Diversity Among Cotton Cultivars by Polyacrylamide Gel Electrophoresis.

P145. Stem elongation plasticity in the alpine and prairie ecotypes of *Stellaria longipes*: preliminary comparative studies with an α-expansin SlEXPA1 expressed in both the ecotypes.

P146. Phenology and biometry of 10 populations of barrel medic (*Medicago truncatula* Gaertn)

P147. Patterns in terrestrial bryophyte and lichen species in young and old sub-boreal spruce forest

P148. Aspects of Seed dispersal of the Gulf of Saint Lawrence Aster, *Symphyotrichum laurentianum*: disturb to preserve.

P150. Landscape pattern and persistence of grassland-forest ecotones:

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Poster abstracts

P101. Transformation of *Brassica napus* with a cDNA encoding diacylglycerol acyltransferase-1 results in increased seed oil accumulation.

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Diacylglycerol acyltransferase (DGAT, EC 2.3.1.20) catalyzes the acyl-CoA-dependent acylation of sn-1,2-diacylglycerol (DAG) to generate triacylglycerol (TAG) in the sn-glycerol-3-phosphate pathway. The level of DGAT activity in the developing seed may have a substantial effect on the flow of carbon into TAG. Therefore, the DGAT-catalyzed reaction represents a target for engineering oilseeds with enhanced seed oil content. *B. napus* cv. Westar was transformed with genetic constructs containing a cDNA encoding DGAT1 isoform driven by seed-specific napin (pD1) or cruciferin (pD2) promoters. Seed oil content analysis of T_2 and T_3 generation seeds revealed several transformants with increased seed oil content compared to Westar and/or control plants transformed with the null vector. The D1-2 family of transformants gave rise to several progeny with up to a 12% increase in seed oil content and up to four-fold increase in DGAT activity compared to Westar. This family of transformants also showed a reduced DAG:TAG ratio compared to controls, which is consistent with a more efficient conversion of DAG to TAG anticipated with elevated DGAT activity. The converse was true of lines such as D1-1 and D1-11, which showed an overall decrease in seed oil content and DGAT activity relative to controls. Field trials are currently underway to further investigate the most promising lines. As well, we are continuing this work using a double haploid breeding line of *B. napus*.

P102. The Toc159 family of chloroplast protein import receptors in *Arabidopsis thaliana*

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The Toc (translocon at the outer envelope membrane of chloroplasts) complex is of critical importance to plants, as it is responsible for recognizing and initiating the import of many essential plastid preproteins. This complex acts coordinately with the Tic (translocon at the inner envelope membrane of chloroplasts) complex to complete translocation of preproteins into the stroma. The core of the Toc complex consists of Toc159, Toc75 and Toc33. Toc75 forms at least part of the channel through the outer membrane, whereas Toc159 and Toc33 are related GTPases thought to be involved in preprotein recognition and regulation of import. In Arabidopsis thaliana, these GTPases are represented by small gene families. There are four Toc159 family members (Toc159, Toc132, Toc120 and Toc90) and two members of the Toc33 family (Toc34 and Toc33).

We have established that Toc159 interacts directly and specifically with transit peptides of photosynthetic preproteins destined for the chloroplast, through a binding site that is comprised at least in part by the GTPase domain of the receptor. In addition, we have demonstrated that Toc159 and Toc132/Toc120 differentially associate with Toc33 and Toc34, and therefore constitute structurally distinct Toc complexes. Toc132 and Toc120 also appear to specifically recognize and bind non-photosynthetic preproteins. These findings are consistent with the observation that Arabidopsis null mutants lacking one of Toc132 or Toc120 have no discernible phenotype, whereas double knockouts lacking both Toc132 and Toc120 are lethal. Collectively, these data have led to the hypothesis that members of the Toc159 family are transit peptide receptors that represent distinct routes for targeting preproteins to plastids, and that these separate pathways are required to ensure balanced import of proteins that are essential to the many biochemical pathways housed within plastids.

P103. The Role of DET1 and Damaged DNA Binding Proteins (DDB1 and DDB2) in Arabidopsis DNA Repair and Light Signaling

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During plant development, plants use light to decide the time and duration of each developmental stage. Around 7% of the total solar radiation is ultra violet (UV). UV affects plants by interrupting the physiological processes or by damaging the DNA which may lead to mutations in the plant genetic material. Reversal and excision of DNA damage are the two mechanisms used in plants to repair UV-induced DNA damage. In humans, proteins called damaged DNA binding protein 1 and 2 (DDB1 and DDB2) are implicated in the disease Xeroderma pigmentosa (XP). XP patients are at risk for skin cancer due to an inability to repair the UV-damaged DNA. DDB1 and DDB2 are found also in plants. Arabidopsis has two homologs of DDB1: DDB1A and DDB1B. DDB1A is found in a complex with another protein called DET1 which has a role in visible light signaling. We are interested in the effect of loss of function of DET1, DDB1A and DDB2 on the light signaling and DNA repair. In this study, different mutant genotypes of *Arabidopsis thaliana* were used: wildtype (Col-0), single mutant (*det1-1, ddb1a* and *ddb2*) and double mutants (*det1 ddb1a*, *det1 ddb2*, and *ddb1a ddb2*). The light regulated phenotypes (flowering time, rosette diameter, number of shoots, height and chlorophyll content) of these genotypes was investigated under long day visible light conditions. Results showed that there is no significant difference between wildtype, *ddb1a*, *ddb2* and *ddb1a ddb2* plants in all of the above mentioned parameters. *det1-1* plants exhibit early flowering and reduced plant height compared to wildtype. The double mutant *det1 ddb2* suppresses the *det1-1* early flowering phenotype in both number of days and number of leaves and enhances the short phenotype of *det1-1* plants. Currently, we are performing preliminary experiments to screen these genotypes for UV light (UV-C) sensitivity and DNA repair mechanisms. Further work will be done on these genotypes to study the interaction between light signaling and DNA repair.

P104. Sulfide-Resistant Respiration by Alternative Oxidase and the Potential for Sulfide Oxidation by Sulfide Quinone Oxidoreductase in Plant and Animal Mitochondria

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Besides cytochrome (cyt) oxidase, all plant mitochondria have a second terminal respiratory oxidase called alternative oxidase (AOX). Surprisingly, we have also found AOX genes in sequence data from seven different animal species representative of three phyla (Mollusca, Nematoda, Chordata), consistent with sporadic reports of CN-resistant respiration within species of these phyla. Hydrogen sulfide is a potent toxin, due to its reversible inhibition of cyt oxidase. However, using tobacco suspension cells and isolated mitochondria, we have established that AOX is fully resistant to sulfide. In addition, treatment of tobacco cells with sulfide results in a rapid induction of both AOX protein and activity. These results suggest that one role of AOX may be to provide a sulfide-resistant pathway of respiration. A wide taxonomic range of organisms (including bacteria, cyanobacteria, fungi and animals, but not plants) have been shown to possess a sulfide quinone oxidoreductase (SQR) that is able to oxidize sulfide and pass the electrons to the respiratory chain. Particularly in marine invertebrates, this has been previously described as an important detoxification mechanism. However, since the sensitivity of cyt oxidase to sulfide inhibition could hinder or disable this detoxification mechanism, it has been hypothesized that another oxidase is responsible for accepting electrons associated with sulfide oxidation. Interestingly, we have found that several plant genomes also encode SQR-like proteins similar to that described in other organisms. These proteins include the conserved FAD-binding domain, as well as candidate cysteine residues required for catalysis. We hypothesize that some plants and animals may use the combination of SQR and AOX as a system to tolerate and remove toxic levels of sulfide that can accumulate in tissues due to its prevalence in some environments (marine sediments, marshes, rice paddies) or due to metabolic activity in the mitochondrion (eg. ß-cyanoalanine synthase activity).

P105. Mitochondrial Alternative Oxidase, Reactive Oxygen Species and Cell Death in Tobacco Leaves

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In animals, the mitochondrion can play an active role in the signal integration and execution of programmed cell death (PCD). Recent research suggests that the mitochondrion may also play some role in plant PCD. However, plants harbor additional electron transport chain components that may alter the specific role of mitochondria in PCD. One such component is alternative oxidase (AOX), which catalyzes the oxidation of ubiquinol and reduction of oxygen to water. Previously, we established that antisense tobacco suspension cells lacking AOX exhibit increased susceptibility to PCD induced by several different treatments. To investigate this in planta, we used post-transcriptional gene silencing to generate tobacco plants lacking leaf AOX. Upon treatment with Complex III or IV inhibitors, transgenic (RI9) leaves showed extensive cell death, while no death was evident in wt leaves. However, the RI9 death could be blocked by an antioxidant (flavone), suggesting that the death is dependent upon elevated levels of reactive oxygen species (ROS). It further suggests that the protective effect afforded by AOX in wt leaves is not simply due to its ability to maintain energy metabolism. To investigate this further, in situ staining techniques and Northern analyses were used to compare ROS levels and ROS scavenging enzymes between the wt and RI9. In untreated plants, ROS levels were reduced in RI9 plants compared to wt, due to the up-regulation of scavenging enzymes. However, treatments always resulted in lower levels of O_2^- and higher levels of H_2O_2 in RI9 than wt, even regardless of whether the treatment caused differential death. However, those treatments that induced RI9 death had the highest levels of H_2O_2 relative to O_2^-, accompanied by reduced catalase gene expression. Our results suggest that the mitochondrial AOX has a significant impact on the cellular balance that exists between rates of ROS generation and the level of antioxidant defenses. A lack of AOX disrupts this balance, perhaps with implications for ROS-dependent PCD pathways.

P106. Root exudate proteomes of *Arabidopsis thaliana* and *Brassica napus*: An analysis of extracellular proteins by MudPIT and LC-MS/MS.

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The ability of plant roots to target a wide array of compounds to the rhizosphere results in a set of extracellular proteins that are strategically positioned to play a role in plant responses to biotic and abiotic stress. We have employed a sterile, hydroponic system and advances in the fields of proteomics and bioinformatics to collect and identify root exudate proteomes from two species, *Arabidopsis thaliana* and *Brassica napus*. The *B. napus* proteins were first separated by 2D-PAGE, isolated spots were then analyzed using liquid chromatography coupled to tandem mass spectrometry (LC/MS/MS) followed by de novo peptide sequence determination. A total of 16 *B. napus* spots were identified. Multidimensional Protein Identification Technology (MudPIT; on-line 2D LC/MS/MS) was used to identify 52 A. thaliana proteins. Bioinformatic analysis, including an effort to determine signal peptides and the presence/absence of transmembrane domains was completed for these extracellular proteins. Functional classification grouped the proteins into various families. An on-line Extracellular Plant Proteins Database (EPPdb) is being developed to provide information about this project and the identified extracellular proteins. We have also modified our hydroponic system to collect proteins exuded from *B. napus* roots under phosphate-deficient and aluminum-stressed conditions. The proteins from each of these treatments will be compared to control conditions using 2D-PAGE to identify proteins of interest. This work is a first step towards analysis of the extracellular proteomes exuded by roots under nominal and stress conditions.

P107. Revealing the secrets of antifreeze protein promoters

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Many of the antifreeze proteins in winter rye (*Secale cereale* L.) have both antifreeze and pathogenesis-related activities. The antifreeze activity of these proteins is only exhibited after a period of cold acclimation. To examine the possibility that this difference is due to differential expression of specific genes, the genes for several winter rye chitinases and glucanases with and without antifreeze activity will be sequenced along with their 5′ promoter regions. This is being done using using nested-PCRs and genomic walking techniques. Promoters will be analyzed for the presence of cis-acting regulatory elements using bioinformatic tools. To date, the genes and promoter of one winter rye ß-1,3-glucanase designated M2GLU with antifreeze activity has been fully sequenced and analyzed for cis-acting regulatory elements. Sequencing of another antifreeze protein gene coding for a ß-1,4-glucanase designated M1GLU, is nearly complete. Preliminary analysis indicates the promoters contain an abundance of putative cis-acting regulatory elements involved in light regulation, cold and dehydration tolerance, and some elements involved in hormonal regulation. The ultimate goal of this project is to compare to the elements in the promoter region of antifreeze proteins to those of pathogenesis-related non-antifreeze proteins in order to infer differential expression patterns. The gene and promoter sequences of these genes, along with the gene and promoter regions of related grasses will be used in subsequent phylogenic analysis in an attempt to find evolutionary links between antifreeze proteins.

P108. Plant responses to petroleum coke

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A greenhouse experiment was designed to study the effects of petroleum coke on plants.

Petroleum coke, a by-product of the oil sand industry in Alberta (Syncrude Canada Ltd. and Suncor Energy Inc.), contains relatively high concentrations of arsenic, boron, iron, nickel, sulphur, titanium, and vanadium embedded in a carbon matrix. Through weathering and biological degradation, these components may be released from coke and absorbed by plant tissues where they may inhibit growth and interfere with physiological activities. The low macronutrient content and physical properties of coke may also reduce plant growth. Tufted hairgrass (*Deschampsia caespitose*) and wheat (*Triticum aestivum*) were grown in peat (control), Syncrude coke, Suncor coke, Syncrude coke capped with 3 cm of peat, and Suncor coke capped with 3 cm of peat. Germination, plant growth, pigment analysis, and element content were determined over a 2 and 3 months period for wheat and hairgrass respectively. Hairgrass treated with Syncrude coke showed a decreased germination after 14 days whereas wheat showed no differences between treatments. Decreased height and shoot biomass were observed in all coke treated plants while a reduction in root biomass was only observed in coke treated hairgrass. Wheat and hairgrass showed a decrease in chlorophyll a, chlorophyll b, and carotenoid content in all coke treated plants. Roots of wheat grown in Syncrude coke and hairgrass grown in Syncrude and Suncor coke showed significant increases in nickel concentration at levels which may be toxic to plants. Wheat roots grown in Syncrude coke and all uncapped coke treated hairgrass roots showed increased accumulation of vanadium at levels which may also pose a phytotoxicity risk. These results suggest that petroleum coke has an inhibitory effect on plant growth and physiology which is likely due to nutrient deficiency, drought stress and potentially ionic toxicity.

P109. A Functional Genomics Analysis of the Phenylpropanoid Pathway in Arabidopsis

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The identification of rate limiting steps in metabolic pathways is a prerequisite for designing strategies to modify plant metabolism through genetic engineering. To identify target genes in the phenylpropanoid pathway, we carried out chemical analyses on a collection of Arabidopsis mutant lines with knockouts in genes affecting metabolic steps leading to the synthesis of sinapoylcholine

(sinapine) from phenylalanine. Chemical analysis of seed phenolic extracts from homozygous lines of these mutants revealed that genes affecting later steps in the pathway caused significant alterations in the phenylpropanoid profile. These results provide insight into critical steps in phenylpropanopid biosynthesis, and suggest strategies that could be used to modify the profiles of phenylpropanoids in related crops.

P110. Membrane-Based Heat Signaling And Hsf Activation In Tobacco Cells

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BY2 tobacco cells were grown at 25C in liquid suspension cultures according to published procedures. Cells from 7-day old cultures, which were growing rapidly, were used in all experiments reported here. De novo heat shock protein synthesis was monitored by labeling with ^{35}S-[Methionine] after 3-hour heat shock (37°C). Heat shock proteins synthesized at 3 hours of heat shock were visualized autoradiography. We found that HSP101, HSP90, HSP70 and several small HSPs were synthesized and accumulated to substantial levels. Heat shock factor-1 (HSF1) and heat shock factor 2 (HSF2) increased to maximum levels at 3h of heat shock but declined thereafter. HSF1 was selected for further analysis. We have previously demonstrated a role for membrane fluidity in heat signaling. HSF1 expression was inhibited at 37C by the membrane rigidifier DMSO, microfilament stabilizer jasplakinolide (JK) and the microtubule stabilizer taxol (TX). However, HSF1 expression was caused at 25C by treating cells with the membrane fluidizer, benzyl alcohol (BA), microfilament destabilizer latrunculin B (Lat B) and the microtubule destabilizer oryzalin (Ory). Furthermore, the heat shock induced expression of phosphorylated HSF1 and HSF2 was inhibited by the inhibitors of MEK, a MAPKK.

These observations show that heat shock factor (HSF) expression is regulated by heat-triggered membrane fluidization and requires the activity of a heat shock activated MAPK (HAMK) and the activity of MEK-related MAP kinase kinase (MAPKK).

P111. Inorganic carbon acquisition by the chrysophyte alga *Mallomonas papillosa*.

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It is now widely established that a number of microalgae, and all cyanobacteria, express CO_2 concentrating mechanisms which consists in most species examined of the active uptake of either, or both, bicarbonate and CO_2. Little is known about the mechanism of inorganic carbon uptake of chrysophyte algae. Photosynthetic characteristics of the chrysophyte alga *Mallomonas papillosa* were investigated to determine whether this species has some form of CO_2 concentrating mechanism. The effect of external pH on the photosynthetic oxygen evolution rate of air-grown cells demonstrated an optimum in the range pH 5.0 to 7.0. This species lacked external carbonic anhydrase, the cells has no capacity for direct bicarbonate uptake and had a low affinity for dissolved inorganic carbon. Measurement of the fluxes of CO_2 and O_2 in photosynthesizing cells at pH 7.0 by mass spectrometry, displayed no rapid uptake but only a slow depletion of CO_2 from the medium upon illumination. Furthermore, CO_2 uptake and O_2 evolution by Mallomonas was greatly reduced by iodoacetamide, an inhibitor of CO_2 fixation. The overall internal pH of Mallomonas was determined by distribution between the cells and medium, of ^{14}C-benzoic acid over the pH range 5.5 to 6.0, and [2-^{14}C]-5,5-dimethyloxazolidine-2,4-dione over the pH range 6.5 to 7.0. As the external pH was lowered from 7.0 to 5.5, there was a decrease in the internal pH of Mallomonas cells, from 8.31 to 7.75. The ΔpH was great enough to allow the accumulation of inorganic carbon by the diffusive uptake of CO_2.

P112. Immunohistochemical localization of phospholipase D in strawberry (Fragaria ananassa Duch.) fruits

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Sub-cellular localization of Phospholipase D (PLD), the key enzyme involved in membrane phospholipid catabolism, was conducted in strawberry (*Fragaria ananassa*. Duch) fruits during development using immunohistochemical techniques. Tissue segments of strawberry fruits from green expanding, mature white and ripening stages were fixed, processed and incubated with polyclonal PLD antibodies and gold-labelled double antibodies and examined using a transmission electron microscope. At the green expanding stage, the cells are much smaller and reveal typical ultrastructural features that include cell wall, plasma membrane, nucleus, mitochondria and the vacuole, while very few gold particles are detectable at this stage. At the mature white stage the cell wall region became a predominant structural feature. The sub-cellular distribution of PLD in the membrane and the cytoplasm increased as shown by the increased gold labelling with the IgG-gold conjugate. The cell wall region appears to be swollen further and the cytoplasm appears as a thin structure surrounding the vacuole in the ripening fruits. Increased gold labeling can be observed at the ripening stage and gold particles are especially visible in the vacuole and the cell wall region. At the ripening stage, several microvesicular structures can be seen near the plasma membrane, which may originate from the damaged regions of the membrane and be released into the cytoplasm to preserve the integrity of the membrane. The patterns of PLD localization may indicate the important role of PLD in strawberry fruit development and ripening.

P113. Expression of heterologous *PREPHENATE DEHYDRATASE-LIKE* (*PDL*) of Arabidopsis for biochemical analysis.

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The aromatic amino acid, phenylalanine is a product of the shikimate pathway. In plants, phenylalanine is thought to be synthesized from prephenate via a two-step reaction, with prephenate aminotransferase catalyzing the first step and arogenate dehydratase (ADT) the second. Most bacteria use the classical pathway for phenylalanine biosynthesis in which the enzymes prephenate dehydratase (PDT) and phenylpyruvate aminotransferase catalyze the first and second steps, respectively. Six genes from Arabidopsis thaliana have been identified based on their high sequence similarity to bacterial prephenate dehydratase genes. Due to this sequence similarity, the Arabidopsis genes were named *PREPHENATE DEHYDRATASE-LIKE* (*PDL*). However, based on the biochemical evidence for phenylalanine biosynthesis in plants, we would expect that these genes encode arogenate dehydratases. Both PDTs and ADTs have very similar catalytic functions but are proposed to use different, although structurally similar substrates. Hence it is possible that both enzymes share a high degree of amino acid sequence similarity and/or have the ability to use more than one substrate. To test these hypotheses, we are planning to use a combined molecular and biochemical approach to determine the enzymatic specificity of the six PDLs. Previous attempts to express the Arabidopsis PDLs in E. coli have been unsuccessful, indicating that the expression of these proteins might be toxic to this organism. Therefore the current study tests if a yeast expression system can be used as an effective alternative to express the plant PDLs. Characterization of the purified proteins will determine whether we have identified the first genes in plants known to encode arogenate or prephenate dehydratases and will provide further evidence as to which pathway is used for the synthesis of phenylalanine in Arabidopsis.

P114. Expression and characterization of 7 abiotic stress inducible genes in Arabidopsis

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Abiotic stresses including extremes of temperature and water availability are major factors that determine the natural geographical distribution of plants and limit the agricultural production of crops. Microarray profiling of the model plant Arabidopsis identified hundreds of cold, drought, ABA, mannitol and salinity inducible genes, of which the function of many is unknown. Among the stress inducible genes, RD29A/COR78/LTI78, RD29B/LTI65, KIN1, KIN2 and ERD10/LTI29/LTI45, which all encode proteins similar to those of Group 2

late embryogenesis abundant (LEA) proteins, and two functionally unknown, At2g24100 and At1g780709 containing a WD-40 repeat were chosen for further study. Previous study showed that RD29A/LTI/COR78 displays the greatest induction under abiotic stresses and is induced in roots greater than 250-fold for cold, 40-fold for mannitol, and 57-fold for NaCl. The responses of the former 5 genes to cold, drought, salt and ABA are different. To further characterize these 7 genes during abiotic stress tolerance, first, drought and stress tests as well as root elongation analysis under salt stress of RD29A and RD29B T-DNA insertion SALK lines were conducted, which showed no significant difference from wild-type Arabidopsis seedlings, indicating the functional redundancy of many LEA proteins exist. For protein purification and protein-protein interaction analysis, a hydrophilic 8-amino-acid FLAG-epitope tag (DYKDDDDK) was introduced to the N-terminal of these 7 genes under the drive of CaMV35S promoter and transformed into wild-type Arabidopsis with over 50 T_2 lines obtained for each construct and about 30 lines screened by PCR and RT-PCR for each. PCR showed that all 7 genes were integrated into genome in more than 90% of hygromycin resistant lines, while RT-PCR demonstrated that in more than 80% of the screened lines, FLAG-tagged fusion genes are transcribed correctly. However, Western blotting analysis of 5 genes failed to detect the expression of corresponding fusion proteins, with cy3-conjugated M2 anti-FLAG antibody showing cross reactivity with a ~110 kDa protein from Arabidopsis. We propose that the expression levels of those 5 FLAG-tagged fusion proteins may be too low to be detected or post-translational modification(PTM) may render the tag to be undetected, or even the N-termini are removed after PTM. Continuous analysis is still under way.

P115. Engineering enhanced nitrogen use efficiency in rice (Oryza sativa L.)

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Nitrogen (N) is the most important factor limiting crop productivity worldwide and therefore, improvement of nitrogen uptake, translocation and assimilation has been a long term goal in agriculture research. Isolation and characterization of the genes/promoters involved in nitrogen assimilation and their efficient expression in target tissue via genetic transformation technique provide tools for new strategies to improve nitrogen use efficiency. With the aim of improving nitrogen use efficiency, genetically modified rice (Oryza sativa L.) plants that over express alanine aminotransferase (AlaAT) were engineered by introducing a barley AlaAT cDNA driven by a rice root specific promoter into rice. AlaAT is known to be induced when plants are subjected to drought stress or recovering from nitrogen stress. Following the successful construction of desired transformation vectors, an improved Agrobacterium tumefaciens-mediated transformation protocol was developed for generating the large number of transgenic plants that are required for functional analysis of desired gene of interest. The developed protocol enabled the generation of a large number of transgenic plants in two cultivars within a short period, thereby reducing time and labour input. Currently, the transgenic plants are being analyzed to see if they display a nutrient use efficient phenotype under drought/nitrogen-deficient conditions.

P116. Developmental regulation of the GA biosynthesis genes, GA20ox, GA3ox, and GA2ox during germination and young seedling growth of pea (Pisum sativum) L.

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To understand the role of gibberellins (GAs) during germination and early post-germinative stages of large-seeded dicotyledonous plants, we profiled the expression pattern of genes encoding three regulatory GA biosynthesis enzymes (PsGA20ox1, PsGA3ox1 and PsGA2ox1) in pea (*Pisum sativum* L.) using real-time RT-PCR. To broaden our inferences on the role of GAs in these processes, we compared the GA biosynthesis gene expression patterns in two distinctly different genotypes of pea ('Alaska' a model cultivar for vining pea containing the wild-type internode length gene LE and 'Carneval' a model cultivar for semi-leafless field pea containing the le-1 mutation producing shorter internodes), both of which germinate readily on imbibition under normal environmental conditions. Residual amounts of PsGA20ox1, PsGA3ox1, and

PsGA2ox1 transcripts were detected in the mature embryos (0 days after imbibition; DAI) of both genotypes. Transcription of PsGA20ox1, PsGA3ox1, and PsGA2ox1 mRNAs occurred in all tissues examined (cotyledons, embryo axis, shoots and roots from 0.5 to 6 DAI) and was developmentally regulated within each tissue. Cotyledonary GA biosynthesis gene transcript patterns suggest that a signal from the axis triggers GA biosynthesis in the cotyledon. The high levels of PsGA20ox1 and PsGA3ox1 mRNA in the embryonic axis at 1 DAI suggests that the embryo axis is a major site for GA biosynthesis for stimulation of axis expansion. GA biosynthesis gene expression in 2 to 6 DAI shoots and roots (when their growth in fresh weight and length increased linearly) indicates a key role for de novo GA biosynthesis in early growth of seedlings. Supported in part by NSERC grant #138166.

P117. Programmed cell death and leaf morphogenesis in *Monstera obliqua* (Araceae).

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The unusual perforations in the leaf blades of *Monstera obliqua* (Araceae) arise through programmed cell death early in leaf development. At each perforation site, a discrete subpopulation of cells undergoes programmed cell death simultaneously, while neighboring protoderm and ground meristem cells are unaffected. Nuclei of cells within the perforation site become terminal deoxynucleotidyl transferase-mediated dUTP nick end labeling (TUNEL)-positive, indicating that DNA cleavage is an early event. Gel electrophoresis indicates that DNA cleavage is random and does not result in bands that represent multiples of internucleosomal units. Ultrastructural analysis of cells at the same stage reveals misshapen, densely stained nuclei with condensed chromatin, disrupted vacuoles, and condensed cytoplasm. Cell walls within the perforation site remain intact, although a small disk of dying tissue becomes detached from neighboring healthy tissues as the leaf expands and stretches the minute perforation. Exposed ground meristem cells at the rim of the perforation differentiate as epidermal cells. The cell biology of perforation formation in *Monstera* resembles that in the aquatic plant *Aponogeton madagascariensis* (Aponogetonaceae; Gunawardena et al. 2004), but the absence of cell wall degradation and the simultaneous execution of programmed cell death throughout the perforation site reflect the convergent evolution of this distinct mode of leaf morphogenesis in these distantly related plants.

P118. HBK2: A homeobox gene with potential for producing high quality somatic embryos in spruce.

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Three homeobox genes HBK1, HBK2 and HBK3, similar to the KNOTTED1-like Homeobox (KNOX) genes, have previously been found in Norway spruce (*Picea abies* (L.) Karst.). Although HBK1 and HBK3 are expressed in both embryogenic and non-embryogenic lines, HBK2 is only expressed in embryogenic lines. In spruce only a partial sequence of HBK2 is available. By screening cDNA libraries from white spruce (*Picea glauca* (Moench) Voss) it is the goal of this study to first find the complete sequence of HBK2, then to determine whether HBK2 is required for proper embryo development. RNA in-situ hybridization studies were used to determine gene localization within the embryo. RT-PCR studies were carried out using embryos at different maturation stages to determine when and in what abundance HBK2 is expressed. Transformation studies will be carried out to characterize gene function and to determine whether the embryogenic potential of white spruce lines can be improved.

P119. AUTOBAHN: a gene that has a role in auxin influx in Arabidopsis leaves

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We have characterized a mutation in the *Arabidopsis thaliana* gene named *AUTOBAHN* (*ABN*). *abn* leaves produce leaves proliferate disorganized, overlapping veins that are parallel to the midvein. They show no differentiation of higher order veins. abn floral organs have similar defects in vascular patterning but stems and roots do not show

vascular defects, thus ABN seems to be specific to foliar organs. Auxin is critical for vascular patterning: auxin canalization is proposed to cause files of cells to accumulate higher auxin levels and develop into veins. Thus, the response of cells to auxin and transport of auxin are critical to establish proper cell fate.

To examine auxin response in abn plants, we created an abn line that carries the auxin-inducible DR5:GUS construct. Analysis of the GUS expression pattern in wild type leaves reveals two distinct classes of expression: 1) primary, at the distal tip and, 2) secondary, along the margin, in hydathodes and predicting higher order veins. *abn* leaves show no normal aspects of the secondary response. Additionally, abn double mutants were generated with three classes of mutants: 1) the auxin-response mutants *monopteros*, *bodenlos*, *axr6*, and *fkd*-1, 2) the auxin-resistant mutants *axr1*, axr2 and *axr4* and, 3) the auxin over-producer *rty*. The additive phenotypes of all double mutants suggest that ABN functions independently of these auxin response pathways.
To establish if defects in auxin transport could explain the abn phenotype, abn seedlings were 1) grown on NOA and NPA, auxin influx and efflux inhibitors respectively and, 2) exposed to exogenous NAA and 2,4-D, synthetic auxins that bypasses the influx and efflux carriers respectively. Abn was also analyzed in combination with *aux1*-7 and *pin1*-1, mutants defective in influx and efflux respectively. The results from these experiments have led to the hypothesis that abn is defective in carrier-mediated auxin influx.

P120. Phenotypic characterization of a cambiumless mutant in Arabidopsis thaliana.

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Wood is formed by the meristematic activity of the vascular cambium, a cylindrical meristem located between the xylem and phloem. Little is known about the regulation of the vascular cambium and there are no previous reports of mutants defective in its development. We have identified an Arabidopsis mutant that lacks a cambium and does not undergo secondary growth. This mutant is tentatively called cambiumless and we postulate that the CAMBIUMLESS protein is a positive regulator of vascular cambium development. The phenotypic alterations in this mutant are not restricted to the cambium, it also has an extremely reduced plant size, the leaves are long and thin with involute margins, leaves senesce quickly, the plant flowers much earlier than wild type, floral organs fail to abscise, and both unfused carpels and sepaloid carpels are observed in later flowers. To determine whether the small size of the plant organs were due to decreases in cell size or cell number, scanning electron microscopy was used to analyse the leaves, and the mutant was found to have both smaller and fewer cells than wildtype. At this point we are uncertain of the common function that links all these phenotypes and are therefore anxiously awaiting the identification of the gene by map-based cloning.

P121. Investigating phloem development and differentiation using Microarrays and In situ hybridization.

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Until recently, most research on plant vascular tissue development has focused on xylem. As a result there is little information on the phloem of higher plants. In order to investigate which genes are expressed during phloem development and differentiation, RNA from phloem tissue of Arabidopsis plants that have undergone secondary growth was analyzed using whole-genome microarrays. Candidate genes from the microarray experiments are being investigated using available reverse genetics resources and in situ hybridizations. We are complementing this gene discovery approach with a forward mutant screen using GFP-tagged Arabidopsis lines.

P122. ABI3/VP1 and the regulation of Arabidopsis and tomato (*Lycopersicon esculentum*) seed germination

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The ABSCISIC ACID INSENSITIVE3/VIVIPAROUS1 (ABI3/VP1) transcription factor is a positively acting component of abscisic acid signal transduction

in a variety of seeds. During late seed development ABI3 promotes embryo dormancy, suppressing precocious germination. Whether ABI3 also acts to regulate germination has not been clearly established. The expression pattern of ABI3 was therefore examined in relation to Arabidopsis and tomato (Lycopersicon esculentum) seed germination. A decline in AtABI3 expression in Arabidopsis seed occurs after germination, in a nearly identical manner to that of RGL2. Expression of the tomato ABI3 orthologue (LeABI3) in wild-type embryos peaks at 24 hours after imbibition (HAI) and declines beyond detection following the completion of germination. A similar post-germination decline in LeABI3 expression was observed in GA-deficient gib-1 embryos stimulated to complete germination with GA, while LeABI3 transcripts are maintained in the embryos of non-germinating gib-1 seed on water. Isolated axes from gib-1 seed can germinate in GA or water. The decline in LeABI3 transcripts in these axes also occurs after germination regardless of whether GA is applied. A reduction in LeABI3 expression occurs in GA-treated axes isolated from gib-1 seed relative to the water control. This decline is not observed in embryos from intact seed under the same conditions, suggesting the endosperm in tomato regulates ABA signaling within the embryo.

P123. Microarray analysis of bast fibre gene expression in hemp

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Agricultural fibres are a promising alternative to wood fibres. Industrial hemp (*Cannabis sativa* L.) , in particular, is an environmentally friendly crop with significant potential as an alternative source of fibres in the pulp, textile, and bio-composite industries. However, current knowledge about the biology of fibre production in hemp is very limited. Gene discovery by microarray analysis is a useful approach to building a resource base for crop improvement of hemp (e.g. through marker-assisted breeding and reverse genetics). We have produced cDNA libraries from scutched stems of 3-4 week old plants of the hemp oilseed variety 'Carmen'. Approximately 7,000 cDNAs were selected from these libraries and were spotted in duplicate on glass slides to produce a high-density microarray. Here we report preliminary expression profiling results and DNA sequence information from among our preliminary hybridization experiments with these hemp fibre microarrays.

P124. Transcriptional responses to the xenobiotics 2,4-dinitrotoluene , 2,6-dinitrotoluene, and 1,3-dinitrobenzene

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TNT (2,4,6-trinitrotoluene) is the major component of landmines and other munitions, and is a significant environmental hazard both as an explosive and as a toxin. When TNT is exposed to soil, as occurs during leakage from landmines or unexploded ordnance, it is converted into a range of other compounds including 2,4-dinitrotoluene (2,4-DNT), 2,6-dinitrotoluene (2,6-DNT), and 1,3-dinitrobenzene (1,3-DNB). These compounds are typically more abundant than TNT itself, in the vicinity of a buried landmine. Therefore, as part of a long-term program to develop plants that may function in the sensing or bioremediation of TNT and related compounds, we have used oligonucleotide microarrays to examine the transcriptional response of the Arabidopsis genome to 2,4-DNT, 2,6-DNT, and 1,3-DNB. We have identified distinct inductive responses to these xenobiotics, with a particularly strong respone to 1,3-DNB. We have further analyzed the transcriptional responses of selected genes to 2,4-DNT, 2,6-DNT and 1,3-DNB through quantitative RT-PCR and promoter::GUS (beta-glucuronidase) reporter gene fusions.

P125. Glutathione improves shoot apical meristem identity during white spruce (*Picea glauca*) somatic embryogenesis

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Optimizing propagation of tree species through somatic embryogenesis remains one of the main objectives in sustaining superior quality genotypes for reforestation strategies. In white spruce, improvements of embryo development

were observed following experimental alterations of the endogenous glutathione redox state. An increase in embryos number and quality can be obtained through initial applications of reduce glutathione (GSH), which promotes cell proliferation and the formation of immature embryos, followed by application of the oxidized form (GSSG), which induces proper embryonic development. Structural and ultrastructural studies reveal marked differences between the control embryos and those of the treated counterparts. In addition, RNA in-situ localization studies of a shoot apical meristem marker gene, HBK1, indicate that changes in glutathione metabolism during embryo development improve shoot apical meristem stability, and ultimately regeneration of viable plants.

P140. Impact of isolation on pollination of Tall-Grass Prairie plants.

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This study compares insect pollination activity on small isolated and larger contiguous Tall-grass prairies to better understand the impact of habitat isolation on this process. Floral abundance and pollinator activity was monitored every two weeks for four months in 2004. Total and insect-pollinated plant species richness was lower at the isolated site compared to the contiguous site. The insect species richness and number of visitations of Dipterans were lower and Hymenopterans higher at the isolated site than at the contiguous site. Plants in the Asteraceae were each pollinated by 4-5 different insect species while plants in other families were pollinated by 1-2 species. The isolated site had greater variability in pollinating insect species over the year than the contiguous site. The environmental variables that most influenced insect activity were humidity, wind speed and cloud cover. These results show that insect diversity and pollination activity are different at isolated and contiguous sites, which may affect seed production.

P141. The effect of temperature and photoperiod on photosynthesis and the development of dormancy in contrasting ecotypes of dogwood (*Cornus sericea* L.)

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Dormancy development in plants is synchronized with the cessation of environmental conditions that favour plant growth. The timing of this response is important in trees for growth and survival. Woody plants, such as dogwood (*Cornus sericea* L.) have evolved different ecotypes that are closely adapted to local climatic conditions, which are differentiated by irradiance, photoperiod and/or temperature. While short photoperiods are well known to play a key role in dormancy induction in northern ecotypes, low temperatures alone can by-pass the photoperiodic requirement. It is well established that the combination of low temperature and even low to moderate light intensities to predispose plants to photoinhibition of photosynthesis. However, plants possess numerous photoprotective mechanisms to minimize photoinhibition including increased rates of photosynthesis as well as non-photochemical dissipation of excess excitation energy, a process involving xanthophyll cycle carotenoids. In addition, avoidance mechanisms such as the production of anthocyanins and other flavonoid compounds also exist. We used two ecotypes of dogwood, the northern ecotype of Alaska, U.S.A. (64^{o} 43' N latitude) and the southern Utah, U.S.A. (41^{o}N latitude) ecotype, which exhibit differential responses to photoperiodic- and temperature-induced dormancy. These ecotypes were exposed to various combinations of photoperiod and low temperature treatments and monitored for photosynthetic and photoprotective processes as the plants entered dormancy. Our results demonstrate differential long- and short-term responses to photoinhibition at low temperatures between the two ecotypes. The role of xanthophyll carotenoids and flavonoid compounds such as anthocyanins in mediating this process is discussed.

P142. The study of Alpine plant Ferulago angulata (schlecht.)Boiss essential oil constituents extracted from seeds of two different habitats, Nevakoh and shahoo, Zagross mountain ,West of Iran.

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The genus Ferulago comprises of thirty-five species world wide, of which seven species are found in Iran.In this study, the major components of Ferulago angulata (schlect.)Boiss seeds oil were extracted by hydrodistillation and was measured by gas chromatography and gas chromatography þu mass spectrometry.

From F.angulata growing in Nevakoh habitat 24 constituents were identified. From F.angulata growing in shahoo habitat 26 constituents were identified. The major components were found to be cis-ocimene(64.8%,76.11%), á-pinene(15.4%,7.29%), ã-terpinene(5.9%,2.88%), ñ- cymene (4.1%,1.4%), myrcene (1.9%,1.05%) and bornyl acetate(0.9%,1.69%).

P143. The Cypress Hills as habitat for Crataegus

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The Cypress Hills of Alberta and Saskatchewan lie just north of the US (Montana) border, are centered at 49^{o} 32? N and 109^{o} 50? W and occupy about 10^{4} sq km. The hills trend E-W and have a north-facing scarp slope, rising some 400-600 m to a summit plateau at 1200-1465 m. The southern slopes are gentle and the hills are dissected by some major valleys. The Cypress Hills constitute a mesic island in the surrounding prairie that is caused by interference to cold fronts originating to the north and northwest which increases precipitation from a prairie average of about 375 mm/p.a. to over 600 mm/p.a., while the altitudinal difference at the top of the scarp generates mean temperature differentials during the growing season of about 3^{o}. The soils are broadly chernozemic. This physical arrangement permits the growth of some near-scarp plateau and northern slope forests of lodgepole pine and white spruce in the most favoured areas; extensive aspen forests on the upper northern slopes and sides of major valleys which are often rich in Crataegus; and exceptionally large Crataegus-dominant thickets in the more mesic draws and valleys, which, however, peter out towards drier areas. The remainder of the Cypress Hills, when in natural condition, is short-grass prairie, with no hawthorns or indeed other woody plants other than subshrubs. This remainder constitutes perhaps 80% of the surface area, consequently the 'mesic island' is very fragmented except along much of the upper half of the northern slopes. The presentation illustrates those habitats with respect to Crataegus.

P144. Study of Genetic Diversity Among Cotton Cultivars by Polyacrylamide Gel Electrophoresis.

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Genetic diversity is desirable for long-term crop improvement and reduction of vulnerability to diseases and crop pests. Information about genetic parameters and relationships among the breeding materials has a significant impact on crop improvement. Association between parental divergence and progeny performance has not been well documented in cotton. A cotton study was conducted in the department of Plant Breeding and Genetics, University College of Agriculture, Bahauddin Zakaryia University, Multan, (30.2^{o}N, 71.4^{o}E) Pakistan and National Food Research Institute, Tsukuba, Japan. The objective of the study was to estimate genetic diversity among selected cotton genotypes and their reciprocal crosses along with the performance of genetic parameters. Six U.S. and two local (Pakistani) cultivars all belonging to Gossypium hirsutum L were genotyped by polyacrylamide gel electrophoresis. Cluster analysis revealed low genetic diversity among the parents and pooled the crosses with their parents, indicating success of hybridization. The present study, combined PAGE analysis with cluster analysis confirmed the genetic similarities between parents and their crosses while it also confirmed the dissimilarities between the parents as showed by the morphological characters. While the estimation of genetic parameters by diallel analysis showed the predominance of the additive with over dominance type of gene action among the F_1 and F_2 generations.

P145. Stem elongation plasticity in the alpine and prairie ecotypes of Stellaria

longipes: preliminary comparative studies with an α-expansin SlEXPA1 expressed in both the ecotypes.

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The alpine and prairie ecotypes of *Stellaria longipes* exhibit varying degrees of stem elongation plasticity. The alpine ecotype, found on an open alpine habitat, shows dwarf morphology. In contrast the prairie ecotype - a classic shade avoider, is able to elongate considerably, a beneficial trait in its crowded habitat. Stem elongation in *S. longipes* has been found to primarily involve cell elongation. Plant cell elongation is mostly based on cell wall loosening, mediated by a family of cell wall proteins known as expansins. We decided to investigate the role of expansins in the differential stem elongation plasticity exhibited by the two ecotypes of *S. longipes*. Southern blots using an 800 bp expansin conserved region fragment show the presence of multiple expansin members. An 1186 bp full-length cDNA clone (SlEXPA1) was obtained using 3' and 5' RACE procedures. SlEXPA1 is expressed in both the ecotypes. Plants of both ecotypes when grown under short day cold conditions are dwarf with short stems and ovate leaves. Transfer to long day warm conditions stimulates elongation of both the stem and the leaf. This response is however more dramatic and rapid in the prairie ecotype where elongation continues up to 20 days. The alpine plants do not elongate as much and growth ceases by the 16th day. Uppermost growing internodes were collected from alpine and prairie plants at 3h, 8h, 12h and 24h after placing under long day warm conditions, as well as on the 4d , 8d, 13d and 19d. Trends in the expression of SlEXPA1 was monitored in both ecotypes using semi quantitative RT PCR. SlEXPA1expression remains constant in prairie ecotype throughout the elongation phase. The alpine plants show an increase in transcript levels though these taper off towards the later stages of elongation. This corresponds to the elongation profile of both the ecotypes. A more accurate transcript profile would however be obtained if expression is monitored for specific internodes and leaves. Further studies with SlEXPA1 will primarily focus on the light and hormonal regulation in these two ecotypes.

P146. Phenology and biometry of 10 populations of barrel medic (Medicago truncatula Gaertn)

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The aim of this study is to evaluate the inside and within accessions variability in 10 populations of *Medicago truncatula* Gaertn. The statistical analysis has revealed four groups of populations: early group which characterized by a significant winter vegetative recovering and less leafy branching. However it ensures a good production of healthy pods (big pods and seeds), and presents the frequencies from 1 to 4 of pods per inflorescence.

In opposite, the late group with a weak rate of flowers (abortion) produces a high number of scaled pods (M.T.Y) and less for (M.T.T) and important spring vegetal recovering.

This group is characterized by an important frequency of 1 till 5 (MTT) a 7 pods (MTY) per inflorescence.

- Semi-early group was characterized by abundant foliage, and a rate of high abortion of flowers, otherwise, semi late has presented the mixing between the powerful and those at rates vigour's (low strengths)

- A clear polymorphism was observed in young plants (simple leaves) pods and seeds.

The results of the biometric characters of the pods (and the seeds /pods) by inflorescence are decreasing from one, two... and six pods by inflorescence.

P147. Patterns in terrestrial bryophyte and lichen species in young and old sub-boreal spruce forest

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This study examined the effect of forest age on the diversity, biomass and abundance of terrestrial moss, lichen and liverwort species. The study compared old-growth and young

second-growth sub-boreal spruce forest sites at the Aleza Lake Research Forest in central British Columbia. Terrestrial lichen and bryophyte diversity patterns, and the processes affecting them, have not been extensively studied in the Sub-Boreal Spruce biogeoclimatic zone in B.C. Forest harvesting is a major landscape altering process central B.C. and clearcut logging can affect terrestrial bryophyte and lichen communities through changes to substrate and microclimate and through habitat fragmentation. The effects that extensive forest management is having on bryophytes and lichens in sub-boreal spruce forests is not well documented.

In total, 116 species of bryophytes and lichens were identified in both stand ages. Major differences in species composition were found between forest ages with 49% of lichen and bryophyte species found in common between the forest ages, 30% found only in old-growth forest and 21% found only in young second-growth. Liverwort species were much more diverse in old-growth with half of the liverwort species found only there and 96% of the recorded liverwort cover occurring in old-growth forests. Lichen cover was greater in second-growth stands and different assemblages of lichen species were common in young second-growth compared with old-growth stands. Moss abundance and diversity was similar between forest ages, however, species compositions varied. Second-growth stands were largely dominated by a single moss species.

This study highlights the need for additional study of the impacts of logging practices and rotation times on lichen and bryophyte diversity. The length of time required to accomplish the transition from second-growth to old-growth non-vascular and lichen floristics is unknown for this region.

P148. Aspects of Seed dispersal of the Gulf of Saint Lawrence Aster, *Symphyotrichum laurentianum*: disturb to preserve.

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The Gulf of Saint Lawrence Aster, *Symphyotrichum laurentianum*, is categorized as a 'threatened' plant by the Committee on the Status of Endangered Wildlife in Canada, an elevated designation from its previous classification of 'special concern' prior to May 2004. This halophytic member of the Asteraceae family is endemic to New Brunswick, Prince Edward Island, and the Magdalene Islands. *Symphyotrichum laurentianum* colonizes disturbed habitats and can be found growing in or around saline marshes, tide pools, dune slacks, sandy beaches, and other unstable saline environments. The populations of these asters on P.E.I. have fluctuated greatly in recent years and populations estimates from the autumn of 2004 show an overall increase from the last two years. Since dispersal is believed to be important for pioneer species adapted to disturbance, the main goal of this study was to assess the wind dispersal potential for the achenes of *S. laurentianum*. Although this plant is dispersed primarily by wind, in situ observations and experimental data show that the effectiveness of this method of dispersal may be minimal due to the surrounding canopy of vegetation and flooding events. A larval lepidopteran seed predator was discovered consuming achenes of *S. laurentianum* and its impact where it was present could pose an additional constraint on its dispersal. These data will help better formulate a management plan for this rare plant.

P150. Landscape pattern and persistence of grassland-forest ecotones:

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Aspen (*Populus tremuloides* Michx.) encroachment in the plains rough fescue grasslands of Riding Mountain National Park (RMNP) was examined using historic aerial photographs (1956, 1964, 1978 and 1996) and the spatial point pattern of stems within aspen groves. Four fescue grasslands were examined during 2003 (Grasshopper Valley, Baldy, Deeplake and Bobhill prairie). Results indicate that since the mid-1900's forest cover has increased by at least 20%. The degree of forest encroachment is related to topo-edaphic complexity. Encroached sites are generally characterized as having transitional forest-grassland soils. By contrast, forest edges which have remained stable over the past fifty years are characterized by abrupt changes in soil properties. Soils and topography are considered to be strong determinants of aspen encroachment patterns. The spatial point pattern of aspen was quantified within four clones in RMNP. In every case, clones are

expanding into the prairie. However, recruitment is followed by a high mortality phase (80% mortality) for individuals > 1m in height. All standing dead stems have evidence of repeated browse from ungulate herbivores. We suggest that the pattern of aspen encroachment may be determined by a combination of the spatial extent and persistence of the root system, landscape complexity, ungulate browsing, and fire history.

P151. Frugivory by three large mammals in the Canadian Arctic.

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The diversity of fruit-bearing plant species in the central Canadian arctic is low (3.8% of 342 spp.), however fruit densities may be locally high (up to 200 fruits and 3,600 seeds per m^2). The ability of herbivorous mammals to disperse seeds of such fruits was examined by analyzing the viable seed content of fecal pellets from barren-ground caribou (*Rangifer tarandus*), Arctic hare (*Lepus arcticus*) and Arctic ground-squirrel (*Spermophilus parryi*) from four mid-arctic tundra plant communities, near Baker Lake, Nunavut. Germination tests were conducted on fragmented pellets after a period of stratification (4^oC). Arctic hare and Arctic ground-squirrel were both effective seed dispersers with 15.8 and 43.6 viable, germinable seeds per 10g of dry fecal pellets, respectively. The majority of seeds (87.4%; hare, 94.3% squirrel) were from the soft-fruited species, *Vaccinium uliginosum* (northern blueberry), with small amounts for *V. vitis-idaea* (rock cranberry) and *Empetrum nigrum* (black crowberry). Fruits of each species were different in color (blue, red and black, respectively), but were otherwise similar in shape (spherical), diameter (0.7 cm) and possessed numerous small seeds. Dispersion of hare pellets was widespread on the tundra and assisted by anemochory, but squirrel pellets were confined to the vicinity of their burrows, nevertheless, numerous seeds of all three species were abundant in the dormant soil seed bank. Caribou were not proven to be responsible for the dispersion of any viable seeds, even though their migratory behavior would suggest that they are potentially effective agents.

P152. Bracteoles, a new character in *Crataegus* taxonomy.

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Bracteoles, small leaf or part-of-leaf homologues, are abundant in *Crataegus* inflorescences. They appear to have a role in protecting flowers and their pollinators from ants. Nearly all bracteoles have marginal glands, often on teeth of various lengths, are venous and generally 5-10 mm long. Although noticed as early as 1834 by Spach, we are the first to record their detailed structure. Two apparently fundamentally different kinds of bracteole occur, the first, restricted to a taxonomically coherent group of mainly Asiatic species, is larger in size, falcate in shape, green and persistent. The second, found in all species, is generally smaller than the first type, symmetric, caducous to sub-persistent, and is scattered throughout the inflorescence. This has five sub-types which may be characterized as: broad, ± herbaceous, ± persistent, venous; narrow, caducous, membranous, glands ± sessile; narrow, caducous, membranous, ± eglandular; a little broader, caducous, membranous, bordered with glands on short teeth; as last, glands on long teeth. These subtypes often match major taxonomic groups. The presentation discusses the types in more detail and relates it to the taxonomy.

P165. Stomatal adaptation in native poplars of southern Alberta.

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Several poplar species meet at the margins of their natural distributions in southern Alberta. In this semi-arid region poplars are obligate riparian species, but within this context occupy several intergrading ecoregions. *Populus deltoides* predominates in the warmest and driest eastern prairie ecoregions; *P. balsamifera* occupies the wetter and cooler western parkland and montane ecoregions; and *P. angustifolia* and hybrids between the species occur in intermediate grassland ecoregions. We have investigated stomatal characteristics in these poplars in genotypes collected across the range of ecoregions and grown in a semi-arid common garden.

Stomatal length differed among genotypes within species but did not differ among species, ranging from 19-22 µm. Total stomatal densities (adaxial plus abaxial) differed among genotypes within species but were similar among species (290-420 stomata mm^{-2}). Single-surface densities did differ among species and consequently the ratio of adaxial:abaxial stomatal density also differed, ranging from 0.94 for *P. deltoides* to 0.27 for *P. balsamifera*, with intermediate density ratios in *P. angustifolia* and hybrids. In a subsequent study of a subset of the same genotypes stomatal density was shown to be correlated with stomatal conductance and conductance ratios differed among species in the same manner as seen previously for density ratios. We conclude that: [1] diverse poplar genotypes respond similarly to a semi-arid environment by producing comparatively small and dense stomata; [2] differences in stomatal density underlie differences in stomatal conductance; [3] differences among species in the density ratio or conductance ratio may reflect adaptation to climatic differences among ecoregions; [4] there is considerable variation in stomatal characteristics, which offers opportunities for selection of genotypes that may be particularly well-adapted to prairie conditions.

P160. Natural Product Metabolism in *Monarda didyma*.

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Monarda, members of the mint family (Lamiaceae), are erect perennials native to many regions of North America with over 20 species known. Several varieties (e.g. *M. didyma* and *M. fistulosa*) are grown for essential oil production, and for used in alternative medicine. *M. didyma* (variety Marshalls delight) is an hybrid of *M. didyma* and *M. fistulosa*.

The monoterpene and flavonoid content of some Monarda species have previously been characterized; however, developmental production of these secondary metabolites has yet to been investigated. In this study, we investigated the tissue specificity and developmental production of monoterpenes (main aromatic constituents of the essential oil) and flavonoids in the leaves and flowers of *Monarda didyma* (variety Marshalls Delight). The essential oils of the leaves and flowers contained linalool, carvacrol and gerianol as the major constituents, while cineole, p-cymene, alpha-pinene, and beta-pinene represented minor components of the oils. There was no difference in the linalool concentration in young versus old leaves. However, carvacrol and geraniol concentration, increase in fully mature leaves when compared to young leaves. Preliminary HPLC results indicate that the flavonoids hyperoside, quercitrin and quercetin are present in the Monarda leaf methanol extracts. Improvement of flavonoid extraction process, development of HPLC-MS methods, and additional flavonoid standards are required to identify and quantify the remaining flavonoids present in Monarda leaf tissue.

P161. Composition and antioxidant activities of polyphenols in sour Cherries (*Prunus cerasus* L.)

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Antioxidants have important human health benefits as well as the ability to increase the shelf life of food products. Since consumers have become much more aware of synthetic food additives, there is increasing interest to replace artificial colour additives in foods. Both sweet and sour cherries have been shown to contain significant amounts of several anthocyanins and hence, these have been explored as natural sources of colouring agents and antioxidants. Anthocyanin content and compositions of twenty genotypes of sour cherries (*Prunus cerasus* L.) from the Vineland Research Station, University of Guelph, were evaluated. Based on total polyphenol content, which ranged from 100 mg/100g.fresh weight to nearly 600 mg/100g fresh weight, four high anthocyanin containing genotypes were selected for further analysis. Antioxidant activities, as measured by superoxide radical and hydroxyl radical scavenging capacities showed a dose-dependent increase with increasing polyphenol content. However, varietal differences in antioxidant capacities were observed that might be related to their anthocyanin composition. MALDI-TOF-MS

analysis of anthocyanins showed the presence of glucosides of cyanidin, peonidin, delphinidin, petunidin and malvidin along with their acetate and coumarate derivatives. There were considerable variations in the anthocyanin composition between the varieties that might be related to their unique differences in antioxidant characteristics.

P162. Molecular cloning and characterization of a phospholipase D alpha cDNA from strawberry (*Fragaria ananassa* Duch) fruits

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To study the molecular properties and developmental regulation of PLD, a key enzyme involved in membrane deterioration that occurs during fruit ripening and senescence, we cloned a 2874 bp full-length cDNA for PLD alpha from strawberry fruit mRNA by using RT-PCR and 5'- and 3'- RACE (GeneBank accession number: AY758359). This sequence includes a 2433-nucleotide long ORF, which encodes an 810 amino acid-polypeptide with a predicted molecular mass of 91.4kDa and an isoelectric point of 5.76. Sequence search results suggested that strawberry PLD alpha is a very stable and soluble protein with a grand average hydropathicity of –0.395. Multiple sequence alignments showed a high degree of similarity between strawberry PLD alpha and other PLD alphas from plants, with a 74% similarity to PLD alpha from Arabidopsis and 81% to that from tomato. Phylogenetic analysis showed the relationships between PLD alpha, beta and gamma. PLD alpha possesses the characteristic N-terminal C2 domain and the two phospholipase active site domains known as HKD motifs, conferring calcium sensitivity and the enzyme activity, respectively. Alignment of amino acid sequences of C2 domain showed minor differences in the Ca^{2+}-binding residues, which may explain the different Ca^{2+} affinity between strawberry PLD alpha and other PLD isoforms. Additional motifs of interest include two motifs flanking the C-terminal HKD motif, enriched in basic amino acids asparagine, arginine and glutamine, which may bind the negatively charged modulators of PLD, and two hydrophobic motifs comprising isoleucine, valine and tyrosine. The molecular role of phospholipase D in fruit ripening and senescence is discussed.

P163. Molecular Analysis of Two Banana Genotypes Induced In Vitro Under Gamma Rays Stress

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The aim of the current work is to investigate the effect(s) of Gamma irradiation in Banana on DNA and protein levels respectively. Shoots of banana genotypes, Grand Naine and Shikita were initiated on Murashige and Skoog (MS) medium containing 2.0 mg /L benzyl adenine. Shoots were then irradiated with ^{60}Co, doses 0.0, 15.0, 30.0 and 45.0 Gy respectively. DNA was subject to Random Amplified Polymorphic DNA, RAPD-PCR, using ten random primers. Eight primers (80%) generated polymorphic RAPD profiles. Furthermore, comparative RAPD profiles were dissimilar indicating genomic variations. Protein expression profile was variable due to the absence and/or detection of de novo protein entities. Proteins with different molecular weights were observed in the treated samples if they compared with the controls. Variation in protein expression pattern was influenced by several factors including the genotype and the irradiation dose. Our results suggest that gamma-irradiation have induced variations in the DNA and protein evident to the variable RAPD and protein expression profiles respectively.

P164. In vitro isolation of viable gametes in lupins (*Lupinus albus* and *Lupinus mutabilis*)

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In vitro isolation of viable male and female gametes of *Lupinus albus* and *Lupinus mutabilis* were carried out to facilitate gene transfer between these two species. Germinated pollen tubes in culture medium were used to isolate sperm cells. The effect of concentrations of potassium in the culture medium was found to be critical for pollen germination and pollen tube growth. Effect of concentrations of boric acid, calcium chloride, potassium phosphate, and sucrose on isolation of sperm cells was also reported.

The highest yield of egg cells (20 %) was achieved when cellulase (1 % (w/v)) and pectinase (0.5 % (w/v)) were used for macerating unfertilised ovules, followed by mechanical manipulation. The viability of sperm and egg cells were determined by positive reaction of fluorescein diacetate (FDA).

P165. Fluorodetection of copper toxicity in sunflower plants

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UV-induced chlorophyll (ChlF) and blue-green (BGF) fluorescences emitted by leaves were proposed as useful indicators of the physiological state of plants and of their responses to environmental stress such as mineral deficiencies, pathogens, prolonged exposure to UV, etc. Considering the increasing occurrence of copper in ecosystems, it was important to also evaluate the potential of UV-induced fluorescences to detect the toxic effect of this heavy metal on plant growth. Sunflower plants were cultivated during a 11 day-period in a growth chamber in presence of normal (2 µM) and toxic (75 µM) concentrations of Cu^{2+} in the nutrient solution (Hoagland). During the experiment, biomass accumulation, CO_2 assimilation rates, chlorophyll and soluble polyphenols concentrations, and ChlF and BGF intensities were regularly measured. On the 2nd day of the experiment, lower biomass accumulation and lower root/shoot ratio were already apparent in plants treated with 75 µM Cu^{2+} compared to control plants. On the 4th day and thereafter, Cu-inhibited plants showed lower CO_2 assimilation rates, lower chlorophyll concentration but higher concentration of soluble polyphenols compared to control plants. Concerning fluorescence intensities, it is noteworthy that only one day after the start of the experiment, it was possible to detect a decrease of UV-induced ChlF in Cu-inhibited plants. However, no difference in BGF from control and Cu-inhibited plants were observed throughout the experiment. Therefore, these results indicated that the more pronounced inhibitory effects of Cu^{2+} on plant growth relative to photosynthesis may increase carbon allocation toward polyphenol synthesis. The accumulation of these UV-absorbing compounds in the cell vacuoles of leaf epidermis will then decrease UV-excitation of chlorophylls in leaf mesophyll.

P166. Factors affecting enhanced resistance to aluminum among wild and cultivated plant species on the Island of Madeira.

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Diversity of flora in island ecosystems is determined by both natural and anthropogenic factors such as geographical isolation, climate, colonization events and industrialization or urbanization pressure. Madeira, the volcanic island on the Atlantic Ocean exhibits a great diversity of soil and edaphic conditions. Physical and chemical properties of soils including their pH, texture and structure, content of metals and organic matter play an important role in the distribution of plant species. These factors can either severely restrict occurrence or enforce adaptation of plants to local conditions. In acid volcanic soils with pHs below 5.0, toxicity of aluminum (Al) is considered to be a major growth limiting factor. One hundred and sixty top-soil samples obtained from different locations throughout the island were analyzed for pH, Al and organic matter content. The pH of topsoil ranged from 3.84 to 7.44, with a mean soil pH_{KCl} of 5.01. On average, the soils contained 0.79 $cmol.kg^{-1}$ of Al and 3.02% of organic matter. Three plant species, two agricultural (wheat: *Triticum aestivum*, corn: *Zea mays*) and one wild (Sinapidendron), were studied. Seeds of 48 genotypes of wheat, 40 genotypes

of corn and 10 populations of Sinapidendron were collected from sites of known soil chemistry and the resistance to Al of hydroponically grown seedlings was evaluated using the eriochrome cyanine test. We found that soil conditions had profound effects on the distribution of both cultivated and wild species. Enhanced resistance to Al was observed among the Madeiran plant species, especially among the cultivated ones. Al content in soil solution was the main environmental factor involved in development of Al resistant genotypes and at the same time it could be a factor determining the distribution of cultivars/ecotypes across the island. A large number of Al resistant genotypes found among the tested species suggests a possible involvement of environmental and anthropogenic factors in diversification processes and the development of new ecotypes highly adapted to local edaphic conditions.

P167. Excitation energy distribution of the mesophilic and psychrophilic strains of *Chlamydomonas raudensis.*

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Chlamydomonas raudensis Ettl. (UWO 241) was isolated from Antarctica's permanently ice-covered Lake Bonney. Recently, this psychrophilic green alga was identified as a strain of *C. raudensis* Ettl. (SAG 49.72), a mesophile. Excitation energy distribution between photosystem II (PSII) and photosystem I (PSI) was examined by low temperature (77K) chlorophyll fluorescence in response to growth of these strains at different temperatures. The psychrophile, previously found to be deficient in state I – state II transitions, was unable to regulate the light energy distribution between the two photosystems in response to changes in temperature. In contrast, the mesophile showed a gradual increase in the PSI-related chlorophyll fluorescence emission maxima at 713 nm (F713) relative to PSII associated fluorescence at 682 nm (F682) with decreasing temperature. Interestingly, the relative abundance of PSI reaction center polypeptide PsaA gradually decreased with decreasing growth temperature from 28°C to 11°C, while the abundance of PSII reaction center polypeptide D1 did not change. This indicates that the enhanced fluorescence emission at 713 nm could not be due to temperature induced changes in the abundance of PSI-related proteins, but rather to temperature dependent excitation energy redistribution in favour of PSI. This was accompanied by an increase in PSI cyclic electron flow. Increased cyclic flow around PSI in the mesophile positively correlated with increasing NPQ as temperature decreased. In contrast, the psychrophile showed relatively constant higher levels of NPQ and higher capacity for PSI cyclic electron transfer regardless of growth temperature. Thus, acclimation of the mesophile shows the ability to optimize the light energy used towards photosynthesis by regulating imbalances in energy distribution between PSII and PSI and modulating the capacity for non-photochemical quenching, while the psychrophile exhibited a limited capacity for temperature acclimation.

P168. Endogenous gibberellin (GA) levels in response to varied light irradiance in *Stellaria longipes* alpine (sun) and prairie (shade) genotypes.

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Plants grown under low light irradiances (low PAR [photosynthetically active radiation]) show a shoot elongation growth (etiolation) similar to that observed under enriched far red light (FR). Individuals of the Stellaria longipes alpine population (1D non-shade [sun] genotype) grow on an open alpine plateau at 2,453 m in the southern Alberta Front Ranges. They have very short internodes and small, but wide leaves. In contrast, plants in a lower elevation population (7B [shade] genotype) grow in an open "prairie-type" grassland in the lower foothills. They have long internodes and long and narrow leaves. We used 1D and 7B as model sun and shade plant systems to study the role of GAs in the etiolation associated with low PAR. Both genotypes responded to low PAR treatment in a similar manner with regard to increased stem growth. However, reduced PAR modified the endogenous GA_1 levels of the two genotypes quite differently. The 7B "shade" genotype showed increased GA_1 (the "growth effector" GA) levels over time under low PAR, while the 1D genotype showed just the opposite trend. Exogenous application of growth-active GA_3 and GA_4 significantly promoted etiolation of 7B

genotype, even when grown under low PAR, but had no significant effect on the 1D "sun" genotype under low PAR. Thus, while the growth responses to reduced PAR are similar (in direction, but not magnitude) for these two genotypes, which have very different phenotype characters, their endogenous phytohormone (GAs) responses are quite different. It appears that the phenotype plasticity associated with the "shade" 7B genotype is directly related to its ability to synthesize GA_1 and respond to applied GAs. In contrast, the "sun" 1D genotype, with a very reduced stature, appears to have muted its response to relatively high endogenous GA_1 levels, and also to applied GAs, e.g. some other factor is controlling the magnitude of its shoot growth response.

P180. Seed yield improvement in fenugreek (*Trigonella foenum graecum* L.) using mutation breeding.

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Fenugreek is an annual legume widely grown as spice. It can produce high quality, bloat free forage and is a source of diosgenin. The first North American forage cultivar "Tristar" has indeterminate growth habit and needs 120 days to produce mature seed. To generate mutants that would produce high quality seed we used Ethyl Methane Sulphonate (EMS) on Tristar fenugreek. Seed were presoaked in distilled water and treated with varying EMS concentrations between 10 and 300 mM for 2, 4, 6, 8, 12, 16 & 24 hours. Plants were allowed to grow for 85 days, treated with a desiccant, allowed to dry before seed yield and quality was determined. Plants with high seed yield and uniform golden yellow colour seed were selected for producing the next generation. Progenies of some large seeded high yielding M_2 plants have shown the ability to mature early. One M_3 line derived from a selected plant has grown well under greenhouse condition with determinate growth habit and high seed yield. Since fenugreek is a self pollinated crop this line has the potential to become a new cultivar. This work indicates usefulness of mutation breeding in fenugreek improvement.

P181. Targeting Better Hemp Traits Using Reverse Genetics

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Reverse genetics is being broadly applied as a tool for establishing large populations of non-transgenic plants that can be targeted for desirable traits such as oil quality, fibre ratios, and agronomic traits. Key methods for reverse genetics include RNAi, fast neutron deletion, and transposon insertional mutagenesis. We are using an alternative and high-throughput approach called Targeting Induced Local Lesions in Genomes (TILLING) to generate novel plant traits in hemp.

We have generated a 15,000 EMS-mutated hemp population and are using TILLING to identify viable genetic mutations which improve hemp fibre profiles, all while circumventing the traditional definition of a Genetically Modified Organism (GMO). TILLING has several advantages over other reverse genetics approaches including the generation of allelic series of mutations, elimination of lethal mutations, is relatively inexpensive and rapid in the detection of mutations, can be applied to nearly any species of plant, can be used in the screening of populations for nearly any trait, and through the use of EMS as a mutagenic treatment does not cause excessive collateral damage across the genome.

Hemp is being increasingly recognized in Canada as a valuable agronomic commodity because of its high quality fibre and healthy oil product. Due to its illegal status as an agricultural product prior to 1998, hemp has been vastly under-researched and under-bred in North American. Hemp fibre is a superior alternative to traditional forest products in North America because it can yield four times as much paper per acre than poplar. We are filling a gap in the breeding of hemp in Canada quickly and effectively, all while generating products that have a viable place in today's emerging bioproducts industry and eco-conscious market. I will be discussing the on-going hemp reverse-genomics program at the Alberta Research Council Inc.

P182. The Development and Characterization of an Inter-Specific Hybrid Lineage Derived from *Brassica carinata* and *Brassica napus*.

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1)
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Each modern crop species has its own set of important fungal, bacterial and viral pathogens. Many research groups around the world work on improving our chemical and logistical control of pests, as well as the handling of possible side-effects from using these controls within our environment. Other research avenues concentrate on improving the genetic resistance of crops. Of all approaches, increasing a crops' genetic resistance to pathogens is now regarded as the most viable means to controlling pests of all kinds. To achieve genetic resistance to pathogens, it is common practice within crop breeding programs to attempt to "move" certain traits from one variety to another. Often, the 'donor' lines are from a different genus or species than the 'recipient' cultivar. This approach serves to increase the diversity of crop varieties, and can result in the transfer of a valuable trait. However, often these attempts are unsuccessful, or take many more generations than expected due to our limited understanding of recombination within complex crop genomes.

To increase our knowledge in this area, we have developed advanced backcross families from *Brassica carinata* (genome symbol BBCC) and *Brassica napus* (genome symbol AACC).The ambitions of this project are to transfer a genetic disease resistance trait from one species to another, to record the events of recombination occurring in this wide cross using molecular markers, and to record the morphology of one of these lineages. Initial hybridization required ovule rescue tissue culture, followed by a few generations of backcrossing to seed. Fertility, as well as other characteristics of interest, were variable, and were found to be related to the novel chromosome arrangements in the families. Here, we document the process and the phenotypic results of creating interspecies lineages between *B. carinata* and *B. napus*.

P183. Diallel Analysis of Vernalization Response in Spring Wheat

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We investigated the vernalization response of five selected Canadian spring wheat (*Triticum aestivum* L.) cultivars ('AC Taber', 'AC Foremost', 'AC Barrie', 'AC Intrepid' and 'Cutler') and their F_1 crosses made in a one-way diallel mating design. The parents and F_1 crosses were evaluated under vernalized (6 weeks) and un-vernalized long-day (16 hours photoperiod) conditions in a controlled environment. Final leaf and spikelet number on the main stem, days to anthesis and maturity, and tiller number and yield per plant were mainly controlled by additive gene action, as indicated by the significance and preponderance of general combining ability effects. Specific combining ability effects were not significant for all the traits. Some of the F_1 crosses deviated significantly from the mid-parental values suggesting the existence of non-additive gene effects. The significance of general combining ability and non-significance of specific combining ability indicated that general combining ability alone could adequately predict the single-cross progeny performance. Narrow-sense heritability was low for tiller number (0.25) and days to maturity (0.32), medium for yield (0.53) and relatively high for FLN (0.80), spikelet number (0.86) and days to anthesis (0.93). Flowering time in the absence of vernalizing temperatures could be the best selection criterion for the elimination of vernalization response in the early stages of a spring wheat breeding program.

P184. The impact of seeding rate and row spacing on productivity of canola

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Canopy manipulation implemented by varying seeding rate and row spacing has been suggested as a potential method to reduce the severity of sclerotinia stem rot of canola. The impact of these measures on

canola productivity was examined in the absence of disease in experiments at Melfort, SK and Lacombe, SB from 2002-2004. The effect of 4 row spacings (23, 31, 46 and 62 cm) and three seeding rates (0.5, 1.0 and 1.5 X the recommended rate of 6 kg ha^{-1}) was examined for two glyphosate tolerant varieties (hybrid and open-pollinated (OP)) of canola in a direct seeding system. The effects of these factors were determined on days to first flower (DTSF), flowering period (FP), lodging, plant population, yield, % green seed (GS) and thousand seed weight (TSW). The DTSF and plant population were greater for the Hybrid than the OP, while FP, lodging score and GS varied inconsistently between cultivars. Yield was greater for the hybrid than the OP cultivar under dry conditions, but the OP was greater at 4 site-years under normal moisture conditions. The TSW of the OP was consistently greater than the hybrid. Seeding rate had few consistent effects on the factors measured except plant population, which increased with increased seeding rate. Yield of the 0.5 and 1.5 X rates were not different from the 1.0X rate, except at 1 site-year where yield of the 0.5X treatment was reduced compared to the 1.0X rate. Row spacing had the greatest impact on most measured factors. DTSF, FP and lodging score tended to increase and plant population decrease as width of row spacing increased. Yield at the 23 cm row spacing was always greater than or equal to yield at other row spacings. At 2 site-years, yield at the 61 cm row spacing was less than at other row spacings. The GS and TSW were not usually affected by row spacing. The results indicted that the use of wider row spacing may have a detrimental impact on crop productivity in the absence of disease and may not be a useful routine practice to reduce stem rot risk. Moreover, there was an indication of enhanced lodging with wider row spacing, which can increase disease risk.

P185. The effect of cultivar, seeding rate and row spacing on seed yield of kura clover (*Trifolium ambiguum*) in central Alberta.

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Kura clover (*Trifolium ambiguum*) is a perennial forage legume recently introduced into Canada as it exhibits several desirable traits for use in mixed species pastures. Recent studies have shown that this clover is persistent and productive under Alberta growing conditions, however, wide adoption of the species would be enhanced by the development of a local seed source.

The purpose of this study was to determine if kura clover will produce seed under central Alberta growing conditions, and to determine the agronomic practices that maximize seed yield. Two cultivars of kura clover (Endura and Cossak) were sown at four seeding rates and three row spacings in Edmonton, Alberta. Seed yield, 1000 seed weight, flowers per head and seeds per head were measured for each treatment. In the first production year seed yield ranged from 84 kg/ha for cultivar 'Cossak' sown at 12 kg/ha and 45 cm spacing, to 342 kg/ha for cultivar 'Endura' sown at 9 kg/ha and 45 cm spacing. Averaged across all treatments the cultivar 'Endura' had higher seed yields, and 1000 seed weights, than the cultivar 'Cossak'.

Kura clover can successfully flower and produce mature seed under the short growing season of central Alberta, however, chemical desiccation of the stand may be necessary to ensure a timely harvest and seed loss at harvest may be significant.

P186. Salt Stress Tolerance in Diploid Potato Clones

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Incorporating salt stress tolerance of wild diploid potato species into cultivated tetraploid potatoes are hampered by a lack of quick, efficient and representative salt stress tolerance screening methods to evaluate diverse potato genotypes. The objectives of this research were to; a) identify reliable indicators of stress tolerance in potato and b) evaluate mechanisms of stress tolerance. 22 diploid salt-tolerant (T) and sensitive (S) clones of early-maturing (EM) and late-maturing (LM) diploid potato lines were stressed for 5 d at the tuber initiation stage with 150 mmol NaCl in a

hydroponic sand culture under greenhouse conditions. Relative growth index (shoot growth under stress/shoot growth under control conditions) was easily measured and was a good predicator of overall salt stress tolerance compared to the degree of similarity cluster analysis. However, method of stress application impacts responses. To evaluate mechanisms of salt tolerance, potato clones were exposed to 150 mM NaCl for 5 days at the tuber initiation stage. Under salt stress, early-maturing (EM) clones accumulated Na^+ while late-maturing (LM) clones excluded it from the leaf tissues. Auto- and hetero-grafting was used to further examine the effect of rootstock on exclusion of Na^+ from the shoots and the effect of abscisic acid (ABA) on expression of the Ca^{2+}-storage protein calreticulin (CR). Increased CR expression was induced by NaCl stress and associated with salt resistance in LMT and EMT clones with higher levels of CR in the LMT clone. The salt tolerance of the EMS clone increased when grafted onto LMT rootstocks but not onto EMT rootstocks. ABA deficient clones (AD) were characterized by low CR levels, which did not increase after salt stress. However, grafting AD scions onto LMT rootstocks increased CR expression in the AD portion of the graft combination. Salt-stress induced CR expression and is positively associated with the presence of ABA and salt-stress tolerant phenotypes.

P187. Nitrogen use efficiency of field-grown cucurbits.

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As Ontario nutrient management legislation requires producers to strictly adhere to provincial guidelines for nutrient applications, it may be more critical to optimize nitrogen (N) use efficiency (NUE) in order to maintain crop yields and minimize environmental N losses. Three field experiments each of once-over simulated machine-harvested processing cucumbers and processing winter squash were conducted to determine the effects of different N rates, timing of application, and technology (i.e. nitrification inhibitor combined with an urease inhibitor) on yield and NUE. Preplant ammonium nitrate was broadcast applied at 0, 70, 110, 165, and 220 kg N ha^{-1}, as well as a split application of 70 plus 40 kg N ha^{-1} applied preplant and before the vines run, respectively. A preplant application of UMAXX®, a urea-based fertilizer that contains a nitrification inhibitor and an urease inhibitor, was broadcasted at 110 kg N ha^{-1}. At harvest, the following parameters were evaluated: total and marketable yield; crop quality including cucumber brine quality; shoot and fruit percent total N and phosphorus; and soil mineral N. Results presented here are based on the first year of a four year study. In 2004, although plants in zero N plots displayed symptoms of N deficiency, the quantity of N applied (0-220 kg N/ha), the method of application (preplant vs split) and N source (ammonium nitrate vs nitrification and urease inhibitor) did not influence total and marketable yield nor crop quality. Generally, NUE (expressed as the difference in plant N from fertilized vs. non-fertilized plots divided by quantity of fertilizer applied) decreased as N rates increased from 110 to 220 kg N ha^{-1} for both cucurbits. Squash NUE was considerably higher than cucumber at =37% compared to =14%, respectively, which was likely due to the stage of plant development at harvest. At harvest, the quantity of N remaining in the field (i.e. crop N residue plus soil mineral N at 0-75 cm depth) in the N non-fertilized plot varied greatly with location from between 129 to 318 kg N ha^{-1}. In one cucumber field, the non-fertilized plots had more residual N than plots receiving 110 or 220 kg N ha^{-1}. Although further research is underway, preliminary results indicate an opportunity to reduce N rates and thus increase NUE and decrease potential N environmental losses.

P188. Liming and calcium fertilizer application for clubroot control in canola (*Brassica napus* L.)

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Clubroot, caused by *Plasmodiophora brassicae* Woronin, is an important soilborne disease of cruciferous crops worldwide. It was identified in several canola (*Brassica napus* L.) fields near Edmonton, Alberta in 2003, in the first report of clubroot on canola in the Canadian Prairies. Agricultural lime ($CaCO_3$),

calcium cyanamide ($CaCN_2$) and two $CaCl_2$-based commercial fertilizers (Soil-Cal and Ferti-Cal) were evaluated in greenhouse tests for their effectiveness in reducing disease severity. Due to high disease pressure, symptom severity was high in all treatments compared to non-inoculated controls. Nevertheless, a significant reduction in root galling and less stunting of the plants was observed in plants grown in soil amended with $CaCO_3$. This may have been related to the fact that the highest pH was detected in the $CaCO_3$-amended soil, since clubroot disease development is favored in acidic soils.

P189. Effect of harvesting methods on dry edible bean production

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Dry bean (*Phaseolus vulgaris*) production in Northern Quebec could be limited by the humid and cool climate. During 3 years (2002 to 2004), 13 cultivars and 2 harvesting methods (direct combining and conventional) were evaluated to: 1) determine the adaptability and productivity of dry bean in Northern Quebec and 2) evaluate grain yield loss according to harvesting methods. Treatments were laid out in a factorial design with 3 replications. Grain yields were affected by dry bean cultivars. Pinto bean had higher average grain yield in 2002 compared to other types of bean. Nevertheless, pinto bean grain yield was low in 2003 and 2004 because this type of bean was severely affected by sclerotinia stem rot. In 2002, 2003 and 2004, direct combining reduced grain yields by 31 %, 39 % and 28 % compared to conventional harvesting method

P190. Intercropping barley, triticale and fababeans at Breton, Alberta.

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Intercrops of cereals and pulses have the potential to increase silage yield and protein content, and to decrease N fertilizer requirements compared to the same crops grown in monocultures. Barley, triticale and fababeans were grown alone and in mixtures on a Gray Luvisolic loam at Breton, Alberta in 2002, 2003 and 2004. Experiments assessed biomass yield and protein concentration at silage-stage of barley-triticale-fababeans intercrops compared to monocrops. The effects of seeding fababeans prior to cereals, changing the within-row and between-row seeding arrangement, and adding nitrogen fertilizer were also assessed. Drought conditions in 2002 and 2003 greatly reduced fababean biomass, producing dry matter yields of 2-3 Mg ha^{-1} for fababeans grown as monocrops. Dry matter yields of intercrops ranged from 3-8 Mg ha^{-1} and usually did not provide a yield advantage compared to monocrops of triticale or barley. With normal rainfall in 2004, one barley-triticale-fababeans intercrop treatment produced a greater biomass yield than a barley monocrop. Protein concentrations of whole plants at silage-stage were 14-19% for fababean and 6-9% for barley and triticale. The addition of nitrogen fertilizer at a rate of 100 kg N ha^{-1} decreased the fababean component of intercrops. Seeding the fababeans prior to cereals increased the fababean component of intercrops. In 2003, intercrops with fababeans seeded in separate rows had greater yields than intercrops with fababeans and cereals seeded in the same row. In 2004, intercrop yields did not differ with fababeans seeded in separate rows versus in same rows with cereals. Further research under conditions of normal rainfall is needed to adequately assess the potential of intercropping fababeans with barley and triticale for forage in west-central Alberta.

P191. Impacts of a preceding oat crop on stem rot of canola and the pathogen *Sclerotinia sclerotiorum*

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Research suggests that oats may reduce pathogen populations and disease incidence in subsequent crops in rotation; however, impacts on sclerotinia stem rot of canola caused by *Sclerotinia sclerotiorum* have not been reported. Field studies were conducted in Manitoba from 2001-04 to determine the impact of preceding crop management on the survival of sclerotia, and on stem rot incidence, yield and quality of canola. Treatments consisted of oats managed as green manure

(tilled or mowed) or greenfeed harvested at heading, and oats and wheat managed as a grain crop (straw removed or returned, tilled or untilled post-harvest). Subplots consisted of +/- fungicide during the canola year. Microplot studies were also conducted from 2001-03 to compare the effects of oat cultivars to wheat, managed as green manure or grain, on the survival and germination of sclerotia.

In 2002 and 2003, environmental conditions were not favourable for stem rot, and treatments had little effect on disease, yield and quality of canola. However, sclerotia levels measured in soil before and after the preceding oat treatments showed the largest losses in pathogen viability were in tilled green manure treatments. In 2004, preceding crop management influenced the total number of apothecia and germinated sclerotia, but disease levels did not vary significantly. Neither preceding crop management nor fungicide significantly affected canola yield in 2004, although fungicide significantly reduced sclerotinia disease, and increased (P=0.08) test weight at both sites. In the microplot trials, mean apothecia production for all oat cultivars managed as a green manure was less than for wheat in 2003. While results regarding effects of preceding crop on sclerotinia disease, yield and quality of canola were inconclusive, a trend of reduced numbers of apothecia and germinated sclerotia in tilled oat green manure treatments suggests that such agronomic practices may be useful in managing *S. sclerotiorum* in canola.

P192. Impact of six fungicide soil treatments on potato scab incidence in two potato soils.

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A great variety of soil treatments are being tested by various potato growers for control of potato scab [*Streptomyces scabiei*]. Some Ontario growers have been testing fungicides and have reported mixed results. It is difficult, however, to evaluate the validity of these results because so many variables can affect scab severity. At the request of some Ontario potato growers we evaluated some of these fungicides under controlled conditions. Soils from two commercial potato fields (sites V and B2; pH 6.1 and 7.6, respectively) with a history of potato scab were brought to our station, mixed with six fungicides separately, placed in micro-plots, potato cultivar Snowden planted, and the resulting scab severity determined. Treatments for each soil included a control, Blocker (76 L/ha), Omega (26.7 L/ha), Flint (3.1 kg/ha), Gavel (20 kg/ha), Reason (7.8 L/ha), and Quadris (6 L/ha). The rates used were those recommended by the suppliers of the fungicides. This experiment was set up in 2003 and again in 2004 with six replicates per treatment. None of the fungicides had any significant effect on potato scab in either soil in either year. Thus, these fungicides do not appear to be useful soil treatments for reducing scab, at least at the rates used in this study.

P193. Impact of seeding and disease initiation dates on seed yield and severity of mycosphaerella blight in field pea

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Mycosphaerella blight (Mycosphaerella pinodes) reduces field pea (Pisum sativum) production throughout Canada. Although disease severity depends to a great extent on environmental conditions, especially moisture, the plant growth stage at which the pathogen attacks also affects disease outcome. Field trials were conducted at Edmonton, AB and Morden, MB in 2004 to assess the impact of seeding date on blight severity in eight field pea cultivars. Foliar and stem disease severity was significantly greater where pea plots were seeded 3-4 wk earlier than in later-seeded plots. However, yield potential was reduced at later seeding dates. Field plots were established at Vermilion, Vegreville and Edmonton, AB in 2004 to determine the effects of disease initiation date on the severity of mycosphaerella blight on field pea and the potential to cause yield loss. Disease initiation in early July caused the greatest level of

disease severity. Disease severity was significantly reduced by a one-week delay in disease initiation, and was further reduced by an additional week of delay in disease initiation. However, delay of disease initiation beyond the third week of July did not result in further reduction in the disease level. Relatively dry weather may have mitigated disease pressure during late July and early August. Disease pressure caused a 13% reduction in yield in plots with the earliest disease initiation date at the Vegreville site, compared to plots where disease was initiated in mid-July. However, differences in yield were not statistically significant at the Vermilion and Edmonton sites.

P194. Genotypic differential spike sprouting in advanced yield trials of barley, triticale and winter wheat

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Pre-harvest sprouting of cereal grains can cause economic losses as it decreases grain quantity and quality. The objective of this study was to determine if there are genotypic differences in grain sprouting in the spikes of advanced lines of barley, triticale and winter wheat. In 2004, the cultivars were planted in 3-m by 8-row plots in three replicates arranged in a randomized complete block design in each test. Barley was planted in five separate tests comprising 103 entries, triticale in one test of 25 entries and winter wheat in one test of 20 entries. A sample of 10 spikes, mainly from the primary tillers, was harvested from each plot shortly after physiological maturity when grain moisture content was about 25%. The spikes were immediately dried in a forced air dryer at 37 ^{o}C for 72 h to reduce the moisture content to about 12%. The spikes were kept in a freezer at –20 ^{o}C until analysis, to reduce dormancy decay due to after-ripening process. From each replicate, a sub-sample of three spikes was placed in a rain simulator at 18 ^{o}C and saturated with humidity. Each day for 8 days, the spikes in the rain simulator were rated visually on a 1-5 scale (1= no visible sprouting, 5= 100% sprouted) taking into account the percentage of sprouted. The visual ratings were converted to spike sprouting indices (SSI) taking into account the promptness of spike sprouting. The genotypes were designated as resistant (R) to sprouting if they had a SSI range of 3.0 to 4.0; moderately resistant (MR) if 4.1 to 5.0; susceptible (S) if 5.1 to 6.0; and very susceptible (VS) if >6.0. There were genotypic differences in sensitivity to spike sprouting. In barley, the genotypes ranged between 3.1 (R) to 7.3 (VS). In triticale, the genotypes ranged between 3.2 (R) and 4.8 (MR) comparable to the winter wheat genotypes that ranged from 3.1 to 5.8. The cultivars Vivar and Xena consistently appear to have a R - MR dormancy to spike sprouting. The triticale and winter wheat genotypes had higher levels of resistance to spike sprouting than the barley genotypes. This was expected because in barley, rapid germination is one of the requirements of the malting industry.

P195. PlantProNet - A Canadian Plant Protection Network of Expertise

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This initiative represents an important step towards national coordination of our existing scientific capability and capacity to respond to the threat of introduction into Canada of invasive alien plants and plant pests and pathogens. Integration of science and technology efforts across government and academia to protect Canada's forests, crops and ecosystems through a coordinated national effort is an initiative that supports federal science vision and priorities. The aims of the Network are to strengthen national expertise and capacity for plant protection research and diagnostics. This would be achieved by sharing best practices and information, discussing plant protection issues with other experts, and jointly developing improved and standardized procedures.

P196. Real-time PCR analysis of pal, hmgr, pr-1 and pr-5 in potato inoculated with Phytophthora infestans (Mont.) de Bary genotype US-1 and US-8

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Defense responses were investigated in two potato cultivars with different responses to late blight, Russet Burbank (RB, susceptible) and Kennebec (KB, moderately tolerant), after inoculation with two P. infestans genotypes, US-1 (old, mildly aggressive) and US-8 (new, highly aggressive). The occurrence of four defense-related genes (pal, hmgr, pr-1 and pr-5) in the genome of RB and KB was assessed using southern blot analysis. Their expression in three leaf strata (local, proximal and distal) and different times post inoculation were evaluated using Real-time quantitative RT-PCR. Southern blot analysis indicated that the distribution of these genes was reasonably conserved between the two cultivars and multi-copies of the same gene were present in both cultivars. Real-time PCR analysis showed that the activation of these defense-related genes was different in each cultivar and was affected not only by genotypes of P. infestans, but also by the proximity from the inoculation site. These genes were induced earlier in KB than in RB and in response to US-1 as compared to US-8. In general, the earliest and strongest induction of these genes was observed in KB inoculated with US-1. Furthermore, induction of pal and hmgr seemed to be suppressed at the site of infection while such possible suppression was not observed with pr-1 and pr-5. Our results showed that changes in either the activation or possible suppression of defense responses were related to late blight severity in the tested potato cultivars.

P197. Exploring biocontrol treatments for the management of potato Verticillium wilt

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Verticillium wilt is a major soil-borne constraint to potato cropping worldwide, and is considered as the main component of the potato early dying syndrome (PED). In the past few years, PED has been recognized as one of the most concerns to Canadian potato growers. Managing such a complex disease requires the use of costly noxious methods such as fumigation, along with cultural practices. However, even such methods don't provide a complete disease control. Therefore, more reliable, eco-friendly and low-cost alternatives are needed. In our study, we assessed the use of biocontrol agents for the control of *V. dahliae*. As a first step, a collection of *V. dahliae* isolates from potato fields in Manitoba was tested for the level of pathogenicity under controlled condition. The two most pathogenic isolates from these assays were used in further biocontrol studies. They were challenged in vitro, on nutrient media, to 18 bacteria previously reported to display either antagonistic or host defense-inducing abilities. Three of the tested bacteria have shown a strong inhibition of *V. dahliae* in vitro. They were further included in growth room and field trials using two cultivars 'Russet Burbank' (moderately susceptible) and 'Kennebec' (highly susceptible), during two growing seasons, in two different locations. In these trials, were also included two other bacteria, previously shown to enhance plant defense responses, along with additional plant extracts treatments with potential biocontrol activity. Treatments were applied by seed-coating before sowing. Over all the treatments tested in this study, one bacterium and one plant extract were shown to significantly and reliably reduce Verticillium wilt on 'Russet Burbank' and 'Kennebec', respectively.

P198. Evaluation of field pea cultivars for resistance to root and stem rot and seedling blight caused by *Rhizoctonia solani*

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Rhizoctonia solani Kühn is a persistent and major cause root and stem rot of field pea (Pisum sativum L.) in Alberta. Substantial losses in plant populations and yield have been reported. Twenty-eight varieties/lines of pea were evaluated for reaction to Rhizoctonia infection by assessing seedling survival, root rot, seedling dry weight, and seedling height in one greenhouse trial, and seedling survival, root rot, and seed yield in two field trials conducted in 2004. The varieties/lines

comprised three major types: 10 brown/mixed peas, 9 yellow peas and 9 green peas. 'JI2117' and 'PI6922' consistently showed the lowest infection levels and highest seedling survival. Varieties/lines with high susceptibility to Rhizoctonia infection included 'JI188', 'Mozart', 'Minuet' and 'JI1252'. Among the three types of peas studied, the brown/mixed peas showed the lowest infection levels and highest seedling survival, while the yellow peas were the most susceptible to seedling blight and root rot caused by R. solani.

P199. DON Content in a Long Term Barley Monoculture System: Effect of Tillage Practices

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Six tillage systems in barley (Hordeum vulgare L.) monoculture have been studied to compare chisel and no-till with conventional tillage. The experiment, conducted at the Research Farm of Agriculture and Agri-Food Canada at Normandin (Quebec), was initiated in 1990. The treatments were: T1: Conventional (fall molboard plowing and spring harrowing – 2 passes with a cultivator); T2: Chisel (fall) and spring harrowing (2 passes with a cultivator); T3: Chisel (fall) and spring harrowing (1 pass with a cultivator); T4: No tillage (fall) and spring harrowing (1 pass with a rotative harrow); T5: No tillage (fall) and spring harrowing (1 pass with a cultivator); T6: No-till (no tillage the previous fall and no harrowing in spring). Treatments were laid out in a complete randomized block design with four replications. Plot size was 10 m X 10 m. Barley seeding rate was 170 kg ha^{-1}. Because fusarium head blight (FHB) has become the most important cereal disease in Northern Quebec, DON content was measured in 2003 and 2004 to determine the effect of soil tillage on FHB incidence in barley. In 2003, mycotoxin content varied from 2.2 to 7.4 ppm. DON content was higher for no-till treatment (T6) than for other treatments (T1, T2, T3, T4, and T5). In 2004, the treatments had no significant effect on DON content. Because fungus that caused FHB survives on residue left on soil, and according to the results of this trial, tillage practices that bury cereal residue could be used to reduce the amount of inoculum.

P200. Competitive ability of historical and modern Canadian Western Red Spring (CWRS) wheat varieties under conventional and organic management

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Competition from weeds plays a role in reducing grain yields in conventional and organic management systems. Numerous studies have reported genotypic differences in the competitive ability of wheat, thus varietal selection may be a useful tool in preventing yield loss. Plant height, tillering capacity, and elevated PAR interception are a few of the traits thought to play a role in conferring competitive ability. It is also thought that crop varieties developed before the advent of modern, high-input agriculture may be better suited to lower soil nutrient levels and elevated weed competition. Twenty seven bread wheat varieties representing 114 years of Canadian wheat breeding were grown at four locations in Alberta under both conventional and organic management in 2002, 2003 and 2004. Overall conventional yields were 63% higher than organic yields and overall weed biomass was significantly higher under organic management. The highest yielding variety under conventional management was Sinton, while Park was the highest yielding under organic management. Earlier flowering and early maturity were found to be more important for achieving high grain yield on organic land than on conventional land. Greater numbers of spikes m^{-2} were also found to be associated with increased grain yield on organic land, but were not on conventional land. Increased plant height and early maturity were found to be associated with reduced weed biomass. High early season vigour was related to increased yield, increased spikes m^{-2} and reduced weed biomass on organic land. A competitive crop ideotype for spring wheat in organically managed systems may include fast early season growth, early maturity, high tillering capacity and tall height.

P201. Comparison of organic and inorganic nitrogen fertilizer on the morphology and

anatomy of industrial fibre hemp (*Cannabis sativa* L.) grown in Northern British Columbia, Canada.

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The effect of organic and inorganic nitrogen fertilizer on the morphology and anatomy of *Cannabis sativa* var. fédrina was investigated in both a greenhouse and field setting in Northern British Columbia. Plots (90 stems/m^2) treated with 0, 75, 150 or 300 kg N/ha of inorganic nitrogen or fishmeal, bloodmeal or sea star organic fertilizer were also replicated with 90 kg inorganic P_2O_5/ha application. The application of 150 and/or 300 kg N/ha of any nitrogen fertilizer type benefited field-grown plant morphology, secondary phloem fibre and xylem development, while greenhouse-grown plant morphology, secondary phloem fibre and xylem were positively influenced by 90 kg P_2O_5/ha. Primary phloem fibre characteristics of both greenhouse and field-grown plants were positively affected by the absence of nitrogen or phosphorus fertilizer. This study determined that organic can be used in place of inorganic nitrogen fertilizer for the production of a majority of fibre characteristics of *C. sativa* var. fédrina.

P210. Wild-harvested and cultivated commercial mushrooms of the Republic of Korea and British Columbia

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"Non-timber forest products (NTFPs) are botanical and mycological products and associated services of the forest other than timber, pulpwood, shakes or other wood products. Examples include wild mushrooms, floral greenery, craft products, herbs, ethnobotanical teaching and forest tourism." Centre for Non-Timber Resources, Royal Roads University.

As mycological non-timber forest products, such as pine mushrooms (matsutake), chanterelles, and morels, gain importance in British Columbia, there is a growing need for reliable information to guide the development of the wild-harvested commercial mushroom industry. At the same time, deregulation of the marketing of cultivated commercial mushrooms in BC is creating opportunities and challenges for that industry and a need for further research and development. Much of this research is best done here under local conditions, but we can also look to other countries with different traditions but similar ecosystems for opportunities and cautions. The Republic of Korea is such a country; it is an important producer of the commercially important and wild-harvested matsutake as is BC, yet it grows many more mushroom species commercially, and mushrooms are much more important than in BC. In this study we explore the similarities and differences in commercial mushroom production in these two jurisdictions and try to distill from this some direction for this industry in BC.

P211. The University of Alberta Microfungus Collection and Herbarium – a Canadian Microbial Resource Centre Conserving and Supplying Mycorrhizal and Other Fungi

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The roles of microbial culture collections are to collect, conserve, and characterize microorganisms and to distribute isolates and information about them to foster research and training. The University of Alberta Microfungus Collection and Herbarium (UAMH) is a specialized, general service culture collection of filamentous fungi that is part of a Canadian network involved in the conservation and supply of microbes worldwide. Established in 1960, the UAMH now holds over 10,500 accessions of living isolates and dried specimens representing more than 3200 species. The UAMH holds the largest collections of mycorrhizal and root associated fungi established from different habitats, regions and hosts in Canada. These have been built up through the inhouse and external research of many Canadian scientists and their students. Our rich collection of ericoid mycorrhizal fungi represents only part of the enormous and still largely unexplored diversity within this community. Ectomycorrhizal inoculants established over many years of research in the boreal and montane regions of Alberta are of value in regenerating forests in

sites such as the Alberta tar sands. Isolates obtained from orchids are used in symbiotic seed germination of threatened species. Other significant collections include stain and decay fungi, fungi associated with insects (alfalfa leafcutter bees, mountain pine beetles), and from specific habitats or substrates (soil, dung, building materials etc). In addition, the UAMH is renowned for the depth of its collections of medically important fungi (pathogenic, opportunistic, toxigenic, allergenic fungi). Information on cultures and services is available through the website [devonian.ualberta.ca/uamh] that includes web-based and print 2005 Catalogues.

P212. The effect of conditionally dispensable chromosomes on rhizosphere colonization by the fungus Nectria haematococca MPVI

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Nectria haematococca Berk. & Br. mating population (MP) VI (anamorph Fusarium solani Mart.) is an ascomycetous filamentous fungus that exists in various plant associations including those of plant pathogen as well as rhizosphere colonizer. *N. haematococca* MPVI possesses "conditionally dispensable" (CD) chromosomes that are not needed for axenic growth, but carry genes that appear to affect the habitats which individual isolates can occupy. Previous results in our lab show that *N. haematococca* isolates highly virulent on pea (*Pisum sativum* L.) are able to utilize homoserine, an amino acid found in pea root exudates, as a sole carbon and nitrogen source whereas isolates that are not pathogenic on pea are rarely able to utilize this compound. The gene(s) for homoserine utilization (HUT) is located on a CD chromosome of this root rot pathogen. We used a replacement series approach to determine if the CD chromosome increases the ability of *N. haematococca* to colonize plant rhizospheres. Tomato, pea, and *Medicago truncatula* plants were grown in soil inoculated with pairs of *N. haematococca* isolates that were isogenic except for CD chromosomes. The rhizospheres were evaluated for the degree of colonization by each isolate using real-time PCR. Preliminary results indicate that rhizosphere competence is increased in isolates with a CD chromosome.

P213. *Cryptosporiopsis ericacea* and *C. rhizophila* dark septate endophytes (DSE) from the roots of trembling aspen

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A series of ascomycetes with melanized hyphae are common in the roots of a large number of plants and comprise an artificial group known as the dark septate endophytes or DSE. These fungi are apparently neither mycorrhizal nor pathogenic. Species of *Cryptosporiopsis* are best known as pathogens of woody plants (especially fruit trees and ornamentals) but several species are reported as apparently harmless endophytes in roots and may be included among the DSE. Recently we obtained numerous isolates of this genus from the apparently healthy, fine roots of *P. tremuloides*. Ten isolates, from 3 different, forested sites in central Alberta, were identified as *Cryptosporiopsis ericacea* Sigler, a recently described species from the roots of ericaceous plants collected in western North America. This species produces phialides on simple conidiophores that are solitary or borne in sporodochial tufts and produce aseptate, cylindrical, slightly curved, macroconidia (22-35 x 6.2-7.5 µm) and microconidia (3.5-6 x 1.5-2 µm). Setae are often diagnostic for these fungi but are uncommon in this species. Comparisons of 5.8S nuclear rDNA and flanking internal transcribed spacers ITS1 & 2. were used to match many of our isolates with *C. ericacea*. Two isolates of a second species, *C. radicicola*, a taxon described originally from the roots of oak in Europe, were also identified from our collections of P. tremuloides roots and was recognizable on the basis morphological characters of cultured material. Our sequences also support the suggested placement of both species in the inoperculate genus *Pezicula*.

P215. Wheat non-specific lipid transfer proteins (ns-LTPs) are inhibitory to fungal plant pathogens.

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Although many non-specific lipid transfer proteins (ns-LTPs) have been found in plants,

their functions are not understood. Two main families of ns-LTPs, ns-LTP1 (ca. 9 kDa) and nsLTP2 (ca. 7 kDa), are known. Twenty one cDNA clones corresponding to the ns-LTP1 gene were differentially expressed in cDNA libraries of wheat infected with *Tilletia tritici*, or subjected to low temperature stress, including eleven full-length different ns-LTP1 clones, all possessing coding sequences of 348 bp. Their coding regions were highly conserved with 78-99 % and 71-100 % identity at the nucleotide and amino acid levels, respectively. The deduced proteins coded by the eleven cDNAs have molecular masses of about 9 kDa with 115 amino acids, and contain four pairs of conserved disulfide bonds, characteristic of ns-LTP family. To study their antifungal activity, 8 ns-LTP1s selected among 22 full-length wheat ns-LTP1 from our own databases and GenBank were cloned into an expression vector and transformed into *Pichia pastoris* strain GS115. Western blots confirmed the expression and the molecular weight of the fused ns-LTP1s. In vitro lipid binding activity for all ns-LTP1s was demonstrated. The various ns-LTP1s differentially inhibited spore germination and mycelium growth of the fungal plant pathogens *Botrytis cinerea, Sclerotinia sclerotiorum*, a Low Temperature Basidiomycete, *Typhula incarnata, Fusarium graminearum, Verticillium dahliae, Puccinia graminis, Puccinia recondita, Tilletia tritici, Pyrenophora tritici-repentis,* and *Stagonospora nodorum*. These results demonstrated that the different ns-LTP1s are differentially toxic to a wide range of plant pathogens and may be involved in specific defence functions in wheat.

P216. Water stress and the *Septoria musiva*-hybrid poplar pathosystem.

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Short rotation high yield hybrid poplar plantations are becoming more frequent in North America as demands for pulp and fiber increase. *Septoria musiva* Peck. a fungal pathogen of hybrid poplar has caused extensive damage and occasional plantation failure in Canada and the United Sates. Greenhouse inoculations may be useful to screen for resistant individuals. In order to test the applicability of greenhouse inoculations to field conditions an experiment was conducted to evaluate the importance of the genotype by environment interaction in determining canker development. Four clones of hybrid poplar and four isolates of *S. musiva* were inoculated in all possible combinations and then exposed to either [1] water stress (soil water content of 10%) [2] no stress (soil water content >35%). Height and root collar diameter differed between stressed and unstressed trees indicating that the water deficit affected tree growth. However, there were no two- or three-way interactions involving clone, isolate or the clone by isolate interaction, and water stress. These results imply that greenhouse inoculations may be useful for selecting clones resistant to Septoria canker.

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P217. Variation in *Cylindrocarpon destructans* isolates causing root rot of American ginseng (*Panax quinquefolius* L.).

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The fungus *Cylindrocarpon destructans* (Zins) Scholten can cause extensive root rot (disappearing root rot) of American ginseng in Canada. Isolates of *C. destructans* collected from typical disappearing rot lesions were divided into highly and weakly virulent groups depending on their ability to cause lesions on wounded and unwounded roots. Among the 80 isolates recovered, 51% were weakly virulent (WV) and the remaining 49% were highly virulent (HV). The 80 isolates were tested using PCR primers (CdU3 5'-GACGATTCGGGCCGTATCTGTG -3' and CdL1b 5'-CAGCGGCG CCCACTAACAAC-3'). PCR amplicons were obtained only from isolates which were categorized as HV based on the root inoculation test. None of the WV isolates showed any amplification product. After 2 weeks of growth on PDA, HV isolates became dark to rust brown in colour with a velvety colony surface, whereas WV colonies were beige to light brown with cottony aerial mycelium. Spore production on PDA by 3 WV isolates was significantly higher compared to 3 HV isolates. Out of three WV isolates, two did not produce any detectable pectinase and one produced a significantly lower amount (0.48 units/mg dry mycelial weight) compared to HV isolates. All 3 HV isolates produced pectinase levels ranging from 2.63 units in isolate #1 to 1.38 units for isolates #16 and #17. Polyphenoloxidase activity was also markedly different between the two groups of isolates. Consistent with the levels of pectinase

production, highest PPO was produced by isolate #1 (72 units) followed by 56 and 55 units for isolates #16 and #17, respectively. Direct penetration of the epidermis by HV isolate of C. destructans hyphae was observed by histopathological analysis. Hyphal growth from the point of infection occurred mainly intracellularly and extended down through 7-8 cell layers. Higher enzyme production by HV isolates appears to facilitate direct penetration of ginseng root cells by *C. destructans*.

P218. Utility of a cultural method for identification of the ericoid mycobiont *Oidiodendron maius* confirmed by ITS sequence analysis

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Correct identification of the mycobiont is essential to gaining a true understanding of the species involved in forming mycorrhizas. Identification of *Oidiodendron* species is often difficult, in part due to variation in both colonial and microscopic features when isolates are grown under differing conditions. *Oidiodendron* maius is reliably reported to form ericoid mycorrhizas, but some reports concerning other species have been based on misidentified isolates. A simple cultural method was investigated for its reliability in distinguishing *O. maius* from selected other species of Oidiodendron. Forty three isolates belonging to different species were grouped by morphology after 28 days growth on cereal agar overlaid with a cellophane membrane. All isolates of *O. maius* and its close relative *O. citrinum* expressed characteristic colonial morphologies allowing recognition regardless of sporulation. Isolates grouped by colonial features correlated with strongly supported groupings obtained by analysis of nuclear ribosomal internal transcribed spacer (ITS) region sequences.

P219. Two new Cryptosporiopsis species from roots of ericaceous hosts in western North America

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Cryptosporiopsis species are coelomycetous anamorphs of ascomycetes in the genera *Pezicula* and *Neofabraea* (Dermataceae). These fungi are occasionally isolated from roots of woody plants but may be difficult to identify due to absence of sporulation. Some isolates obtained from roots of ericaceous hosts had previously been linked phylogenetically to *Pezicula* and, when regrown, revealed conidiomata and conidia typical of *Cryptosporiopsis* species. Cultural and molecular data allowed for the recognition of two new species. These species appear to be uncommon in roots of western North American ericoid plants as judged by their low recovery in culture by isolation from surface-sterilized roots and by the inability to detect them in direct PCR amplification from roots. Although both species have been obtained only from surface-sterilized roots, little is known about their roles in roots of ericaceous plants.

P220. Proteome-level analysis of *Brassica napus* following infection with *Alternaria brassicae*

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Interactions between plants and pathogens are specific, complex and dynamic. Pathogen invasion triggers recognition and response in the plant leading to the induction of signaling cascades and the up- or down-regulation of numerous proteins involved in the interaction that in turn may lead to resistance, or susceptibility of the plant. The main objective of this study was to identify the proteome-level changes that occur following infection of a blackleg-resistant *Brassica* doubled-haploid line (generated from an interspecific cross between *B. napus* and *B. carinata*) by *Alternaria brassicae* (causal agent of alternaria blackspot of canola). Proteomics-based analysis was performed to gain insight into the molecular basis of disease induced processes for the comprehensive understanding of the molecular events mediating plant-pathogen interactions. Two-dimensional gel electrophoresis followed by tandem Mass Spectrometry led to the identification of 18 differentially-expressed proteins. The biochemical significance of our findings and

the potential roles of these proteins during the compatible interactions are discussed.

P221. Two dimensional display of phosphorylated and glycosylated proteins in the Fusarium head blight infected spikes of wheat

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Fusarium head blight (FHB) is a very destructive fungal disease of wheat. Although FHB resistance has been well documented and partially resistant cultivars have been developed to reduce incidence and severity of FHB, there is a limited understanding of the molecular mechanisms involved in plant resistance against the infection and spread of Fusarium graminearum. In our current study, a proteomic approach was performed to investigate protein phosphorylation and glycosylation in relation to FHB infection and spread in the spikes of wheat cultivars with various FHB resistance levels. Two dimensional gel electrophoresis and three compatible protein stain technology were used to directly display and quantitatively compare phosphorylation and glycosylation patterns between protein samples from FHB infected and sterile spikes. Using this technology, phosphorylated proteins, glycosylated proteins from a single sample are sequentially stained by Pro-Q Diamond phosphoprotein gel stain and Pro-Q Emerald glycoprotein gel stain. Different patterns of protein phosphorylation and glycosylation were observed between protein samples from FHB infected and sterile spikes and between spikes of cultivars with different FHB resistant levels as well. Proteins showing significant modification following FHB infection were excised for analysis by LC-MS/MS. Results will be discussed in function of differential post translational modifications and level of resistance to FHB.

P222. The Efficacy of Reduced Risk Fungicides and the TOMCAST Disease Forecasting System as Tools for Control of Septoria Late Blight of Celery

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Septoria late blight (*Septoria apiicola*) is a common, economically important disease affecting celery (*Apium graveolens*) leaves and petioles. Integrated management of the disease could be improved by using reduced-risk fungicides and disease forecasting. Field trials were conducted in 2003 and 2004 to determine the efficacy of reduced- risk fungicides, calcium chloride and a foliar fertilizer, Alexin, in controlling the disease on two celery cultivars. In 2004 an additional trial was carried out to evaluate disease forecasting based on the TOMCAST system for tomato leaf blight. Trials were inoculated by spreading diseased celery tissue between rows.Severity of leaf disease, petiole disease, and yield were assessed at harvest. . In 2004 disease progress was monitored weekly by assessing % leaf area diseased. In 2003, Quadris (azoxystrobin 23%) and BAS 516 (pyraclostrobin 12.8%, boscalid 25.2%) were most effective at reducing disease incidence, petiole disease severity index (DSI) and leaf blight severity. In 2004 petiole DSI was lowest on celery treated with BAS 516, Quadris, Cabrio (pyraclostrobin 20%), and Bravo 500 (chlorothalonil 50%). Alexin treatment reduced petiole DSI and percent diseased petioles compared to the untreated check. Trimmed weight of the celery decreased as DSI increased ($r = 0.20$, $P < 0.03$). The threshold treatment of 10 disease severity values (DSVs) under the TOMCAST system using BAS 516, resulted in lower petiole DSI than the check, calendar spray program (Bravo 500), and 20 DSV treatment (Bravo 500).

P223. Testing of *Xylella fastidiosa* in chemically defined media indicates that xylem chemistry may play a role in biofilm formation and disease susceptibility

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The bacterium *Xylella fastidiosa* (XF) causes Pierce`s disease (PD) in grapes and various diseases in other hosts. Despite the wide host range, the mechanism for symptom development is similar. XF is injected into the

plant vessels by xylem-feeding sharpshooters. They may remain as free cells (planktonic) or small clusters until adhering and forming complex biofilms. Biofilms are microbial communities attached to surfaces, physiologically distinct from planktonics and showing enhanced resistance to antimicrobials. Biofilms can plug the xylem, causing symptoms, and their development is highly influenced by the xylem chemistry of the hosts. We compared the growth of grape strains of XF as planktonics or biofilms in three chemically-defined media, XF-26 (22 components), CHARD2 (10 components) and 3G10R (9 components). PW^{+}, a rich non-defined medium, was used as control. Media were inoculated with standardized cell suspensions in glass tubes, and wooden matrixes were added as surfaces for biofilm formation. Cultures were incubated during a period of 9 days, under shaking, and both planktonic growth and biofilm formation were sampled at time intervals (2, 4, 7 and 9 days) by cell counts. Biofilm formation was also compared by examining size and structure. CHARD2 was by far the best medium capable of inducing biofilm formation, followed by PW, XF-26 and 3G10R. CHARD2 and XF-26 also differ in planktonic growth; CHARD2 exhibited no planktonic growth, whereas XF-26 cultures were predominantly planktonic.CHARD2 is unique because it contains the reducing agent cysteine (0.01 g/L); XF-26 has cysteine as well (0.2 g/L), but it has 16 additional amino acids, sodium citrate and dissodium succinate. We hypothesize that the differences observed are related to the redox environment in different formulations, reinforcing the contention that xylem chemistry in resistant and/or susceptible plants may play a significant role in determining biofilm formation and PD susceptibility.

P224. Suppression of early blight (in vivo) and germination of *Alternaria* spp. conidia (in vitro) with the novel fungicide, azoxystrobin

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Experimental potato fields were established in New Annan, Prince Edward Island, to assess the efficacy of strobilurin fungicides (Quadris® and Headline®) to suppress *Alternaria solani*, causal agent of potato early blight. At the end of the growing season, these field trials were rated according to the Horsfall-Barrett scale (a qualitative assessment of defoliation) and an obvious fungicide treatment effect was observed. The incidence and severity of early blight was low in plots treated with strobilurin fungicides compared to untreated plots where disease was more severe. These ratings also revealed differences in disease based on cultivar type and nitrogen availability, although these differences were less marked in comparison to the fungicide treatment. In addition to the foliage ratings, isolates of *A. solani* as well as *Alternaria alternata*, another potato leaf spot pathogen, were collected from the experimental trials and several other potato fields for subsequent laboratory work. An in vitro spore germination assay was used to measure sensitivity of these isolates to azoxystrobin, the active ingredient in Quadris® fungicide. The effective concentration that inhibited spore germination by 50% (EC_{50}) was determined for each isolate. EC_{50} values ranged from 0.003 to 0.014 parts per million (ppm) for A. solani while the values for A. alternata ranged from 0.001 to 0.023 ppm. These results suggest that the isolates tested are sensitive to azoxystrobin and there was no indication of resistance. This sensitivity is likely due to the limited exposure of these two pathogens (*A. solani* and *A. alternata*) to strobilurin chemistry in Prince Edward Island potato fields.

P225. Stripe rust resistance in Iranian wild wheat (*Triticum boeoticum*)

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Twenty-five germplasm lines of wild wheat (*Triticum boeoticum*, diploid wheat, A genome) populations originating from the warmer, non-stripe rust regions of western Iran (Fertile Crescent) and five germplasm lines originating

from the high mountain, stripe rust regions of northwest Iran. The lines were classified on various floristical (ecological) characters using the ANAPHYTO software, which combines correspondence factorial analysis with cluster analysis. The 30 populations were classified into seven ecological groups; the five germplasm lines originating for the high mountain rust areas were classified into a single group while the 25 lines originating from the non-rust areas were distributed among the other six floristical groups. Lines originating from rust and non-rust regions were evaluated for stripe rust resistance at the two-leaf stage under controlled environments and in the field using two different isolates. The five lines originating from the high mountain, rust regions were rated as resistant while seven lines originating from the non-rust regions were rated moderately susceptible to susceptible. Seven germplasm lines, one from each of the seven ecological groups were grown under controlled environment conditions and subjected to a phenol extraction protocol. A large green phenolic spot was consistently associated with the rust-resistant lines while three smaller phenolic spots were associated with the rust-susceptible wheat lines following two-dimensional thin-layer chromatography on D.C.6 gel. The potential role of phenolic substances in stripe rust resistance of these two ecological groups of T. boeoticum will be discussed.

P226. Effect of crop management on Sclerotinia rot, canopy rowth and yield of carrots in Prince Edward Island.

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Field trials were established in Brookfield, Prince Edward Island to examine the impact of crop fertility, seeding rate and canopy mowing on yield and the incidence of Sclerotinia rot in carrots. Increasing the applied rate of nitrogen increased total and marketable carrot yield, canopy mass and subsequent disease development in petioles and carrot roots. Application of a nitrogen top-dress at mid-season had no affect on yield or disease severity. Lobster processing waste was not adequate as an alternative source of nitrogen for plant growth and yield. Increasing seeding rate from 14 to 34 seeds/ft increased canopy mass as well as the severity of Sclerotinia rot in the foliage and roots. Marketable carrot yield was maximized at 25 seeds/ft and declined on either side of this optimum rate. Lateral clipping of the carrot canopy at row closure significantly reduced the severity of Sclerotinia rot in the foliage as well as in stored carrots, although total and marketable yield was also reduced. These studies suggest that managing carrot canopy by adjusting crop fertility, seeding rate and with canopy clipping may be viable methods for controlling Sclerotinia rot of carrots.

P227. Potential alternatives to CIPC for potato sprout suppression and disease control

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Potential CIPC (chloropropham) alternatives on potato sprout and fungal disease suppression were evaluated in stored potatoes and in vitro. Study 1: (+), (-) carvone, diallyldisulfide (DDS) of garlic, dill, caraway and garlic seed extracts on sprout growth in potatoes under 20, 12 or 10^{o}C storage for 4 months in each of four years. Study 2 compared (+), (-) carvone and DDS on growth of Fusarium sambucinum, F. solani, F. culmorum, Sclerotinia sclerotium, and Rhizoctonia solani under in vitro conditions at 23^{o}C. Study 3 focussed on (-) carvone and its efficacy on fungal control under both in vitro and in vivo conditions at 4^{o}, 10^{o} and 23^{o}C. Carvone can be applied in powder, aqueous or vapour form to suppress sprouting and sprout number without affecting tuber weight loss in 'Norland', 'Russet Burbank' and 'Snowdon' potatoes. Both (+) and (-)-isomers of carvone were active in sprout suppression and all three methods of treatment application were effective in controlling sprout growth at all three storage temperatures. Liquid and powder forms of carvone were more effective in limiting sprout growth than the vapour form. DDS was more effective than carvone in vapour form. A double application of DDS at 60 ml/m^{3} completely inhibited sprout growth for at least 12 weeks at 10^{o}C and was equivalent to CIPC

and maleic hydrazide. None of the carvone treatments produced an unacceptable flavour in baked, boiled or chipped potatoes. In the second study, (+), (-) carvone and DDS treatments similarly inhibited *Fusarium solani* growth but (-) carvone was the most effective on suppressing *F. sambucinum, F. culmorum, Sclerotinia sclerotium* and *Rhizoctonia solani*. In the third study, (-) carvone treatments significantly suppressed all diseases tested (*S. sclerotiorum*, *F. sambucinum* and *R. solani*) at all temperatures in vitro. After 6 weeks at 10^{o}C, (-) carvone significantly inhibited depth of infection F. sambucinum growth in inoculated potatoes by 30-40%.

P228. Pathogenicity of *Fusarium oxysporum lini* isolates from flax.

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Wilt caused by *Fusarium oxysporum* Schlechtend f.sp. *lini* (Bolley) Snyder and Hansen, is a worldwide disease affecting flax (*Linum usitatissmium* L.) and causing major losses in yield and quality. Ninety-five isolates of this pathogen obtained from infected flax crops in Manitoba, Saskatchewan and North Dakota, were characterized for pathogenicity on forty flax cultivars with diverse origin. Eighteen isolates were characterized as highly virulent with >60% disease incidence and >15% reduction in plant height. These isolates were more virulent than all isolates recovered from the fusarium wilt testing nurseries established in Manitoba and Saskatchewan. Sixteen of these isolates were from a single field near Treherne, Manitoba, and had been previously characterized as the vegetative compatibility group (VCG) 0441. Testing results showed that VCG 0441 is highly virulent, similar to a virulent isolate from Europe. Such isolates should be used in screening for resistance in the Canadian flax breeding programs to ensure that future flax cultivars have resistance to the most virulent isolates of this pathogen.

P229. Pathogenic and molecular diversity in populations of *Ascochyta lentis*

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Isolates of *A. lentis* were obtained from 1978 to 1985 and from 2001 to 2004 for a comparative study of pathogenicity and molecular diversity. A total of 65 isolates were tested on 16 lentil genotypes that potentially contain different sources of resistance. Thirty-one isolates originated from pre-1986 and 34 isolates came from recent collections. On average, isolates from post 2000 caused more disease on lentil than those from pre-1986 with the exception of Indianhead and Richlea. The magnitude of the difference in disease caused by the two populations varied. Largest differences were seen on 1156217a, ILL 1704, ILL 4605 and PI 468901. Significant effects of lentil genotypes, isolates and the interaction between both factors were observed and contributions of isolates, genotypes and the interaction were comparable suggesting that ranking of lentil genotypes based on resistance differed among isolates. Plotting relative disease of isolates for each lentil genotype showed a continuum rather than distinct groups though a few single isolates of both populations caused distinctly more disease on some lentil genotypes compared to most other isolates. Comparing the reaction of each isolate on all lentil genotypes also showed a more or less pronounced continuum in the amount of disease caused, though isolates differed in the overall amount of aggressiveness, and in the specific response of lentil genotypes. Molecular analysis of isolates by RAPD-PCR revealed high levels of similarity (= 90 %) among isolates collected more than 20 years apart. Isolates from different time periods grouped separately with a few exceptions. Some isolates were identified with identical banding patterns that originated from different years or different locations, suggesting that isolates may persist in the population in time and space.

P239. Effect of stress on nahG gene in Wheat

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Transgenic wheat lines were created with nahG in a Blizzard winter wheat background.

The nahG encodes for salicylate hydroxylase and it's presence will destroy salicylic acid in the cells. This should render the plants more susceptible to disease as salicylic acid is involved in SAR (systemic acquired resistance).The population was then tested for oxidative stress using exposure of germinating seed to high salt (NaCl), or high sugar (mannitol). The results of the oxidative tests were compared to PCR tests on the seedlings, confirming the presence of the nahG. Disease tests were performed on a selected few of the population. Preliminary data indicate plants with nahG construct are more susceptible to disease.

P240. New perspectives on white pine blister rust

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A century after its introduction in North America, the white pine blister rust outbreak has yet to stabilize. After having severely depleted important timber species, it is now endangering key ecological species with critical importance for biodiversity, ecological succession, watershed protection, and wildlife food and cover. Molecular epidemiology has revealed a high genetic differentiation between eastern and western rust populations. Analysis of worldwide collections has shown higher diversity outside America, possibly including other species. Interspecific hybridization can serve as an evolutionary fast track as for devastating forest pathogens such as Annosus root rot, Dutch elm disease, Phytophthora disease of alder, and poplar leaf rusts. A hybrid rust between white pine blister rust and comandra blister rust has recently been discovered in three limber pine (*Pinus flexilis* James) stands from Alberta. This hybrid has been characterized with regard to morphology, pathogenicity and genetic composition. In the hybrid, aeciospore morphology is intermediate between *C. comandrae* Peck. and *C. ribicola* J.C. Fisch. So far, inoculations of this hybrid on *Ribes* spp. failed to generate infections and was not found in nearby currant farms. Nevertheless, the hybrid has been found on two years, at frequencies up to 38% of the trees sampled, and aeciospores germinated on agar media. All hybrids were completely heterozygous at 12 loci examined so far, having one allele with complete DNA sequence identity with *C. comandrae* and the other allele with identity to *C. ribicola*. This indicates that the hybrid has probably been formed recently. On the other hand, we have recently begun research on association and gene expression studies for identifying alleles at candidate resistance gene loci, which could greatly accelerate white pine tree improvement. The strategy we will employ is to associate single nucleotide polymorphisms with specific host-pathogen interactions and resistance responses.

P241. Localization of fungal enzymes secreted by the European race of *Gremmeniella abietina* during infection of *Pinus contorta* and *P. banksiana*

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Very little is known about the ultrastructural features of the interaction between *Gremmeniella abietina* and its resistant hosts, *Pinus contorta* and *P. banksiana*. In this study, special attention was paid to the presence and localization of enzymes such as laccases, peroxydases, polygalacturonases and cellulases produced by the pathogen during the infection process. Examination of infected shoots incubated with a polyclonal anti-laccase revealed fungal polyphenoloxydases over the pathogen wall and over the extracellular sheath surrounding the fungus. With a monoclonal anti-peroxidase, labelling was found in the cytoplasm of the fungal cell, principally over an electron-dense region. Peroxidases were also found, slightly dispersed, surrounding the pathogen cell and over the host cell wall when they were in close contact. Similarly, with a polyclonal antibody raised against a fungal polygalacturonase, scattered gold particles were seen over host walls and inside the host cytoplasm when the fungus was surrounding host cells, but intense labelling was observed over the fungal wall. When an exoglucanase-gold probe was used, indirect involvement of cellulases was demonstrated by the observation of degraded host cell walls in the vicinity of the fungus. Control tests aimed at assessing the specificity of the probes resulted in little or no labelling. This study showed the ability of the European race of *Gremmeniella abietina* to produce laccases, peroxydases, polygalacturonases

and cellulases when infecting *Pinus banksiana* and *P. contorta*. Being produced in a context of resistance, those enzymes were, most of the time, found in close association with pathogen cells and no extensive degradation of host tissues was noted. As those enzymes were often localized over the extracellular sheath of the pathogen, it is concluded, however, that this sheath might play a significant role in cell wall degradation during colonization of host tissues.

P242. Inhibitory effects of organic and inorganic salts on the growth of *Erwinia carotovora*, a soft rot causative agent in potato tubers: Physico-chemical basis

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Soft rot (*Erwinia carotovora* subsp. *carotovora* and *Erwinia carotovora* subsp. *atroseptica*) is a major bacterial disease affecting potato (*Solanum tuberosum* L.) tubers. The disease causes significant economic losses worldwide especially during storage. Currently, no chemicals are available for an effective control of potato soft rot, and hence, control and management of this disease rely mainly on cultural practices. However, in most of the cases, cultural practices are not sufficient to control the disease effectively. In this study, 21 organic and inorganic salts were tested in vitro for their inhibitory effect on the growth of the bacteria *E. carotovora* subsp. *carotovora* and *E. carotovora* subsp. *atroseptica*. In nutrient broth medium, at 0.2M concentration, eleven of the salts tested exhibited strong inhibition of the growth of both bacteria. Among them, sodium carbonate, sodium metabisulfite, trisodium phosphate, aluminum lactate and aluminum chloride were bactericidal after one hour of exposure and exhibited low minimal inhibitory concentration (MIC) values (≤10 mM). Sodium bicarbonate, sodium propionate and ammonium acetate exhibited intermediary MIC values (20 mM) and a slower bactericidal effect except sodium propionate which was bacteriostatic. Aluminum dihydroxy acetate, potassium sorbate and sodium benzoate were bactericidal but exhibited MIC of 100 mM. It is shown that the inhibitory action of salts relates to the water-ionizing capacity of their constituent ions (low apparent pK, pK'_a or pK'_b values) and to their lipophilicity (partition coefficient, $P_{o/w}$), the latter mainly contributing to the action of preservative salts, potassium sorbate and sodium benzoate. The relationship between the inhibition of bacterial growth and the addition parameter ($pK' + pP_{o/w}$) was sigmoidal. Results of this study suggest a possibility for exploiting some of the effective salts as antimicrobial agents to control potato soft rot.

P243. Impact of oat plants, oat plant extracts and saponins on *Sclerotinia sclerotiorum*

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Research suggests that compounds in roots and shoots of oats may have the potential to reduce pathogen populations: however, impacts on the life cycle of *Sclerotinia sclerotiorum* have not been reported. Laboratory studies were conducted to determine the effect of oat plants, oat plant extracts, and the saponin ß-escin on mycelial growth, and production of sclerotia of *S. sclerotiorum*.

Oats are a unique crop in that they contain natural substances, including specific saponins, which may directly affect the development and growth of several fungal pathogens. In laboratory studies, growth of *S. sclerotiorum* was evaluated in media with 0, 10, 50, 250, 500, 1000, and 2500 ppm ß-escin, a saponin known to be produced in oat leaves and roots. Colony size of *S. sclerotiorum* was reduced at all concentrations of ß-escin with the greatest reductions occurring at 2500 ppm. Production of sclerotia was also affected with fewer but larger sclerotia produced at 2500 ppm ß-escin, as compared to the untreated control.

Impact of oat plant extracts on mycelial growth and sclerotia production of *S. sclerotiorum* was examined using oat extracts from root and leaves. Two treatments of ß-escin (50 and 1000 ppm) and an untreated control were included. Oat extracts had a direct effect on mycelial growth of *S. sclerotiorum*. which was intermediate between results observed with a high (1000 ppm) and low (50 ppm) concentration of ß-escin. Production of sclerotia was reduced in some cases with oat extract compared to the unamended control.

Volatile substances released from 4-week old oat roots and leaves incorporated into soil

and allowed to decompose for 5 days did not result in consistent reductions in mycelial growth of S. sclerotiorum. This suggests that exposure to volatile substances may not play a significant role in suppressing S. *sclerotiorum*

P244. Effects of *Phytophthora infestans* on the development of other tuber rots revealed using PCR

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Potato diseases such as late blight (*Phytophthora infestans* (Mont.) de Bary), pink rot (*P. erythrospetica* Pethybr.), leak or watery wound rot (*Pythium ultimum* Trow), bacterial soft rot (*Erwinia carotovora* subsp. *atroseptica* (van Hall) Dye), and dry rot (*Fusarium sambucinum* (teleomorph: *Giberella pulicaris*) (Fr.:Fr.) Sacc.), cause, alone or in combination, severe losses of potato stock in storage. Most of these pathogens (e.g. *P. infestans*) are carried from the growing season to the storage area through infected tubers or soil holding to them. Others are contracted during harvest or storage operations. Each of these pathogens is able to cause serious damage alone but severe losses are recorded when more than one pathogen is involved. Notorious pathogens such as *P. infestans* can be facilitating the infection by other secondary pathogens. Currently, before moving potato tubers into storage, only visual diagnosis is practiced in order to avoid potential disease development. However, no data are available about how tuber diseases develop and spread during storage. The aim of this study was to provide accurate data about the concomitant expansion of different diseases, especially when tubers are primarily infected with late blight pathogen. This work was carried out using artificial inoculations of high quality tubers incubated at 15^{o}C, in order to simulate conditions during the transition between harvest and long-term storage at lower temperatures. In this study, the development of tuber rots caused by individual pathogens or their combination was analyzed both on the skin and in the core of the tuber as well as at different time points after inoculation. The detection of pathogens was successfully achieved centimeters ahead of the expanding lesions. Based on the pathogen associations studied up to date, it has been found that the presence of *P. infestans* is enhancing the expansion of *E. carotovora* subsp. *atroseptica*, specifically in the tuber flesh, where the pathogen makes multiple galleries. However, interestingly, the development of *P. erythrospetica* and *P. ultimum* were found to be reduced probably due to their lower competitiveness in presence of *P. infestans*. On the other hand, infection by *P. infestans* had no effect on further development of *F. sambucinum, V. dahliae* Kleb. or *V. albo-atrum* Rienke & Berthier.

P245. Effect of seed treatments on control of dwarf bunt of winter wheat in Ontario.

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Dwarf bunt, caused by *Tilletia controversa* Kühn, is a destructive disease of winter wheat in Ontario. To determine the effect of seed treatments, trials were conducted with eight cultivars of winter wheat at two locations each year from 2002-2003 in central Ontario, under T. controversa inoculated field conditions. The seed treatments including three rates of Dividend XL RTA (difenoconazole + metalaxyl-M, 0.13, 0.26, and 0.39 g a.i. kg-1 seed), Vitaflo 280 (carbathiin + thiram, 0.83 g a.i. kg-1 seed), and bioagent ACM941, a strain of Clonostachys rosea (C. rosea ACM941, 2.5 x 106 cfu kg-1 seed). Untreated seed was used as controls. Dwarf bunt was observed in the untreated controls for all trials, at average of 12% in 2002 and 18% in 2003. All seed treatments significantly ($P < 0.05$) reduced disease incidence. The three rates of Dividend XL RTA were all highly effective, providing 98-99% control. Vitaflo 280 and ACM941 were partially effective, reducing disease incidence by 51 and 31%, respectively. All seed treatments increased emergence by 5-8%, which was significantly different from the untreated control. Although the data indicate a trend of higher yield (0.6-2.6%) and lower thousand seed weight (0.4-1.5%) with seed treatments over the untreated controls, the

differences were not significant at P = 0.05. The wheat cultivars differed in susceptibility, but all were considered moderately susceptible to the disease. There were no significant cultivar × treatment interactions in both disease incidence and emergence, suggesting that Dividend XL RTA is an effective fungicide for controlling dwarf bunt for all winter wheat.

P246. Does increased air temperature affect the cold hardening process of photosynthesis in *Pinus banksiana*?

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Temperature and day length are important drivers of physiological changes in boreal evergreen conifers since they determine the length of the growing season. Climate change and an increase of land surface air temperature are likely to increase the length of the growing season in the northern hemisphere. Evergreen trees might benefit from warmer air temperatures in early spring or late autumn, thus increasing the photosynthetically active period and net carbon gain. Our aim is to characterize the physiological mechanisms of the down regulation of photosynthesis in Jack pine under current and anticipated climate change conditions with increased air temperatures in autumn. Here we present results from a factorial experiment using controlled environment facilities to dissect the effects of day length (long day vs. short day) and temperature (20°C vs. 5°C). Day length did not affect photosynthetic capacity and electron transport in the 20°C treatments. However, under long day/cold temperature conditions pigment analysis revealed higher Chl a : Chl b ratios and fluorescence data an increased thermal dissipation of excitation energy as compared to the 20°C treatments, thus indicating increased excitation pressure on PSII. This was due to differences in thylakoid protein composition and in the enzymatic activity of the Calvin cycle. We conclude that in contrast to the autumn cessation of growth, which is supposed to be triggered by a critical day length, photosynthetic capacity in autumn is rather controlled by temperature.

P247. Disruption of water movement through stem galls resulting from infection by western gall rust on lodgepole pine.

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Western gall rust *Endocronartium harknessii* (J.P. Moore), Y. Hiratsuka (WGR) is an important forest pathogen of lodgepole pine [*Pinus contorta* Dougl. ex Loud.]. The fungus causes spherical galls on the main stem and lateral branches following the infection of expanding shoots. To determine whether or not stem galls disrupt water flow to tree crowns, seven WGR infected trees were harvested in each of two lodgepole pine stands approximately 10-years-old. Only galls encircling at least 75 percent of the stem circumference were sampled, as small stem galls were not thought to result in tree mortality. The flow rate, Q (m^3s^{-1}) of degassed water was measured through galled, and non-galled (above-and below-gall) stem sections using a hanging water column apparatus. Nonbonding 10% (w/v) acid fuchsin dye was then run through the stem sections to enable calculation of the conducting surface area, and determination of the flow path of water. The hydraulic conductivity (K_ψ, $m^2Pa^{-1}s^{-1}$) was calculated according to Darcy's Law for all stem sections. K_ψ values of the above-and below-gall sections were not significantly different from one another, and were averaged to calculate the percent reduction in K_ψ at the gall location for each sample tree. The flow of water through the galled sections occurred on the non-galled side of the stem; galls resulted in a 41 to 99 percent reduction in K_ψ.

P248. Differential induction of glucanase and peroxidase by antagonistic bacteria towards biocontrol of Leptosphaeria maculans in canola.

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Pseudomonas chlororaphis 190 (strain Pc190), *Bacillus cereus* E4 (strain BcE4) and bacterium DFE16 suppressed blackleg of

canola (*Brassica napus* L.), caused by *Leptosphaeria maculans* (Desm.) Ces & de Not, at both cotyledon and 3-4 leaf stage in the greenhouse. These bacteria, known for their antibiotic production, were also tested for their ability to induce plant defense enzymes at the cotyledon stage. The bacteria exhibited differential induction of glucanase and peroxidase. Only strain Pc190 exhibited positive induction of glucanase. When co-inoculated with the pathogen, strain Pc190 induced 1.5, 7 and 5 fold increase in glucanase activity at 72, 96 and 120 h respectively, compared to the pathogen control. At all points of time peroxidase induction by the bacteria, when inoculated alone or with the pathogen, was higher than the pathogen and water control. Peroxidase activity induced by strain Pc190, co-inoculated with the pathogen, gradually increased over time and peaked at 120 h, compared to strain Pc190 alone, which peaked at 72 h and decreased thereafter. Peroxidase activity induced by strain BcE4 alone and strain DFE16 alone was higher than when co-inoculated with the pathogen, except at 96 h when the co-inoculated treatments had higher activity. For the biocontrol of blackleg, these bacteria appear to exhibit a combination of mechanisms such as antibiotic production and induction of defense enzymes.

P249. Developing a protocol for large-scale inoculation of lentil germplasms with *Stemphylium botryosum*

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Stemphylium blight, a fungal disease caused by *Stemphylium botryosum* has been reported increasingly in lentil in Saskatchewan, but is not considered a major limiting factor to the production of lentil at this point. Severe losses of up to 94% have been reported in India and Bangladesh. The disease may develop into a more serious problem with the use of ascochyta blight and anthracnose resistant lentil varieties that maintain a more healthy canopy throughout the growing season thus creating a more conducive environment for pathogens like *S. botryosum*. A research program was initiated to study the biology of the pathogen on lentil. As spore production of *S. botryosum* was problematic, experiments were conducted to optimize sporulation conditions. Results showed that *S. botryosum* did not sporulate well under laboratory conditions compared to other lentil pathogens. As a consequence, a new protocol was developed to use mycelium as inoculum in replacement for conidia of *S. botryosum* to allow large-scale screening of lentil germplams for resistance to the pathogen. Isolate SB 19-203 was grown on modified Richard's medium, harvested, air dried and re-diluted in water using a standardized protocol. A calibration curve was developed by measuring the optical density and numbers of colony forming units of a dilution series of the mycelial suspension. The susceptible cultivar CDC Milestone was planted in 4 replications, and inoculated 2 weeks after seeding with each of the 6 dilutions of the mycelial suspension. Plants of CDC Milestone were also inoculated with suspensions of conidia at concentrations of 1 x 10^2, 1 x 10^3, 1 x 10^4, 1 x 10^5 and 2 x 10^5. A correlation analysis was conducted between both approach and the optimal concentration of the mycelial suspension was identified for resistance screening. Inoculation of lentil using mycelial suspensions provides a reliable screening system for testing lentil cultivars and breeding lines for resistance to stemphylium blight.

P250. The silencing suppressor protein P19 from Tomato Bushy Stunt Virus induces defence responses in tobacco.

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Plants attract a variety of intruders, from viruses, bacteria and fungi to insects. To protect themselves, plants have an armoury of defence mechanisms, including the PR protein accumulation and the release of antimicrobial secondary metabolites, induced by the recognition of elicitors. The virulence protein P19 from Tomato Bushy Stunt Virus is known to suppress the posttranscriptional gene silencing. Here we show the first conclusive evidence that a suppressor of silencing P19 acts as an elicitor of defence mechanisms in tobacco plants, including hypersensitive

response, antimicrobial secondary metabolites. This finding may provide a powerful tool for discovering the common ways between silencing and disease resistance mechanisms induced by both general and specific elicitors.

P251. Characterization of Differentially expressed genes of the *Rhizoctonia solani* AG3 during interaction with potato.

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Rhizoctonia solani Kühn (teleomorph: *Thanatephorus cucumeris* Frank Donk) is a soil-borne pathogen responsible for many diseases of several economically important crops world-wide. This complex species is divided into 13 anastomosis groups (AGs) based on hyphal fusion. Isolates belonging to AG-3 infect only potatoes and are responsible for black scurf disease. Irrespective of the AG, the infection process of *R. solani* is well established (attachment, infection structure formation, penetration), however no information is available on why a particular AG group such as AG-3 infects and causes disease on a specific crop species and not on another. The recognition process between the pathogen and its host at the onset of the infection is crucial for host-plant specialization. The understanding of this process and the characterization of implicated genes would enhance our knowledge on how to fight the disease. In this project, we will identify potato and fungal genes that are specifically implicated in the host plant recognition during the interaction of R. solani AG-3 with its host, and study their expression at the onset of the host-pathogen interaction. To carry out our objectives, suppression subtractive hybridisation (SSH) methodology will be performed to construct a first cDNA clone bank of differentially expressed (up-regulated) genes between a pathogenic (AG3 on potato as the host) and a none pathogenic (AG-3 on soybean as the non-host) interactions. To identify differentially down-regulated genes of AG-3, a second SSH will be performed between constitutive genes of AG-3 (expressed when AG-3 is grown without its host on synthetic medium containing no carbon) and a pathogenic (AG3 on potato) interaction. The differentially expressed genes will be screened using microarray technology. A subset of genes will be sequenced and their temporal expression will be estimated by quantitative RT-PCR in order to gain an in-depth understanding of specific interaction genes.

P252. Change of SODs isoforms by *Mycosphaerella fragariae* infection in two resistant and susceptible cultivars in strawberry.

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Relationship between induction of different superoxide dismutase (SOD, EC 1.15.1.1) isoforms and susceptibility of strawberry cultivars was examined during a time course infection of leaves by *Mycosphaerella fragariae* in two cultivars, i.e. “Joliette” (resistant) and “Kent” (susceptible). Infection of the strawberry leaves with the pathogen resulted in increase in specific SOD activities in both cultivars, which higher levels were observed in Joliette. Several SOD activity bands were detected in both Joliette and Kent infected leaf extracts, including a manganese SOD with 19-kD molecular weight and several isoforms of iron SOD. During 20 days of infection, SOD isoform profile of both cultivars differently changed. Induction of copper-zinc SOD with a low molecular weight (16-kD) in Joliette, which was not observed in Kent was notable, suggesting a major role for this molecule in the mechanism of defense against the pathogen attack and regulation of oxidative damage in strawberry.

P253. Bacterial natural product as a selective herbicide

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A new natural herbicide for selective control of weeds has been discovered. The active ingredients in this natural herbicide were partially identified. Under greenhouse conditions, a dry weight reduction by 61.06%, 61.8%, 68.95%, 72%, 76.48% and 92.09% were achieved on white clover (*Trifolium repens*), black medic (*Medicago lupulina*), chickweed (*Stelloria media*), Bermuda grass (*Cynodon dactylon*), dandelion (*Taraxacum officinale*), and crabgrass (*Digitaria* spp.) respectively. Under field conditions, a 68% reduction in dry weight of dandelion seedlings

was achieved with this bacterial natural product, while a 70% reduction in dry weight was achieved with a commercial herbicide Killex. No damage to Kentucky blue grass (*Poa pratensis*), Perennial ryegrass (*Lolium perenne*), Tall fescue (*Festuca arundinasea*), and creeping red fescue (*Festuca rubra* var Boreal) tested in the greenhouse was observed indicates that this bacterial natural product could be a potential herbicide for weed control in turf grass.

P254. Bacterial biofilms: impact on plant and post-harvest disease control and on the need for novel sanitation products and protocols

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Bacteria are usually described as free floating single cells (planktonic); however, most grow as complex multicellular-like communities attached to surfaces and immersed in polysaccharides (biofilms, or "slime"). Biofilms on surfaces of equipment, tools, transport vehicles, containers and storage facilities, as well as in vascular, seed and post-harvest infections, are very costly to agricultural industries. Current pesticides and protocols, developed based on killing of single-celled organisms, are often ineffective against microbial biofilms, which are much more resistant to disinfectants and antibiotics than planktonics. We evaluated the efficacy of currently used disinfectants and antimicrobials against soft-rot Erwinia, Pseudomonads, Xanthomonads and other plant pathogenic bacteria biofilms using the MBEC™ and BEST™ Assays, which allow for testing of antimicrobial susceptibility directly against biofilms, on various surfaces. Erwinia bacteria in the biofilm state were over 250 times more resistant to quaternary ammonium than the same bacteria in suspension, and Pseudomonads were much more resistant to silver and copper based compounds. These biofilms also showed increased resistance to hydrogen peroxide and some grew better on organic surfaces (such as wood) on opposed to innert ones (ex., polystyrene). Similar trends were observed for other roganisms and antimicrobials, suggesting that novel pesticides and disinfectants, as well as new and/or revised decontamination procedures, are necessary for the effective treatment and prevention of plant diseases and post-harvest decay, and for proper surface sanitation. In addition, we report the identification of two compounds with superior efficacy against biofilms than currently available ones..

P255. A novel generation of flower preservatives, potential pesticides and surface disinfectants for agricultural applications – anti-biofilm compounds

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We identified new compounds for treatment and prevention of plant microbial infections associated with biofilms. Biofilms ("slime") are complex communities of microorganisms, attached to surfaces, which are more resistant to antimicrobials than free single-cell form (planktonics), Biofilms impact plant health through vascular infections, shortening the shelf life of cut flowers and produce, and recurrent contamination of surfaces leading to spread of diseases. In cut flowers, biofilms clog the xylem inducing water stress, leading to wilting, ethylene production and premature death. Proper decontamination of surfaces is important for minimizing contamination during post-harvest handling, transportation and storage. Many currently available antimicrobials used for these purposes are not fully effective against biofilms, as they were designed and tested against planktonic growing organisms, more susceptible to their action. We present a more efficacious and practical way to maximize quality of plant materials and floricultural products. We characterized the efficacy of continuous release silver-based antimicrobials, successfully applied in the medical field, against biofilm infections in plants and horti-floricultural products. The efficacy of inorganic and organic silver salts as antimicrobials is known, but they do not afford prolonged protection due to removal or complexation of the free silver ions. Our patented compound, nanosilver, is superior to silver soluble salts, as it allows sustained release of silver ions at antimicrobial active levels over a period of days, and enhanced antimicrobial efficacy than compounds such as silver nitrate. The vase life of roses was extended by 30-100% by nanosilver depending on testing conditions and level of microbial challenge. We demonstrate that stable, slow release nanocrystaline silver compounds, can be used as antimicrobials

against bacteria and fungi pathogens, including biofilms, growing on plant surfaces.

P256. A growth chamber method for screening canola for resistance to fusarium wilt

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Fusarium oxysporum f.sp. *conglutinans*, causes fusarium wilt in canola and is usually identified in later stages of plant development which may make chemical treatment difficult. Using cultivars with resistance is the only major form of control currently available. Therefore, quick and simple methods of screening lines could benefit breeding for resistant cultivars. Two temperatures (22^{o}C, 24^{o}C), two soil types (soil-less mix (Sunagro), 1:1 Sunagro : Vermiculite), four cultivars; 45A55, Bianca II, DS-Roughrider (susceptible) and InVigor 5020 (resistant) were used to develop a simple protocol for screening resistant canola lines/cultivars using a split-split-plot design. Inoculum grown on wheat grain was applied to soil at seeding and plants were rated weekly for 28 days after seeding. Plants at 24^{o}C had a significantly higher wilt severity than those at 22^{o}C, suggesting the higher temperature of 24^{o}C would provide increased disease severity for screening. Soil type had no significant effect on wilt severity. 45A55 developed the highest wilt severity with a final rating of 83%. Bianca II and DS-Roughrider showed no significant differences in wilt severities of 77% and 78%, respectively, while InVigor 5020 was significantly more resistant with a severity of 1% at the final rating. This protocol repeatedly distinguished resistant and susceptible cultivars at each temperature and soil type and showed that it could be used for screening canola lines/cultivars for resistance to *F. oxysporum* f.sp. *conglutinans*. Although no differences were observed in wilt severity between soil types, working with the Sunagro soil alone is more manageable than the more porous 1:1 Sunagro-Vermiculite mix which requires frequent watering.

P257. Susceptibility of lentil varieties to rhizoctonia seedling blight and root rot in Alberta

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Fourteen lentil (*Lens culinaris* Medik.) varieties were evaluated for susceptibility to rhizoctonia seedling blight and root rot (*Rhizoctonia solani* Kühn) in field experiments conducted at Brooks, AB in 2003 and 2004. Seedling survival was compared among the varieties in artificially infested and non-infested plots. Significantly higher survival rates were observed in 2004 compared to the previous year. Overall survival in infected plots was 12% in 2003 and 46% in 2004 compared to the non-infected plots. 'Indianhead' was the most susceptible cultivar, followed by 'Richlea'. 'Milestone' and 'Plato' were relatively tolerant. However, 'Lemay', the most tolerant of the cultivars in 2003, was relatively susceptible in 2004. Cluster analysis showed two major groups of lentil varieties (relatively susceptible and tolerant varieties) with seven varieties in each group.

P258. Yeasts of lowbush blueberry: potential biocontrol agents for blueberry diseases

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Foliar and blossom blights, mummy berry and fruit rot caused by *Monilinia vaccinii-corymbosi* and *Botrytis cinerea* are major concerns for growers of lowbush blueberry (*Vaccinium angustifolium* Ait. and *V. myrtilloides* Michx.) in the Atlantic provinces. Yield and fruit quality are compromised by these diseases. The epiphytic yeasts and yeast-like fungi were explored as sources of biological control agents for these pathogens. Yeasts were washed from leaves and fruit collected in Nova Scotia and were isolated on semi-selective yeast-malt-glucose agar (YMGA) medium containing antibacterial compounds. Colony forming units were counted after incubation for 14 days at 12 °C.

Representative isolates of pink, white and black yeasts were identified using colony and cell morphology and information on D1 and D2 regions of the large rDNA subunit after whole-cell polymerase chain reactions (PCR) and DNA sequencing. Identifications were based on comparisons with sequences in GenBank using the Basic Local Alignment Search Tool (BLAST). The ubiquitous black, yeast-like ascomycetous fungus, *Aureobasidium pullulans*, was identified based on morphology alone . The yeasts were all basidiomycetous species. White yeasts were morphologically similar to *Cryptococcus albidus* and were found in all plant samples. White yeasts included *C. terreus*, *C. terricolus*, *C. wieringae* and a new species similar to *C. saitoi*. In addition, a pink yeast, *Rhodotorula nothofagi* was found on mummified (*Monilinia*) blueberry fruit only . All of these epiphytes were capable of inhibiting the growth of the pathogen *Botrytis* in dual culture bioassays. Biological control potential was verified on potted, artificially infested blueberry plants in controlled environment chambers

P259. Differentiation of *Colletotrichum* spp. from scentless chamomile and lentil based on infection characteristics and ITS sequences

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Previous studies showed that isolates of *Colletotrichum truncatum* (Schwein) Andrus & W.D. Moore from scentless chamomile (SC), a noxious weed in western Canada, caused disease on the weed but not on commercial cultivars of lentil, where *C. truncatum* was reported as a major pathogen. Taxonomy based on fungal morphology placed SC and lentil isolates into the same species despite the apparent differences in pathogenicity. The current study compared fungal isolates based on rDNA sequencing of two moderately conserved internal transcripted spacer (ITS) regions: ITS1 and ITS2. The ITS regions from a total of 50 *Colletotrichum* isolates were sequenced and the results showed that isolates from SC were different from the others, including the isolates from lentil. SC isolates were most closely related to *C. truncatum* isolates from lentil, but the divergence was so significant that they formed their own phylogenetic group. Inoculation of lentil plants with isolates from SC and lentil showed little difference in spore germination and appressorium formation, but the isolates from SC developed fewer and more limited infection hyphae in epidermal cells when compared with the isolate from lentil. The data suggest that the *Colletotrichum* isolates from SC are distinct from those of lentil.

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