
# Program:

# CSPP Western Regional Meeting

**February 22 - 24, 2007**

**Manteo Resort**
**Kelowna, British Columbia**

**Sponsored by:**

**UBC Okanagan**

**SemBioSys Genetics Inc.**

**Varian Canada, Inc.**

**Ariadne Genomics**

**BC Herb growers Association**

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# Agenda

**Abbreviations:**

| | |
|---|---|
| MR | Manteo Resort |
| MHW | Mission Hill Winery |
| UBC O | UBC Okanagan |
| NPC | UBC O Natural Products Laboratory (room Sci339) |

**Important Notices:**

i) The wine / UBC O tours must be registered for, only using the online system until February 19th. Unregistered individuals wishing to participate will be responsible for own transportation, registration, lunch etc.
ii) Please see detailed schedules for exact location of events.
iii) This program is subject to minor changes. Please check for updated version before the conference.

## *Events schedule:*

Feb 22, 2007

| TIME | FUNCTION | PLACE |
|---|---|---|
| 17:00 – 20:00 | Registration | MR Foyer |
| 19:00 – 21:00 | Mixer | MR Upper Lounge |

Feb 23, 2007

| TIME | FUNCTION | PLACE |
|---|---|---|
| 07:00 – 08:00 | Registration | MR Foyer |
| 07:45 – 08:00 | Opening remarks | MR |
| 08:00 – 10:00 | Session I and II | MR |
| 10:30 – 12:15 | Session III and IV | MR |
| 12:15 – 13:15 | Lunch | MR |
| 13:15 – 15:15 | Sessions V and VI | MR |
| 15:15 – 15:45 | Coffee and Poster setup | MR |
| 15:45 – 17:45 | Poster presentations | MR |
| 18:30 – 21:00 | Dinner | MR Upper Lounge |

Feb 24, 2007

| TIME | FUNCTION | PLACE |
|---|---|---|
| 10:30 – 12:00 | Wine Tour | MHW |
| 12:00 – 13:00 | Lunch / Pizza | UBC O NPC |
| 13:00 – 14:00 | UBC O Tour | UBC O |

## *Speaker schedule*

| Concurrent Session I: Metabolic Pathways<br>Location: Waterfront Room III<br>Chair: Joerg Bohlmann | | |
|---|---|---|
| Time | Speaker | Presentation Title |
| 08:00–<br>08:30 | Lange, BM | Regulation of peppermint essential oil biosynthesis - development of a mathematical model and experimental validation |
| 08:30–<br>09:00 | Page, J | Trichome genomics: Dissecting terpenophenolic biosynthesis in *Humulus lupulus* (hops) |
| 09:00–<br>09:30 | Murch, S | Metabolomics / Metabonomics and chemodiversity in endangered plants |
| 09:30–<br>09:45 | Mahmoud, SS | Genomics of aroma formation in lavenders |
| 09:45–<br>10:00 | Covello, PS | Functional genomics and biosynthesis of artemesinin |
| **10:00 – 10:30 Coffee Break** | | |

| Concurrent Session II: Rhizosphere and Pathogen Interactions<br>Location: Theatre<br>Chair: Elizabeth Schultz | | |
|---|---|---|
| 08:00–<br>08:30 | Punja, ZK | Biochemical changes and gene expression induced in ginseng roots infected by *Fusarium equiseti* |
| 08:30–<br>08:45 | Letts, MG | Aspect-related differences in seasonal acclimation to environmental stress in *Pinus flexilis* (Limber Pine) |
| 08:45–<br>09:00 | Jones, M | Physiological diversity of ectomycorrhizas on Douglas-fir seedlings regenerating after wildfire or clearcutting |
| 09:00–<br>09:15 | Hawkins, BJ | A comparison of ammonium, nitrate and proton net fluxes along seedling roots of Douglas-fir and lodge pole pine |
| 09:15–<br>09:45 | Peterson, CA | The role of root epidermal suberin in partial resistance of soybean to *Phytophthora* root rot |
| 09:45–<br>10:00 | Kathiria, P. | Increased trans-generational tolerance in pathogen infected plants |
| 10:00 – 10:30 Coffee Break | | |

| Concurrent Session III: Metabolism and Stress Pathways<br>Location: Waterfront Room III<br>Chair: Jonathan Page | | |
|---|---|---|
| Time | Speaker | Presentation Title |
| 10:30–<br>11:00 | Bohlmann, J | Secondary metabolites in the chemical defense of conifers |
| 11:00–<br>11:30 | Lund, ST | iTRAQ analysis of the grape proteome at ripening initiation reveals a previously unidentified protein controlling a downstream step in anthocyanin biosynthesis |
| 11:30–<br>12:00 | Clouston, J | Plant Pathway Analysis for Non-Model Organisms |
| 12:00–<br>12:15 | Samuels, L | ABC transporters at the plant surface export protective lipids of the cuticle |

| Concurrent Session IV: Development and Physiology<br>Location: Theatre<br>Chair: Igor Kovalchuk | | |
|---|---|---|
| 10:30–<br>11:00 | Muench, DG | Functional characterization of the core exon junction complex (EJC) proteins in plants |
| 11:00–<br>11:15 | Hongwei, H | Vein patterning in Arabidopsis |
| 11:15–<br>11:30 | Weger, HG | Differential biological availability of iron to green algae |
| 11:30–<br>11:45 | Gray, GR | Growth and photoinhibitory responses of *Arabidopsis* and contrasting ecotypes of *Thellungiella salsuginea* |
| 11:45–<br>12:00 | Schuetz, M | Role of *MP* mediated auxin signaling and *PIN1* mediated auxin transport in leaf vascular pattern formation |
| 12:00–<br>12:15 | Chen, JG | TRICHOMELESS1 Controls Trichome Patterning by Suppressing the Expression of *GLABROUS1* in *Arabidopsis* |
| 12:15 –13:15 Lunch<br>Waterfront Room III | | |

| Concurrent Session V: Agriculture and Biotechnology<br>Location: Waterfront Room III<br>Chair: Astrid Boeckelmann | | |
|---|---|---|
| 13:15–<br>13:45 | De Luca, V | Agricultural biotechnology of crops for producing commercially valuable natural products |
| 13:45–<br>14:15 | Moloney, MM | Production of human insulin and other biopharmaceuticals in oilseed crops: achievements and challenges. |
| 14:15–<br>14:30 | Natarajan, S | Biosafety of transgenic soybean using proteomic technology |
| 14:30–<br>14:45 | Wally, O | Evaluation of promoter activity and over-expression of a rice peroxidase gene in transgenic carrot (*Daucus carota* L.) |
| 14:45–<br>15:15 | Facchini, P | Opium poppy: blueprint for an alkaloid factory |
| **15:15 – 15:45 Coffee Break and Poster Setup** | | |
| **15:45 – 17:45 Poster Presentations: Theatre** | | |

| Concurrent Session VI: Gene Stability and Expression<br>Location: Theatre<br>Chair: Melanie Jones | | |
|---|---|---|
| 13:15–<br>13:30 | Boyko, A | Somatic and transgenerational changes in genome stability of salt-exposed plants |
| 13:30–<br>13:45 | Malik MR | Gene expression profiling of microspore embryogenesis in *Brassica napus* |
| 13:45–<br>14:00 | Gagne, S | Elucidation of acetylenase functionality |
| 14:00–<br>14:15 | McKinnon, DJ | Dual targeting of $NADP^+$-isocitrate dehydrogenase to mitochondria and chloroplasts |
| 14:15–<br>14:45 | Kovalchuk, I | Genetic and epigenetic regulation of genome stability in plant response to stress |
| 14:45–<br>15:00 | Ellis, BE | Non-canonical stromal targeting of an Arabidopsis mitogen-activated protein kinase kinase |
| 15:00–<br>15:15 | Cole, I | *Scutellaria racemosa*: An important species for study of medicinal compounds |
| **15:15 – 15:45 Coffee Break and Poster Setup** | | |
| **15:45 – 17:45 Poster Presentations: Theatre** | | |
| **18:30 – 21:00 Dinner<br>MR, Upper Lounge** | | |

# Presentation abstracts

## *Concurrent Session I: Metabolic Pathways*

### (001) Regulation of peppermint essential oil biosynthesis - development of a mathematical model and experimental validation

Lange, B.M. Institute of Biological Chemistry and Center for Integrated Biotechnology, Washington State University, Pullman, WA 99163

The essential oil of peppermint (Mentha x piperita) is synthesized and stored in specialized glandular trichomes (oil glands) on aereal surfaces. Methods for the bulk isolation (using a mechanical abrasion technique) and individual cell sampling (using microcapillaries) have been developed and allow direct access to genes, enzymes and metabolites of these specialized cell factories. Based upon the kinetic properties of the biosynthetic enzymes and measurements of relevant transcripts, enzyme activities and metabolites, a mathematical model that describes the regulation of monoterpene biosynthesis in oil glands has been generated. We present evidence that the essential oil composition of peppermint oil glands does not depend solely on enzyme kinetics and gene/protein expression patterns but that transport of metabolic intermediates also plays an important role. We have tested the mathematical model by predicting how changes in the expression of certain biosynthetic enzymes (brought about by transgenic overexpression or silencing of the corresponding transcripts or by environmental factors) impact essential oil composition, compared these predictions with experimental data, and further optimized the model. Using such an iterative approach we are now able to use mathematical modeling to guide transgenic approaches aimed at modulating essential oil composition so that the levels of undesirable components are reduced (e.g., pulegone and menthofuran), whereas the levels of desirable components (e.g., menthone and menthol) are increased.

### (002) Dissecting terpenophenolic biosynthesis in *Humulus lupulus* (hops)

Page, J. and Nagel, J. NRC Plant Biotechnology Institute, 110 Gymnasium Place, Saskatoon, SK, S7N 0W9

Prenylated acylphloroglucinols and prenylflavonoids found in the hop plant, *Humulus lupulus* L., contribute bitter flavour to beer and possess chemopreventative properties, respectively. Most terpenophenolics found in hop are synthesized and stored in glandular trichomes that develop on female flowers. We analyzed Expressed Sequence Tags (ESTs) derived from trichome-specific cDNA libraries as a means to understand the biosynthetic

systems at work in these specialized secretory structures. EST sequencing of the hop lupulin gland transcriptome has clarified many of the metabolic pathways by which hop terpenophenolics are synthesized and has led to the discovery of genes potentially involved in their biosynthesis. Functional characterization of trichome-specific *O*-methyltransferases from hop has revealed a novel enzyme, desmethylxanthohumol *O*-methyltransferase, which catalyzes a key step in xanthohumol formation, and another *O*-methyltransferase that methylates phenylpropanoid substrates. Understanding how terpenophenolics are synthesized may allow the metabolic engineering of this important, and potentially valuable, branch of secondary metabolism in plants.

**(003) Metabonomics & chemodiversity in endangered plants**

Murch, S. J. Chemistry, UBC Okanagan, Kelowna, BC, Canada

Metabolomics is the study of the whole complement of small compounds in a biological sample. In an average leaf, it is common to detect 4,000 – 14,000 unique phytochemicals. About 1,150 of these can be identified as common primary metabolites while the majority are products of plant secondary metabolism, many with no known chemical identity. Further, the phytochemical profile of a plant tissue fluctuates with time, environment, ecotype, individuality, insect and microbial interactions and many other factors. Endangered species can tell us a great deal about those aspects of the phytochemistry that are important for adaptation and survival of a species. The metabolomic analyses of Cycads, Wollemi Pine (*Wollemia nobilis*) and several medicinal plants are beginning to provide new insights into plant secondary metabolism, the conservation of important metabolites across species and the potential for discovery of important new plant chemistry.

**(004) Genomics of essential oil and aroma formation in lavenders**

Mahmoud, S.S., UBC Okanagan, Kelowna, BC, Canada.

Lavenders (*Lamiacea*, the mint family) are grown worldwide both as ornamental plants, and for the production of lavender essential oil, which are extensively used as additives to numerous cosmetic, pharmaceutical and food products. We are using the English Lavender (*Lavandula angustifolia*) as a model system for the study of regulation of essential oil production, and floral aroma development in plants. The essential oils and floral aromas contain a complex mixture of mono- and sesquiterpenes produced in glandular trichomes present on the surfaces of the leaves and flower parts of the plant. Currently, the genetic and biochemical factors that control the production of these metabolites are poorly understood. We are using a genomics approach to identify and study genetic elements that regulate this process in lavender leaves and flowers. We have recently obtained sequence information for approximately 10,000 genes from English

lavender, and are using the gene chip (or microarray) technology to identify those involved in the biosynthesis of terpenoids in this plant. Once identified, the sequences could be used in precision breeding experiments in order to develop agronomically improved lavender varieties.

**(005) Functional genomics and the biosynthesis of artemisinin**

Covello, P.S., Teoh, K.H., Polichuk, D.R. Reed, D.W. and Nowak, G.
Plant Biotechnology Institute, 110 Gymnasium Place, Saskatoon, SK, Canada S7N OW9

Artemisinin, a sesquiterpene lactone endoperoxide derived from *Artemisia annua*, forms the basis of the most important treatments of malaria in use today. In an effort to elucidate the biosynthesis of artemisinin, an expressed sequence tag approach to identifying the relevant biosynthetic genes was undertaken using isolated glandular trichomes as a source of mRNA. Progress in the elucidation of genes involved in artemisinin biosynthesis will be discussed, including the isolation of a cDNA encoding a cytochrome P450 involved in amorpha-4,11-diene oxidation.

## *Concurrent Session II: Rhizosphere and Pathogen Interactions*

**(006) Biochemical changes and gene expression induced in ginseng roots infected by *Fusarium equiseti.***

Punja, Z.K., Goswami, R.S. and Rahman, M. Department of Biological Sciences, Simon Fraser University, 8888 University Drive, Burnaby, British Columbia V5A 1S6, Canada

Ginseng roots inoculated with *Fusarium equiseti* developed superficial reddish-brown lesions within 7 days. Affected cells stained a bluish-green with Toluidine blue O, indicating an accumulation of phenolic compounds, and hyphae were observed in the uppermost 10-12 cell layers. HPLC analysis revealed higher levels of cinnamic acid, quercetin, catechin and chlorogenic acid in diseased tissues compared to healthy tissues. A zone of cells, resembling phellogen or cork cambium, developed in a zone below the diseased area. These cells were shown to contain lignin. Suppressive subtractive hybridization of mRNA from healthy and diseased tissues was used to identify differentially expressed EST's. Sequence analysis identified a large number of genes involved in host defense responses. These included members of signaling pathways, a range of peroxidases, catalases and PR proteins, as well as proteins related to cell death and phenolic production. The gene expression pattern suggested an involvement of reactive oxygen species and the jasmonic acid pathway. The upregulation of genes encoding for products potentially involved in lignification, detoxification, apoptosis, anthocyanin production and cell wall reinforcement highlight the likelihood of disease symptoms being a manifestation of a defense response in the roots.

**(007) Aspect-related differences in seasonal acclimation to environmental stress in *Pinus flexilis* (Limber Pine).**

Letts, M.G. and Johnson, DRE Department of Geography, University of Lethbridge, Lethbridge AB Canada, T1K 3M4

Microclimatological and plant physiological data are presented for *Pinus flexilis*, growing in shallow soils on windswept slopes of four distinct aspects at Lakeview Ridge, Waterton Lakes National Park (1938 m). Higher solar radiation receipt leads to higher near-surface temperature, which results in lower soil moisture and higher vapour pressure deficit on SE and SW-facing slopes than on NW-facing or NE-facing slopes. This results in a temporary shutdown of net photosynthetic uptake during a late-summer drought on southerly aspects. Seasonal patterns of plant acclimation to moisture stress are assessed for each aspect, using leaf gas exchange, leaf reflectance and stable carbon isotope techniques. The bimodal seasonal pattern of net photosynthetic uptake suggests that shoulder seasons may be important for net carbon uptake in this environment, especially on south-facing aspects.

**(008) Physiological diversity of ectomycorrhizas on Douglas-fir seedlings regenerating after wildfire or clearcutting**

Jones, M., Biology and Physical Geography, UBC Okanagan, Kelowna, BC

Douglas-fir seedlings often show poor regeneration in dry, interior sites after logging. We initiated a study comparing roots and rhizosphere biology of Douglas-fir seedlings regenerating after wildfire and clearcut logging. Specifically we examined whether the activities of three enzymes used in the breakdown of soil organic matter varied (i) amongst disturbance types and (ii) amongst ectomycorrhizal fungi within disturbance types. Four disturbance types (high severity burn, low severity burn, clearcut logging with forest floor removal, clearcut logging without forest floor removal) were compared with unburned Douglas-fir forest. The activities of phosphatase, chitinase, and ß-glucosidase were measured on 5-7 ectomycorrhizas formed by the same fungus on each of four replicate sites of each disturbance type. When the activities of the ectomycorrhizas were considered together as a community, there was no evidence of physiological specialization for a specific type of disturbance, even though soil chemistries differed considerably. Individual fungal species were, however, associated with different profiles of enzyme activity within the same site or same disturbance treatment. These results suggest that ectomycorrhizas of different fungi may be adapted to different soil microsites or that colonization following disturbance is driven more by the ability to disperse and colonize roots rather than specific physiological attributes.

**(009) A comparison of ammonium, nitrate and proton net fluxes along seedling roots of Douglas-fir and lodgepole pine**

Hawkins, B.J. Centre for Forest Biology, University of Victoria, Victoria, BC, Canada.

$NH_4^+$, $NO_3^-$ and $H^+$ net fluxes were measured with ion-selective microelectrodes along the root length of Douglas-fir (*Pseudotsuga menziesii*) and lodgepole pine (*Pinus contorta*) seedlings. To investigate the influence of N pretreatment on $NH_4^+$ and $NO_3^-$ net fluxes, seedlings were grown with $NH_4^+$, $NO_3^-$, $NH_4NO_3$ or no N, and net fluxes were measured in solutions containing one or both N ions, or no N. Net $NO_3^-$ and $NH_4^+$ influx and net $H^+$ efflux were greater in Douglas-fir than lodgepole pine. Highest net $NO_3^-$ influx occurred 10-30 mm from root apices and highest net $NH_4^+$ influx occurred in the first 5-20 mm, depending on species. Mean $NO_3^-$ and $NH_4^+$ net fluxes did not differ when measured in $NH_4NO_3$ solution or solutions containing the respective N ion. We observed no preference for $NH_4^+$ as a N source, and no inhibition of $NO_3^-$ uptake by $NH_4^+$ in the measurement solution.

**(010) The role of root epidermal suberin in partial resistance of soybean to Phytophthora root rot**

Peterson, C A.[1], Ranathunge, K. [1], Fang, X.[1], Thomas, R.[2], Bernards, M A.[2] and Gijzen, M.[3] [1]Department of Biology, University of Waterloo. [2] Department of Biology, University of Western Ontario. [3]Agriculture and Agri-Food Canada, London Station.

Roots of soybean [*Glycine max* (L.) Merr.] possess suberin in their epidermis, as demonstrated by histochemical tests and chemical analysis. The degree of partial resistance of various soybean cultivars, and also recombinant inbred lines derived from them, to infection by *Phytophthora sojae* correlates with the quantity of root aliphatic suberin, suggesting that it may act as a mechanical impedance to pathogen ingress. This idea was supported by a comparison of early stages of infection in a cultivar with strong partial resistance *vs* a cultivar with weak partial resistance. Zoospores were attracted to the roots of both cultivars equally well, and the encystment and germination of the spores was similarly uniform. However, growth of the germ tubes through the middle lamellae of the epidermis was slower by two to three hours in the more resistant cultivar. Such a delay would allow time for the root to mount chemical defenses that may prevent the hyphae from entering the vascular cylinder.

**(011) Increased trans-generational tolerance in pathogen infected plants**

Kathiria, P., Boyko, A. Zemp, F. and Kovalchuk, I. Department of Biological sciences, University of Lethbridge, Lethbridge, Alberta
Canada.

Plants often receive biological stress in form of pathogens. In this study the response of plants to compatible pathogens was analyzed. The model system of Tobacco mosaic virus (TMV) and *Nicotiana tabacum* plants was used for the experiments. Initial studies revealed production of a mobile signal which travels faster than virus. This signal leads to changes in genome stability. Further analysis was carried out on progeny of plants which received the signal. It was observed that there is higher tolerance to pathogen in the progeny of virus infected plant as compared to mock treated plants. This was consistent with delay in the TMV mediated symptom appearance. The persistence of tolerance for one further generation was also analyzed. The results indicate that the tolerance can last for more then one generation. This new finding may help in our attempts for selective breeding for resistant plants.

## *Concurrent Session III: Metabolism and Stress Pathways*

### (012) Chemical diversity in plant defense against insects: terpenoid synthases and cytochrome P450 in conifers

Bohlmann J. Michael Smith Laboratories, University of British Columbia, Vancouver, V6T 1Z4, British Columbia, Canada

Conifer trees display a large array of defenses against insect pests and insect associated pathogens. Insect induced defenses in species of spruce (Picea spp.) include a myriad of combinations of constitutive and induced, chemical and physical, direct and indirect, as well as local and systemic defenses. Among some of the most prominent insect-induced defenses in conifers are terpenoid (traumatic resinosis; terpenid volatile emissions) and phenolic secondary metabolites. Traumatic resinosis involves methyl jasmonate or ethylene-inducible de novo differentiation of specialized anatomical structures (traumatic resin ducts, TRD) for induced terpenoid accumulation in the developing xylem. Phenolic defenses involve the induction of polyphenolic parenchyma (PP) cells of largely unknown chemical contents. Insect induced volatile emissions are based on passive release of resin terpenoids and on the active de novo formation and emission of non-resin monoterpenes and sesquiterpenes. In targeted biochemical characterization of insect-induced secondary metabolite defenses, we have functionally characterized a large family of conifer terpenoid synthase genes as well as cytochrome P450 genes for conifer diterpene resin acid formation and measured their expression in response to real and simulated insect attack. Large-scale genomics resources for species of spruce (>200,000 ESTs; ca. 6,400 FL-cDNA; and a 22k-cDNA microarray) and optimized proteomics tools have been developed in our conifer genomics program (www.treenomix.ca) and applied for a comprehensive analysis of conifer defense response to insect attack. In parallel, we have established an EST resource for a bark beetle associated fungal pathogen representing a large number of CYP450 potentially involved in detoxification of conifer chemical defenses.

### (013) iTRAQ analysis of the grape proteome at ripening initiation reveals a previously unidentified protein controlling a downstream step in anthocyanin biosynthesis.

Lund, S.T.[1], Lücker, J.[1], Elliot, M.[2], and Smith, D.[2] [1]Wine Research Centre, University of British Columbia, Vancouver, BC, Canada. [2]UVic-Genome BC Proteomics Centre, Victoria, BC, Canada.

iTRAQ is a recent quantitative proteomics technique for comprehensively characterizing protein expression patterns in four biological samples simultaneously. We analysed four stages of both exocarp and mesocarp tissues of *Vitis vinifera* cv. Cabernet Sauvignon (CS) berries during ripening initiation. In 'red' cultivars such as CS, there is a striking

colour change in the exocarp from green to purple during ripening initiation, which is caused by the accumulation of anthocyanin flavonoids. Consistent with this phenotype, we detected strongly up-regulated peptides in our iTRAQ data annotated as flavonoid pathway enzymes only in the exocarp tissue. We also detected a methyltransferase showing a similar expression pattern to the up-regulated flavonoid pathway proteins. The most common anthocyanin in ripe CS is malvidin 3-O-glucoside, but the methyltransferase producing this compound had remained unidentified. Heterologous enzyme assays showed that this methyltransferase is responsible for converting delphinidin 3-O-glucoside to petunidin 3-O-glucoside and malvidin 3-O-glucoside.

**(014) Molecular networks in plants: extraction from literature and pathway analysis**

Clouston J. , Senior Director, Worldwide Sales, AriadneGenomics Inc. Rockville Maryland.

Using a high-content linguistics tool that was orignially designed to assist mammalian pathways analysis and research, Ariadne Genomics has compiled a reference database of 70,000 relationships between plant proteins and small molecules to aid in plant-based research. The resulting database includes facts about protein interactions, promoter binding, molecular biosynthesis and trafficking, and cell process regulation for the primary model organism Arabidopsis thaliana. Using sequence similarity and the topology of interactions, Ariadne has made protein relations predicitons to support research with barley, tomato, potato, rice, corn, and tobacco. The presentation will focus how the current ResNet Plant reference database was constructed and how it may be used and modified to support the seven current plants and/or other plant species.

**(015) ABC transporters at the plant surface export protective lipids of the cuticle.**

Samuels, L., Kunst, L., Jetter, R. and Bird, D. Department of Botany, University of British Columbia, Vancouver, BC, Canada, V6T 1Z4.

Plants are covered with a protective cuticle that forms a critical interface between the organism and its environment. The cuticle is composed of a crosslinked polymer of oxygenated-fatty acids called cutin, as well as waxes, a complex mixture of very long chain fatty acids and their derivatives. Cuticle synthesis requires extensive transport of these lipids out of the epidermal cells across the cell wall to the plant surface. Previously, we showed that an ABC transporter, *CER5,* was required for normal wax export from *Arabidopsis thaliana.* Based on epidermis-specific microarray data, a related ABC transporter, WBC11, was hypothesized to be another candidate for a cuticular lipid transporter. Arabidopsis *wbc11* T-DNA insertional knock-out mutants exhibited lipidic inclusions inside epidermal cells similar to the previously characterized wax transporter mutant *cer5*, but had a slightly stronger reduction in the alkane load in surface waxes. Moreover, the *wbc11* knock-out also showed defects not present in *cer5* including post-

genital organ fusions, stunted growth and a reduction in cutin load on the plant surface. The double knock-out *wbc11 cer5* exhibited the same morphological and biochemical phenotypes as the *wbc11* knock-out. These results demonstrate that WBC11 functions in secretion of surface waxes, probably by interacting with CER5 at the plasma membrane. However, unlike *CER5*, *WBC11* is further required for deposition of the cutin polymer matrix.

## *Concurrent Session IV: Development and Physiology*

### (016) Functional characterization of the core exon junction complex (EJC) proteins in plants.

Muench, D.G., Park, Nam-Il, and Yeung, E.C. Department of Biological Sciences, University of Calgary

The exon junction complex (EJC) plays an important role in post-transcriptional control of gene expression. Mago Nashi (Mago) and Y14 are core EJC proteins that operate as a functional unit in animal cells. The Mago-Y14 heterodimer interacts with other EJC core and peripheral proteins, including a protein called PYM. There is limited knowledge on the biochemical, cellular and functional characteristics of the EJC and its orthologs in plants. Mago, Y14 and PYM have been studied in detail in our laboratory. Protein-protein interactions, subcellular localization, and the identification of nuclear targeting signals have been characterized. In addition, we have initiated a functional analysis of these proteins, including the role of the EJC in non-sense mediated mRNA decay (NMD) in plants.

### (017) Vein patterning in *Arabidopsis*

Hongwei H., Garrett J., Cormack R., Meservy J., Tavernini L, Blackshaw M. and Schultz E.. Department of Biological Sciences, University of Lethbridge, Lethbridge, AB, T1K 3M4

Current models for vein pattern formation suggest that continuous vascular cell files depend upon auxin canalization through directed localization of auxin efflux (PIN) and possibly auxin influx (AUX1) proteins in procambial cells. PIN and AUX1 localization depend on separable vesicle trafficking pathways. Adoption of vascular cell fate also depends on auxin response pathways. We have isolated a set of mutants defective in vein patterning, and our analysis suggests that they identify novel components in both auxin transport and response. Leaves of the *autobahn* mutant have multiple midveins. Expression of the auxin responsive reporter gene *DR5::GUS* and expression of procambial marker *AthB9::GUS* are consistent with defects to auxin transport. Treatment of wild type with the auxin influx inhibitor NOA partially phenocopies the *abn* phenotype and the *abn* phenotype is suppressed by *aux1*; both results are consistent with *abn* being defective in auxin influx as a result of AUX1 mislocalization. Leaves of the *forked1 (fkd1)* mutant show lack distal vein junctions, resulting in an open vein pattern. The *FKD1* gene encodes a protein with a PH domain, suggesting a possible role in membrane trafficking. Moreover, the *FKD1* gene is expressed in a manner consistent with it having an integral role in auxin canalization.

**(018) Differential biological availability of iron to green algae.**

Weger, H.G.[1], Matz, C.J.[1,3], Walker, C.N.[1], Fink, M.B.[1] and Treble, R.G.[2] [1]Department of Biology, University of Regina, Regina, SK. [2]Saskatchewan Health, Regina, SK. [3]Present address: Dept. of Anatomy & Cell Biology and Toxicology Centre, Univ. of Saskatchewan, Saskatoon, SK.

HBED is a very strong $Fe^{3+}$ chelator, and unlike Strategy I vascular plants, iron-limited cells of the green alga *Chlamydomonas reinhardtii* were unable to access iron present as $Fe^{3+}$-HBED. However, $Fe^{3+}$ chelated with HEDTA (a weaker chelator) was rapidly taken up by iron-limited cells. *Chlamydomonas* ferric reduction rates with $Fe^{3+}$-HBED were 15% of the rate observed with $Fe^{3+}$-HEDTA, suggesting that low reduction rates with $Fe^{3+}$-HBED might be one factor in the low rate of iron acquisition. By contrast, iron-limited cells of the green alga *Chlorella kessleri* were able to rapidly assimilate $Fe^{3+}$ chelated by HBED, although ferric reduction rates with $Fe^{3+}$-HBED were 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 (and siderophore analogue) DFB mesylate and the cyanobacterial siderophore schizokinen. These results suggest that green algal species differ in their abilities to access tightly complexed $Fe^{3+}$, and that aquatic iron bioavailability is a function of species.

**(019) Growth and photoinhibitory responses of *Arabidopsis* and contrasting ecotypes of *Thellungiella salsuginea*.**

Gray, G.R. and Khanal, N. Department of Plant Sciences, University of Saskatchewan. Saskatoon SK S7N 5A8.

*Thellungiella salsuginea*is an emerging model plant species which displays a high sequence similarity to that of *Arabidopsis.* The Shandong ecotype of *Thellungiella* grows in the warm-temperate, semi-humid coastal areas of eastern China with temperatures ranging from -5° C to 31° C. In contrast, the Yukon ecotype thrives in the sub-arctic and semi-arid regions of the Yukon Territories in Canada, with temperatures ranging from -18° C to 14° C. We have examined these contrasting ecotypes of *Thellungiella* and *Arabidopsis* under growth regimes optimal for each species/ecotype. Our data indicate differences in growth between the species/ecotypes were evident within each growth regime. Furthermore, there was also a difference in growth characteristics across growth regimes. Changes in pigmentation in the species/ecotypes displayed a similar trend across growth regimes. In addition, a differential tolerance to photoinhibition at low temperature was observed in the species/ecotypes between growth regimes. Preliminary experiments have revealed that this may be attributable to differential photosynthetic responses between these species/ecotypes.

**(020) Role of *MP* mediated auxin signaling and *PIN1* mediated auxin transport in leaf vascular pattern formation**

Schuetz, M., Wenzel, C., and Mattsson, J.Department of Biological Sciences, Simon Fraser University, Burnaby, B.C., Canada

Xylem and phloem constitute the two tissue types of the plant vascular system. The phytohormone auxin has been implicated in the process of vascular differentiation for a long time and the molecular genetic basis of auxin mediated vascular patterning and differentiation are currently beginning to unfold. Genetic evidence links the Arabidopsis *MONOPTEROS* (*MP*) and *PIN FORMED1* (*PIN1*) genes to the patterning of leaf veins. We have assessed the dynamics of *MP* and *PIN1* expression during vascular patterning in Arabidopsis leaf primordia to evaluate their potential roles in this process. Both genes undergo a dynamic process of gradual refinement of expression before overt vascular differentiation. We found evidence that *MP* expression can be activated by auxin exposure and that *PIN1* as well as DR5::GUS expression is defective in *mp* mutant leaves. Taken together the results suggest a feedback regulatory loop that involves auxin, *MP* and *PIN1* and provides support for the canalization of auxin flow hypothesis.

**(021) TRICHOMELESS1 controls trichome patterning by suppressing the expression of *GLABROUS1* in *Arabidopsis***

Chen, J.G., Zeng, Q., and Wang, S. Department of Botany, University of British Columbia, Vancouver, BC V6T 1Z4, Canada

Trichome cell specification is directly controlled by a homeodomain protein, GLABRA2 (GL2), in Arabidopsis. The expression of *GL2* is controlled by a trichome initiation activator complex whose transcriptional activity is regulated by trichome initiation and patterning negative regulators. Here we report the identification of TRICHOMELESS1 (TCL1) as a novel negative regulator. A dominant mutant with elevated expression of *TCL1* has a glabrous phenotype, whereas a loss-of-function mutation in *TCL1* confers ectopic formation of trichomes. We further demonstrated that TCL1 and CPC work synergistically to regulate trichome cell specification. Furthermore, overexpression of TCL1 specifically suppresses the expression of *GL1*, a critical component in the trichome initiation activator complex, whereas overexpression of TCL1-VP16 transactivator enhances it, suggesting that TCL1 negatively regulate trichome cell specification by directly suppressing the transcription of *GL1*. These findings offer another fine-tuning mechanism for the interaction and regulation between the trichome initiation negative regulators and the activator complex.

## Concurrent Session V: Agriculture and Biotechnology

### (022) Agricultural biotechnology of crops for producing commercially valuable natural products.

De Luca, V.; Chen, J.; Hall, D.; Levac, D.; Murata, J. and Magnotta, M. Department of Biological Sciences; Brock University; St. Catharines, ON, L2S 3A1

Our laboratory uses the Madagascar periwinkle (*Catharanthus roseus*) that manufactures powerful anticancer drugs and Concord grape (*Vitis labrusca*) that makes aroma compounds and blue pigments, as models to compare the similarities and differences in cellular specialization required for accommodating their respective pathways. State of the art technology for harvesting individual cell types (laser capture microdissection and carborundum abrasion) has been used to obtain specialized cell factories in order to more easily study the properties that make them different. These differences have been exploited through the use of genomic, proteomic and metabolic profiling tools to study and identify biochemical pathways and their regulation. The results provide a fingerprint of the cellular specialization required to make and accumulate particular specialty chemicals in plants and we are using this fundamental knowledge for enhancing, modulating and modifying the chemistry of plants to produce more and better drugs, aromas and flavours.

### (023) Production of human insulin and other biopharmaceuticals in oilseed crops: achievements and challenges.

Moloney, M.M., SemBioSys Genetics Inc., Calgary, AB.

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### (024) Biosafety of transgenic soybean using proteomic technology

Natarajan, S. Ph.D., U.S. Department of Agriculture, Agricultural Research Service, Soybean Genomics and Improvement Laboratory, Beltsville, MD 20705, USA

Soybean is the second most important cash crop in the US with an estimated annual value of $17.7 billion and provides an inexpensive source of protein for humans and animals. However, modifications of several nutritional deficiencies in soybean would make them a more valuable and nutritious protein source. One way for the US to remain competitive in the world soybean market is through the production of high quality soybeans using transgenic technology in order to achieve rapid benefits that are not available through plant breeding. Transgenic soybean produced approximately $1 billion of income through savings in the production costs associated with the use of Roundup Ready soybean. However, there are consumer concern and issues about the biosafety of

transgenic soybeans, which has caused a loss of export income to the US. There are literatures available on genetic and transgenic approaches applied to eliminate allergens and anti-nutritional proteins. However, there is essentially very little information is available regarding collateral or unintended effects of these transgenic modifications. Such information might serve to allay consumer concerns about the biosafety of transgenic soybean consumption. Since transgene integration in plants is mostly random, it is possible that soybean metabolism may be altered due to cis or trans acting effects of transgene expression, which may be beneficial or harmful. Therefore, the goals of the risk assessment program at USDA, Beltsville, MD are to apply suitable proteomics technologies that will be used to quantify any differences between soy seeds derived from transgenic vs. non-transgenic soybean. This project will provide unprecedented data and information on protein variation in soybean seeds and would enable regulators (USDA-APHIS) who are required to make science-based decisions in their assessment of transgenic soybeans. This information is also extremely important for determining the existence of any potential biosafety concerns and issues associated with transgenic soybean and will eventually lead to enhance consumer confidence, acceptability of US soybeans and for enhancing the soybean export market.

**(025) Evaluation of promoter activity and over-expression of a rice peroxidase gene in transgenic carrot (*Daucus carota* L.)**

Wally, O., Jayaraj, J. and Punja, Z.K.. Department of Biological Sciences, Simon Fraser University, 8888 University Drive, Burnaby, BC V5A 1S6, Canada.

Expression levels of four constitutive promoters (CaMV 35S, double 35s (D35S), maize (*ubi1*) and Arabidopsis (*ubq3*) ubiquitin) and three tissue specific promoters (mannopine synthase, agropine synthase and *rol*D) were quantified in carrot tissues using *Gus*A expression. Histological staining and flurometric assays were performed on 5 independent lines for each promoter construct. The *ubi1* and *ubq3* promoters had the highest GUS activities in carrot roots, while *ubq3*, *ubi1* and D35S had the highest activities in shoots. A rice peroxidase gene (*POC1*) was over-expressed in carrot driven by the *ubi1* promoter with *bar* as the selectable marker. Resistance to two fungal pathogens in 7 transgenic lines was tested using detached petiole assays. Five of the *POC1* over-expressing lines had significant tolerance to *Botrytis cinerea* (95-65% reduction in disease symptoms after 7 days compared to controls). These same lines had 55 - 70% reduction in development of *Sclerotinia sclerotiorum*.

**(026) Opium poppy: blueprint for an alkaloid factory**

Facchini, P.J. , Department of Biological Sciences, University of Calgary, Calgary, Alberta, T2N 1N4

Opium poppy (*Papaver somniferum*) produces a large number of benzylisoquinoline alkaloids, including the narcotic analgesics morphine and codeine, and has emerged as a versatile model system to study alkaloid metabolism in plants. We have taken a holistic strategy – involving biochemical, cellular, molecular genetic, genomic, and metabolomic approaches – to draft a blueprint of the fundamental biological platforms required for an opium poppy cell to function as an alkaloid factory. The capacity to synthesize and store alkaloids requires the cooperation of three phloem cell types – companion cells, sieve elements, and laticifers – in the plant, but also occurs in dedifferentiated cell cultures. We have assembled an opium poppy expressed sequence tag (EST) database based on the attempted sequencing of more than 30,000 cDNAs from elicitor-treated cell culture, stem, and root libraries. Approximately 23,000 of the elicitor-induced cell culture and stem ESTs are represented on a DNA microarray, which has been used to examine changes in transcript profile in cultured cells in response to elicitor treatment, and in plants with different alkaloid profiles. FT ICR-MS and $^{1}$H-NMR spectroscopy are being used to detect and correlate differences in metabolite profiles. Several new genes involved in the biosynthesis and regulation of alkaloid pathways in opium poppy have been identified using genomic tools.

## *Concurrent Session VI: Gene Stability and Expression*

### (027) Somatic and transgenerational changes in genome stability of salt-exposed plants

Boyko, A. and Kovalchuk, I. Department of Biological Sciences, University of Lethbridge, Lethbridge, Alberta, Canada

The influence of salt stress on genome stability was analyzed using transgenic *A. thaliana* plants. Plants were grown on MS media supplemented with NaCl for 3 weeks and then propagated into two subsequent generations. The genotoxic influence of salt stress on somatic tissue was reflected by an increase of the level of DNA strand breaks, 3.9-fold induction of recombination rate (RR) and elevated level of *AtRad51*. Transgenerational effect of salt was reflected by 2.2-fold increase in spontaneous RR, increase in *AtRad51* and decrease of *AtKu70* transcriptional activity, similar levels of strand breaks, changes in methylation pattern and elevated tolerance to NaCl and MMS. Most of these effects were substantially reduced in the 2$^{nd}$ generation, when plants were propagated without stress. Our data suggest that exposure to salt results in both, somatic and meiotic response and that changes in the genome stability, methylation and stress tolerance are transient in nature.

### (028) Gene expression profiling of microspore embryogenesis in *Brassica napus*

Malik M.R., Wang F, Zhou N, Rahman M, Ferrie AMR, Krochko JE
Plant Biotechnology Institute, National Research Council of Canada, 110 Gymnasium Place, Saskatoon, SK, S7N 0W9.

Isolated microspores of *Brassica napus* are developmentally programmed to form gametes; however, microspores can be induced through stress treatments to undergo appropriate divisions and form embryos. We are interested in the identification and isolation of factors and genes necessary, and/or sufficient, for the induction of embryogenesis in isolated microspores. Standard, normalized and subtractive cDNA libraries were constructed from freshly isolated microspores (0h) and microspores cultured for 3d, 5d or 7d under embryogenesis-inducing conditions. Library comparison tools revealed shifts in metabolism across this time-course. Extensive PCR-based expression profiling identified 16 genes as unequivocal molecular markers for microspore embryogenesis in *B. napus*. The quantitative expression values of several of these molecular marker genes were predictive of embryogenic potential in *B. napus* cultivars. Microarray analyses with a small custom array (ABA-, seed-and embryogenesis-related genes) and a *B. napus* 10K array identified genes differentially expressed at the 3d and 7d stages of culture. Functional characterization of two novel embryogenesis-related unknown genes has been initiated.

**(029) Elucidation of acetylenase functionality**

Gagne, S.[1], Gray, G.R.[1,2], and Covello, P.[1,3] [1]Department of Biochemistry, University of Saskatchewan. Saskatoon SK S7N 5E5. [2]Department of Plant Sciences, University of Saskatchewan. Saskatoon SK S7N 5A8. [3]National Research Council of Canada, Plant Biotechnology Institute, Saskatoon SK S7N 0W9.

Oleate desaturases (FAD2) belong to a family of enzymes capable of introducing *cis* double bonds between C12-C13 in (Δ9 unsaturated) oleate esters. Acetylenases are a subset of FAD2 enzymes which introduce a triple bond in the C12-C13 position of linoleate. A total of 50 protein sequences were used to compare the two families of enzymes resulting in the identification of 11 amino acid residues which are conserved within each separate family but differ between the two families. Targeted amino acid residues were then altered by site-directed mutagenesis to test their role in fatty acid modification. Specifically, the wild type acetylenase, Crep1 from *Crepis alpina,* and a number of point mutants have been expressed in *Saccharomyces cerevisiae*, followed by fatty acid analysis of the resulting cultures. Preliminary results point to the importance of certain amino acid position in acetylenase activity.

**(030) Dual targeting of $NADP^+$-isocitrate dehydrogenase to mitochondria and chloroplasts.**

McKinnon, D.J.[1], Froehlich, J.E.[2], Brandizzi, F.[2] and Gray, G.R.[1] [1]Department of Plant Sciences, University of Saskatchewan. Saskatoon SK S7N 5A8 Canada. [2]MSU-DOE Plant Research Laboratory, Michigan State University, East Lansing, MI 48824-1312 USA

Mitochondrial and plastid proteins are encoded in the nucleus and subsequently imported into the organelles via active protein transport systems. While usually highly specific, many proteins are dual targeted to both organelles. Our previous studies examining the gene encoding the mitochondrial isoform of isocitrate dehydrogenase (mtICDH) have identified two ATG start sites, ATG1 and ATG2. Translation from ATG1 generates a precursor protein that has an N-terminal mitochondrial targeting sequence followed by a chloroplastic transit peptide sequence, designated mtICDH. Translation from ATG2 generates a smaller precursor protein containing only an N-terminal chloroplastic transit peptide, designated ICDH2. *In vitro* import assays using isolated pea chloroplasts, confirms that ICDH2 binds and is imported into chloroplasts. Conversely, *in vitro* import assays using isolated tobacco mitochondria, reveals that ICDH2 is not imported into mitochondria. Presently, we are characterizing the import behavior of mtICDH using confocal microscopy to determine how dual targeting is regulated *in vivo*.

**(031) Genetic and epigenetic regulation of genome stability in plant response to stress**

Boyko, A., Zemp, F., Kathiria, P., Titov, V. and Kovalchuk, I. Department of Biological Sciences, University of Lethbridge, Lethbridge, AB Canada.

Plants are sessile organisms not capable of avoiding stress and thus using complex protection mechanisms, including acclimation and adaptation. We analyzed the influence of a variety of abiotic (NaCl, heavy metals, exposure to UV, rose Bengal, flood, drought) and biotic (viral and bacterial pathogens) stresses on plant genome stability and found that the majority of stresses (all but drought) resulted in increase of spontaneous levels of homologous recombination in the progeny of exposed plants. Further propagation of plants without stress seems to reverse the process, whereas propagation with stress maintains the increase in recombination in several generations. These progenies had increased tolerance to stress and exhibited differential methylation patterns. We hypothesize that stress results in the production of small interfering RNAs that "reestablish" the methylation pattern in gametes. We found that response to stress elaborates signaling molecules capable of warning the non-exposed tissue and leading to transgenerational effects.

**(032 Non-canonical stromal targeting of an Arabidopsis mitogen-activated protein kinase kinase**

Ellis, B.E., Michael Smith Laboratories, University of British Columbia, Vancouver, V6T 1Z4, British Columbia, Canada

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**(033) *Scutellaria racemosa*: An important species for study of medicinal compounds**

Cole I. B. and Murch, S.J. Unit 3, UBC - Okanagan, 3333 University Way, Kelowna, BC V1V 1V7

Medicinal plants have very unique secondary metabolism that may not be fully represented in either genome database and finding a suitable model system for study of a medicinal plant genome has been incredibly difficult. The genus *Scutellaria* is a rich source of plants for treatment of a wide range of human ailments and representative species from many parts of the world are used as medicines. *Scutellaria baicalensis* is the most commonly prescribed plant in Traditional Chinese Medicine, *Scutellaria lateriflora* has a long tradition of use by the indigenous people of North America and there is traditional knowledge of the use of *Scutellaria racemosa* in Central America. The current studies were undertaken to compare medicinal plants from *Scutellaria* and other families for genome size, chemical diversity and the potential for application of plant biotechnology techniques. Many medicinal plants have extraordinarily large

nuclear DNA compliments and are not suitable for genomic studies. Other medicinal plants mainly produce phytochemicals from specific classes. Scutellaria racemosa is unique in that the species has a relatively small nucleus with an estimated genomic DNA size of 366 Mbp coupled with an extensive secondary metabolism that produces more than 4,000 unique compounds including melatonin, serotonin, baicalin, baicalein, wogonin and other medicinal metabolites. These results indicate that Scutellaria racemosa may be a good biological resource for future studies of the regulation of plant secondary metabolism.

# Poster session abstracts:

## *Section I: Metabolic Pathways*

### (034) Regulation of monoterpene metabolism in Lavender

Boeckelmann, A., Mahmoud, S. S. Centre for Natural Product Research, UBC Okanagan, Kelowna, BC.

Lavender is mainly grown for its essential oil, which has extensive applications in cosmetics and alternative medicine. The medicinal and olfactory properties of lavender essential oil are attributed to monoterpenes, a class of low molecular weight ($C_{10}$) isoprenoids. The monoterpene composition is species specific, but is also influenced by climate and agricultural practices.
We analysed the monoterpene composition of several lavender species from three different locations in the Okanagan. Our data confirm previous observations that lavender scent is a function of the monoterpenes camphor, limonene and linalool: while high quality oils contain high amounts of limonene and linalool but little camphor, oil scent and quality deteriorates with increasing camphor levels. Our aim is to isolate and identify the genes that mediate camphor synthesis in lavender. High inter- and intra-species homology between monoterpene synthases advocates analogous camphor synthetic pathways in sage and lavender. Hence, we propose to isolate camphor synthases through a homology screen of a lavender cDNA library, using sage genes as probes. Isolation of candidate genes will be followed by identification of the gene product in a functional assay. In order to locate camphor synthesis, the expression pattern of identified camphor synthases will be traced by hybridization. The insights acquired here will build the basis for future studies assessing the regulation of monoterpene synthase gene expression.

### (035) Using genomics approaches to characterize essential oil production in *Lavandula angustifolia* (English lavender)

Lane, A. and Mahmoud, S.S. Centre for Natural Product Research, UBC Okanagan, Kelowna, BC.

*Lavandula angustifolia* (English lavender) is best known for its pleasant odor and its popularity as an herbal remedy for a variety of medical conditions. These factors, combined with the high value of the plants essential oils (mono and sesqui terpenes), have lead to a steady increase in global lavender cultivation. In order to enrich our understanding of terpenoid production in lavenders, two cDNA libraries were constructed and 10,000 ESTs were sequenced. With this resource, we are now in a position to pursue a range of questions which are central to the mandate of our lab. Specifically, we aim to clone and characterize the genes involved in terpenoid synthesis pathways in lavender,

understand the transcriptional regulation of these pathways, and elucidate the mechanisms by which terpenoids are transported within the plant cell. These findings will facilitate the development of improved cultivars, metabolic engineering of terpenoid biosythesis pathways, and enhancement of lavender breeding programs

## *Section II: Rhizosphere and Pathogen Interactions*

**(036) Combinatorial expression of chitinase and lipid transfer protein genes in transgenic carrot plants enhances resistance to foliar fungal pathogens.**

Jayaraj, J and Punja, Z.K. Department of Biological Sciences, Simon Fraser University, 8888 University Drive, Burnaby, British Columbia, V5A 1S6, Canada.

Two pathogenesis-related protein cDNAs consisting of a barley chitinase (*chi-2*) and a wheat lipid-transfer-protein (*ltp*) were introduced into carrot plants by *Agrobacterium*-mediated transformation using the phosphinothricin acetyl transferase (*bar*) gene as a plant selectable marker. Over 80% of the regenerated plants displayed resistance to the herbicide Liberty (0.2%, v/v) in the greenhouse and were confirmed to be positive for the transgene(s) by PCR and RT-PCR. Southern analysis of transgenic plants revealed from one to three copies of the transgenes. Northern analysis and immunoblotting confirmed the expression of transgene proteins in nearly 70% of the plants, with variable expression levels among the lines. Inoculation of plants with *Alternaria radicicola* and *Botrytis cinerea* showed significantly higher resistance to these pathogens in lines expressing both genes compared to single gene transformants. The levels of disease reduction in transgenic plants expressing both genes was around 95% for *Botrytis* and 90% for *Alternaria* infection.

## *Section III: Metabolism and Stress Pathways*

**(037) Assessment of expression and function of *alpha-dioxygenase* in *Arabidopsis* under salinity stress**

Aung, T.S.T.[1], Plant, A.L.[1] and Hamberg, M.[2] [1]Department of Biological Sciences, Simon Fraser University, Burnaby, Canada. [2] Department of Medical Biochemistry and Biophysics, Karolinska Institutet, Stockholm, Sweden.

Alpha-dioxygenases (α-Dox) are involved in fatty acid α–oxidation in plants. The objective of this study is to assess the expression and function of the two α-Dox genes (α-Dox1 and α-Dox1) under salinity stress in Arabidopsis. Alpha-Dox1 is constitutively expressed in the root but not in shoot tissues whereas α-Dox2 is expressed in both root and shoot tissues but at a much lower level than α-Dox1. The expression of α-Dox in

both root and shoot tissue was enhanced by salt stress. Abscisic acid and salicylic acid are major hormonal signals that regulate α-Dox expression in Arabidopsis. Enzyme assays have revealed increased α-Dox products in salt stressed root and shoot tissues and α-Dox1 is responsible for forming most α-Dox products. Alpha-Dox knock-out and over-expressing lines are being assessed under salt and oxidative stress conditions to elucidate a role for α-Dox enzymes in salt stressed plants.

**(038) Functional analysis of the RCD-SRO (RADICAL INDUCED CELL DEATH-SIMILAR TO RCD) gene family in salt treated roots.**

Babajani, G. and Plant, A L. , Department of Biological Sciences, Simon Fraser University, Burnaby, British Columbia, Canada.

JWL-26 is a novel salt-regulated gene that was isolated from tomato roots. BLAST-based homology searches revealed similarity between JWL-26 and several *Arabidopsis* proteins that include RCD1 (Radical-induced Cell Death 1) and the SRO (Similar to RCD-One) family members. These proteins play a role in regulating several hormonal signalling cascades involved in programmed cell death in Arabidopsis. JWL-26 is most closely related to the SRO5 (At5g62520.1) protein. Two full length cDNA clones (LeRCD1-like 1 and LeRCD1-like 2) with similarity to RCD1 were obtained from TIGR. Together with JWL-26 these cDNAs are being analysed to determine their function in salt-stressed roots. Transgenic lines have been generated to overexpress the JWL-26, LeRCD1-like 1 and LeRCD1-like 2 cDNAs in WT, an SRO5 knock out line and the rcd1 mutant, respectively. Preliminary data obtained to date will be presented.

**(039) Interactions between abscisic acid and ethylene in salt-stressed tomato (*Lycopersicon esculentum* Mill) roots.**

Kwok, A. and Plant, A L. , Department of Biological Sciences, Simon Fraser University, Burnaby, BC Canada.

Experiments to characterize the role of abscisic acid (ABA) and ethylene (ET) in regulating a salt-responsive alpha-dioxygenase (*Leα-DOX)* gene uncovered a positive regulatory role for ET in salt-stressed tomato roots. *Leα-DOX1* expression is markedly responsive to ET and we have previously found that ABA antagonizes ET-responsive expression of *Leα-DOX1*. To further investigate the role of ET and the interaction between ET and ABA in salt-stressed roots, the expression of genes encoding transcription factors (*JAMYC2, ERF1, ERF3*) or enzymes for the biosynthesis of ET (*ACS3, ACO1*) and ABA (*NCED1*) was examined. A positive regulatory role for both ABA and ET was substantiated by the salt-induced expression of *ACS3, ACO1, NCED1, JAMYC2, ERF1* and *ERF3* in roots. Furthermore, the expression of *ACO1* and *JAMYC2* was negatively regulated by ABA. Thus, in salt-stressed roots ABA appears to be a "dominant" signal capable of suppressing ET-responsive gene expression.

**(040) Inter- and intraplant stress signaling in *Arabidopsis thaliana***

Zemp, F.J.[1]., Blevins, T.[2]., Bui, T.M.[1]., Meins, F.[2]., Kovalchuk, I.[1],[1]University of Lethbridge, Lethbridge, Alberta, Canada. [2]Friedrich Miescher Institute, Basel, Switzerland.

Stress signaling and response has been discovered to involve a number of intricate molecular pathways. Recently, small RNA species have arisen as important regulators of response and, possibly, resistance. Here we have found the regulation of stress-induced miRNAs in uninfected, systemic tissue 24 hours after biotic and abiotic stress exposure, with two of these miRNAs targeting factors important in auxin response. The regulation of distal miRNAs could suggest a homeostatic or preparatory response to the stress. Further, we have discovered the existence of an interplant signal communicating the genomic instability triggered by UV or X-ray irradiation. These unexposed (bystander) plants exhibit increased homologous recombination rates when grown in media shared with irradiated plants. The communication of radiation-induced instability is a completely novel phenomenon in plant biology.

## *Section IV: Development and Physiology*

### (041) Seasonal patterns of leaf gas exchange in response to moisture availability in riparian Cottonwood hybrids (*Populus* spp.)

Letts, M.G.[1], Rood, S.B. [2], Johnson, D.R.E. [1], Phelan C. [2] and Pearce D.W. [2], [1]Department of Geography, University of Lethbridge, Lethbridge AB Canada, T1K 3M4; [21]Department of Biological Sciences, University of Lethbridge, Lethbridge AB Canada, T1K 3M4

*Populus* spp. tree species often dominate riparian woodlands in arid and semi-arid regions of western North America. The ecological functions served by these phreatophytic trees are threatened in many regions by river damming, water diversion projects and livestock grazing. In this study, we examine seasonal patterns of photosynthesis and physiological acclimation to changing moisture availability, in a southern Alberta Cottonwood grove, using leaf gas exchange, leaf reflectance and chlorophyll fluorescence techniques. A late season irrigation experiment resulted in higher stomatal conductance and lower water use efficiency in irrigated trees, but had no significant effect on net photosynthesis rates. Sex-related differences in physiological acclimation to soil moisture deficits will also be considered.

### (042) Inorganic and organic nitrogen forms in forest soils and their contributions to plant nutrition.

Metcalfe, R. and Hawkins, B. Centre for Forest Biology, University of Victoria, Victoria, BC Canada.

Soil nitrogen (N) is a major factor limiting forest growth. While inorganic N forms have been the focus of plant N nutrition studies, a growing body of research has shown direct plant uptake of organic N forms (including amino acids) is common across the plant kingdom. However, there are few quantitative examinations of the contribution amino acid N makes to plant nutrition. Main objectives of this study are: to determine relative availabilities of amino acid N to inorganic N in local forest soils; to compare uptake of inorganic versus organic N using two economically important conifers *Picea sitchensis,* and *Pseudotsuga menziesii,* and their competitors *Vaccinium* spp. *and Rubus spectabilis*; and to determine how mycorrhizal associations affect N form uptake. Growth and biomass results from plants grown on inorganic and organic (amino acid) N forms will be presented. This information may help explain species abundance patterns, plant diversity and competitive interactions.

**(043) Latitudinal variations in ecophysiological traits of *Populus balsamifera* populations across the boreal forest of North America**

Soolanayakanahally, R. Y[1]. Silim, S[2]. and Guy, R. D[1]. [1] Department of Forest Sciences, University of British Columbia, Vancouver, BC, Canada [2] Shelterbelt Centre, PFRA-AAFC, Indian Head, Saskatchewan, Canada

Variation in plant functional traits results from evolutionary and environmental drivers that operate at a variety of different scales. We describe patterns of ecophysiological trait variation and trait correlations among populations in relations to several environmental and trade-off axes. In so doing we highlight evidence demonstrating that patterns of trait variation across resource and environment gradients (light, water, nutrients, photoperiod, frost free days and temperature) probably reflect adaptation. Significant differences are seen among populations for photosynthesis (A), transpiration (E), stomatal conductance ($g_s$), stomatal density, carbon isotope discrimination ($\delta^{13}C$) and foliar N content. A/gs, A/E, $\delta^{13}C$ and foliar N had strong correlations ($p<0.05$) with latitude and longitude. Population-wide stomatal density ($r = -0.205$) and petiole length ($r = -0.561$) were inversely correlated with latitude. Longer leaf longevity was associated with greater leaf mass, lower photosynthetic capacity, and significant difference in photosynthetic nitrogen use efficiency. These trade-offs suggest that plants adapt N allocation to increase either the rate or duration of carbon assimilation but not both.

## *Section V: Agriculture and Biotechnology*

### (044) Improved somatic embryogenesis yield and biolistic transformation efficiency in *Triticum aestivum* (Superb)

Greer, M.S. [1] Eudes F.,[2] Kovalchuk, I.[1] [1.] University of Lethbridge, department of biology[2.] Lethbridge Research Station, department of cereal biotechnologies

The capability to introduce novel genes into new systems has been shown to be a very powerful tool in humanitarian, scientific and commercial fields. Transformation of crops is still not a routine process and any improvement would of great asset. Our work shows that modification of the media composition leads to 2-2.5-fold increase in the number of somatic embryos regenerated from dissected wheat scutella. We also show that exposure to certain salts are capable of improving the transformation efficiency obtained from microparticle bombardment of wheat tissue. Currently we are analyzed the efficiency of multiple transgene insertions in wheat. In our approach we used the delivery of 3 independent cassettes of transgenes. Preliminary data suggest the increase in the frequency of delivery of these transgenes into wheat tissue. The influence of these salts seems, however, to be cultivar-specific.

## *Section VI: Gene Stability and Expression*

### (045) Cloning of linalool synthase promoter in Lavender

Biswas, K.K. and Mahmoud S.S. Centre for Natural Product Research, UBC, Okanagan, Kelowna, BC.

Monoterpenes are best known as constituents of the essential oils and defensive resins of aromatic plants. They are also found in non-aromatic plants, where they influence the aroma of flowers and fruits. In aromatic species, such as lavenders, monoterpene metabolism is mainly restricted to the glandular trichomes found on the surfaces of the aerial parts of the plant. Currently, little is known about genetic factors that control production of these compounds. We are developing *Lavandula angustifolia* (English lavender) as a model system for studying regulation of monoterpenoid biosynthesis. This plant produces large quantities of linalool, which is also present in fruits and flowers of most other plants. Our strategy includes investigating the role of the promoter regions in linalool synthase (LlS) gene expression. Here we report cloning and sequence determination for a putative LlS promoter from L. angustifolia. The cloned fragment includes regulatory regions typical of plant promoters, including a TATA box and several transcription factor (TF) binding domains. We will use this promoter in reporter gene studies to elucidate its contribution to LlS gene expression. The promoter could also be used for the bioengineering of monoterpenoid metabolism in plants.

# Abstract list

**(001) Regulation of peppermint essential oil biosynthesis - development of a mathematical model and experimental validation**
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**(002) Dissecting terpenophenolic biosynthesis in *Humulus lupulus* (hops)**
Page, J. and Nagel, J

**(003) Metabonomics & chemodiversity in Endangered Plants**
Murch, S. J.

**(004) Genomics of essential oil and aroma formation in lavenders**
Mahmoud, S.S.,

**(005) Functional genomics and the biosynthesis of artemisinin**
Covello, P.S., Teoh, K.H., Polichuk, D.R. Reed, D.W. and Nowak, G.

**(006) Biochemical changes and gene expression induced in ginseng roots infected by *Fusarium equiseti.***
Punja, Z.K., Goswami, R.S. and Rahman, M.

**(007) Aspect-related differences in seasonal acclimation to environmental stress in *Pinus flexilis* (Limber Pine).**
Letts, M.G. and Johnson,

**(008) Physiological diversity of ectomycorrhizas on Douglas-fir seedlings regenerating after wildfire or clearcutting**
Jones, M

**(009) A comparison of ammonium, nitrate and proton net fluxes along seedling roots of Douglas-fir and lodgepole pine**
Hawkins, B.J.

**(010) The role of root epidermal suberin in partial resistance of soybean to Phytophthora root rot**
Peterson, C A., Ranathunge, K. , Fang, X., Thomas, R.` Bernards, M A. and Gijzen, M.

**(011) Increased trans-generational tolerance in pathogen infected plants**
Kathiria, P. Boyko, A. Zemp, F. and Kovalchuk,

**(012) Chemical diversity in plant defense against Insects: Terpenoid Synthases and cytochrome P450 in conifers**
Bohlmann J.

**(013) iTRAQ analysis of the grape proteome at ripening initiation reveals a previously unidentified protein controlling a downstream step in anthocyanin biosynthesis.**
Lund, S.T., Lücker, J., Elliot, M., and Smith, D.

**(014) Molecular networks in plants: extraction from literature and pathway analysis**
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**(015) ABC transporters at the plant surface export protective lipids of the cuticle.**
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