



Fotografía: Gentileza Dr. Almada.

2 al 5
DICIEMBRE
2013

VIII Reunión de Biología Vegetal

Pucón. Región de la Araucanía. CHILE

biología
vegetal

VIII Reunión



INNOVAR ES MEJORAR...
Ejecutando la Estrategia Regional de Innovación, O'Higgins crece.

Proyecto financiado a través del Fondo de Innovación para la Competitividad del Gobierno Regional de O'Higgins y su Consejo Regional enmarcado en la Estrategia Regional de Innovación.

**VIII Reunión
de Biología Vegetal
2013**

Comisión Organizadora – INIA-Rayentué/CEAF

Dr. Boris Sagredo (Presidente de la Comisión Organizadora)

Dra. Paula Pimentel (Coordinadora)

Dr. Ariel Salvatierra

Dra. Adriana Bastías

Dra. Pamela Rojas

Dr. Rubén Almada

Dr. Mauricio González

Dr. Manuel Pinto

Dr. Patricio Hinrichsen

Webmaster

Simón Solís

INSTITUCIONES ORGANIZADORAS



PROYECTO PATROCINANTE



Comisión Científica

Dr. Rubén Almada (INIA-CEAF)

Dra. Adriana Bastías (INIA)

Dra. Francisca Blanco (UNAB)

Dr. Reinaldo Campos (UNAB)

Dra. Liliana Cardemil (UCHile)

Dr. Basilio Carrasco (PUC)

Dra. Marely Cuba (UDEEC)

Dra. María Laura Federico (CGNA)

Dr. Carlos Figueroa (UDEEC)

Dra. Lida Fuentes (CREAS)

Dr. Felipe Gaínza (INIA-CEAF)

Dr. Enrique González (UTALCA)

Dr. Mauricio González (INIA)

Dra. Wendy González (UTALCA)

Dr. Michael Handford (UCHILE)

Dr. Raúl Herrera (UTALCA)

Dr. Patricio Hinrichsen (INIA)

Dr. Cristián Ibáñez (USERENA)

Dra. Carolina Lizana (UACH)

Dr. Claudio Meneses (UNAB)

Dra. Andrea Miyazaka (UNAB)

Dra. Alejandra Moya (UTALCA)

Dr. Claudio Pastenes (UCHILE)

Dr. Jorge Pérez (UTALCA)

Dra. Paula Pimentel (INIA-CEAF)

Dr. Manuel Pinto (INIA-CEAF)

Dr. Patricio Ramos (UTALCA)

Dra. Pamela Rojas (INIA)

Dra. Marlene Rosales (PUC)

Dr. Boris Sagredo (INIA-CEAF)

Dra. Erika Salazar (INIA)

Dr. Ariel Salvatierra (INIA-CEAF)

Dra. Claudia Stange (UCHILE)

Dra. Carolina Torres (UTALCA)

INSTITUCIONES AUSPICIADORAS



Scientific Program

Monday -December 2, 2013

Hotel Check-in starts at 12:00 PM - Gran Hotel Pucón Reception.

11:00 - 17:00 Registration - Gran Hotel Pucón Pre Foyer.

11:00 - 17:00 Poster Set Up (ODD numbered posters) - Lonquimay - Coñaripe Room

15:00 - 15:30 Opening Welcome - Araucanía Room

Pedro Bustos Valdivia, Director Nacional Instituto de Investigaciones Agropecuarias (INIA)

15:30 - 16:30 Plenary Lecture I - Araucanía Room

Dr. Thomas Davies – THE STRUCTURE AND EVOLUTION OF COMPLEX GENOMES: THE FRAGARIA (STRAWBERRY) OCTOPLOIDS AND DECAPLOIDS. Department of Biological Sciences, University of New Hampshire, USA.

Chair: Raúl Herrera

16:30 - 17:00 Coffee Break – Lonquimay - Coñaripe Room

17:00 - 18:20 Oral Session - Araucanía Room

Chairs: Raúl Herrera and Boris Sagredo

17:00 QTL ANALYSIS FOR FRUIT QUALITY TRAITS IN PEACH [*Prunus persica* (L.) BATSCH].

Gerardo Nuñez, Alejandra Cifuentes, Reinaldo Campos-Vargas, Gamalier Lemus, Rodrigo Infante, Ariel Orellana and Claudio Meneses.

17:20 IDENTIFICATION AND CHARACTERIZATION OF PUTATIVE MARKERS DERIVED FROM A TRANSCRIPTOMIC APPROACH (RNA-SEQ) ASSOCIATED TO BERRY SIZE IN TABLE GRAPES.

Claudia Muñoz, Alicia Sánchez, Alex Di Génova, Alejandro Maass, Mauricio González-Agüero, Ariel Orellana and Patricio Hinrichsen.

17:40 STUDY OF RIPENING AND CHILLING INJURY IN EARLY AND LATE VARIETIES OF PEACHES AND NECTARINES (*Prunus persica*) USING RNA-SEQ. Dayan Sanhueza, Claudio Meneses, Paula Vizoso, Christian Silva, Iván Balic, Ariel Orellana and Reinaldo Campos-Vargas.

18:00 LONG-DISTANCE GENES SILENCING BY ARTIFICIAL miRNAs. Daniela Quiroz, Catalina Álvarez, Álvaro Castro and Humberto Prieto.

20:00 - 21:30 Dinner - Gran Hotel Pucón, Calafquén Caburgua Restaurant

21:30 - 23:00 Poster Session I (ODD numbered posters) – Lonquimay - Coñaripe Room

Tuesday - December 3, 2013

7:00 - 11:30 Poster Set Up (EVEN numbered posters) – Lonquimay - Coñaripe Room.

7:00 - 8:45 Breakfast - Gran Hotel Pucón Calafquén Restaurant

9:00 - 10:00 Plenary Lecture II - Araucanía Room

Dr. Aurelio Gómez Cadenas - HORMONAL CROSSTALK IN ROOTS OF PLANTS SUBJECTED TO WATER-STRESS CONDITIONS - Departament de Ciències Agràries i del Medi Natural. Universitat Jaume I, Castelló de la Plana, Spain.

Chair: Adriana Bastías

10:00 - 11:20 Oral Session II- Araucanía Room

Chairs: Liliana Cardemil and Manuel Pinto

10:00 PHYSIOLOGICAL AND PRODUCTIVE RESPONSES MEDIATED BY NITROGEN AVAILABILITY IN *Colobanthus quitensis* and *Deschampsia antarctica* EVALUATED IN GLASSHOUSE. Claudia Rabert, Mario Díaz, Claudia Mella, Carla Alvear, Miren Alberdi, Marjorie Reyes and León Bravo.

10:20 STUDIES OF THE EXPRESSION OF ADAPTATIVE DIFFERENTIAL MECHANISM IN TWO GENOTYPES OF *Solanum peruvianum* L. DURING WATER STRESS. Oscar Arrey, Yazmina Stapung and Gerardo Tapia.

10:40 *Colobanthus quitensis*, A MODEL SPECIE OF GENOMIC RESPONSE TO EXTREME ENVIRONMENT. Marely Cuba-Díaz.

11:00 ASSESSMENT BY MICROSATELLITES OF GENETIC DIVERSITY OF CHILEAN POPULATIONS OF ALGARROBO (*Prosopis chilensis*) IN RESPONSE TO SALINE STRESS. Claus Westphal, Cristian Ibañez and Antonio González.

11:30 - 12:00 Coffee Break – Lonquimay - Coñaripe Room

12:00 - 13:00 Oral Session III – Araucanía Room

Chairs: Lida Fuentes and Mauricio González

12:00 GENE EXPRESSION CHANGES IN NEEDLES AND STEMS OF *Pinus radiata* ROOTSTOCK PLANTS OF TWO AGES WITH DIFFERENT ROOTING CAPABILITY. Carolina Álvarez, Luis Valledor, Patricia Sáez, Manuel Sánchez-Olate and Darcy Ríos.

12:20 CAN FCSEPLIKE REGULATES THE EXPRESSION OF CELL WALL DISSEMBLING GENES DURING RIPENING OF *Fragaria chiloensis* FRUIT? Luis Morales-Quintana, Daniela Urbina, Carlos Figueroa, Raúl Figueroa, Raúl Herrera and María Alejandra Moya-León.

12:40 STRUCTURAL CHARACTERIZATION OF TWO MADS TRANSCRIPTION FACTORS THAT BIND TO THE PROMOTER OF PrXTH, A GENE INVOLVED IN THE INCLINATION RESPONSE OF *Pinus radiata* D. Don. Verónica Latapiat, Luis Morales-Quintana, María Alejandra Moya león and Raúl Herrera.

13:15 – 14:45 Lunch – Gran Hotel Pucón Calafquén Restaurant

15:00 - 16:20 Oral Session IV - Araucanía Room

Chairs: Alejandra Moya and Claudio Pastenes

15:00 EFFECT OF MODIFIED ATMOSPHERE PACKAGING (MAP) ON RACHIS QUALITY OF “RED GLOBE” AND “FLAME SEEDLESS” VARIETIES. Christian Silva-Sanzana, Iván Balic, Adrián Moreno, Dayan Sanhueza, Patricio Olmedo, Bruno G. Defilippi, Reinaldo Campos-Vargas.

15:20 ROLES OF ANTIOXIDANT SYSTEMS ON APPLE SKIN BROWNING DISORDERS POSTHARVEST ASSOCIATED TO HIGH SOLAR IRRADIATION. Carolina Torres, Omar Hernández, Iván Razmilic and Alejandra Moya.

15:40 FLAVONOLS: MOLECULES THAT PLAY AN IMPORTANT ROLE IN THE GROWTH OF PINE. Patricio Ramos, María Alejandra Moya-León, Raúl Herrera.

16:00 EFFECT OF APPLICATION OF PECTIN DERIVED OLIGOSACCHARIDES ON THE ACCUMULATION OF ANTHOCYANIN IN BERRIES OF CABERNET SAUVIGNON. Daniel Villegas, Michael Handford and Alonso Pérez-Donoso.

16:30 - 17:00 Coffee Break – Lonquimay - Coñaripe Room

17:00 - 18:00 Technical Conference – Dr. Romilio Espejo. Advances in the CONICYT Project “Centro Nacional de Genómica y Bioinformática” promoting the application of Next Generation Sequencing in research and technology in Chile. Centro Nacional de Genómica y Bioinformática.

Chair: Rubén Almada.

18:00 - 19:00 Trend and Policy Conference – Dr. José Miguel Benavente. Visión estratégica de la Ciencia e Innovación en Chile. Consejero CNIC.

Chair: Boris Sagredo.

20:00 - 21:30 Dinner – Gran Hotel Pucón Calafquén Restaurant

21:30 – 23:00 Poster Session II (EVEN numbered posters) – Lonquimay & Coñaripe Room

Wednesday - December 4, 2013

7:00 - 8:45 Breakfast – Gran Hotel Pucón Calafquén Restaurant

9:00 - 10:00 Plenary Lecture III – Araucanía Room

Dr. Robert Martin - THE FASCINATING WORLD OF BERRY VIRUSES – MIXED INFECTIONS ARE THE NORM. USDA-ARS Horticultural Crops Research Unit, Corvallis, Oregon, USA.

Chair: Pamela Rojas

10:00 - 11:20 Oral Session V – Araucanía Room

Chairs: Marlene Rosales – Rubén Almada

10:00 DEVELOPMENT OF A MARKER-ASSISTED BREEDING PROGRAM FOR PRUNUS ROOTS-TOCK FOCUSED IN THE NEW CHILEAN AGRICULTURAL REQUIREMENTS. Felipe Gainza-Cortés, Verónica Guajardo, Ismael Opazo, Mauricio Ortiz, Rubén Almada, Paula Pimentel, Boris Sagredo, Manuel Pinto, José San Martín, Gamalier Lemus, Jorge Pinochet and Carlos Muñoz.

10:20 CONSTRUCTION OF A HIGHLY DENSE MAP OF SWEET CHERRY AND ITS COMPARATIVE ANALYSES WITH OTHERS PRUNUS LINKAGE MAPS. Carolina Klagges, Alejandra Guzmán, Levi Mansur, Eduardo Gratacós, Herman Silva, Lee A. Meisel.

10:40 OVERVIEW: IMPROVING SEMOLINA YELLOWNESS OF DURM WHEAT (*Triticum turgidum* L. VAR. DURUM) IN CHILE. Albert Schulthess, Karen Campos, Nicolás Jimenez, Iván Matus and Andrés R. Schwember.

11:00 CHARACTERIZATION OF CHOCLERO MAIZE (*Zea Mays* L.) POPULATIONS ASSESSED BY MORPHOLOGICAL TRAITS AND SSR MOLECULAR MARKERS. Erika Salazar, José Correa, María José Araya, Marco Méndez, Carolina Araya, Boris Sagredo and Basilio Carrasco.

11:30 - 12:00 Coffee Break – Lonquimay & Coñaripe Room

12:00 – 13:00 Oral Session VI - Araucanía Room

Chairs: Francisca Blanco and Patricio Hinrichsen

12:00 LIPOXYGENASE AND APPLE AROMA: GENE EXPRESSION OF LIPOXYGENASE FAMILY MEMBERS IN APPLE PEEL. Carolina Contreras and Randy M. Beaudry.

12:20 EXPRESSION ANALYSIS OF GIBBERELLING SIGNALLING GENES OF SEGREGANTS OF A “RUBY SEEDLESS” X “SULTANINA” POPULATION. Gonzalo Ravest, Sebastián Silva, Maribel Mamani and Patricio Hinrichsen.

12:40 PHOSPHATE SOLUBILIZING BACTERIA NATIVE PLANT GROWTH PROMOTING IN THE AREA OF LA SERENA, IV REGION OF COQUIMBO. Andrés Rodríguez-Aguilera, Stephanie Maldonado, Alexandra Stoll and Jaime Bravo.

13:15 - 14:45 Lunch - Gran Hotel Pucón Calafquén

15:00 – 16:00 Plenary Lecture IV – Araucanía Room

Dr. Gleen Hicks - CHEMICAL BIOLOGY REVEALS INSIGHTS INTO ENDOMEMBRANE TRAFFICKING. Center for Plant Cell Biology and Institute for Integrative Genome Biology, University of California, Riverside, USA.

Chair: Lorena Norambuena

16:00 – 17:00 Oral Session VII- Araucanía Room

Chairs: Lorena Norambuena and Claudia Stange

16:00 FUNCTIONAL CHARACTERIZATION OF TWO NUCLEOTIDE SUGAR TRANSPORTERS IN VITIS VINIFERA. Daniella Utz and Michael Handford.

16:20 bZIP A NOVEL TRANSCRIPTIONAL REGULATOR OF PROTEIN TRAFFICKING INVOLVED IN ENDOCYTOSIS REGULATION IDENTIFIED BY A SYSTEM BIOLOGY APPROACH IN ARABIDOPSIS THALIANA. Lorena Pizarro, Claudio Osorio, Alexander Vergara, Marcela Rojas-Pierce, Rodrigo Gutiérrez, Glenn R. Hicks and Lorena Norambuena.

16:40 THE NAC FAMILY TRANSCRIPTION FACTORS NTL9 AND NAC017 PARTICIPATE IN ENDOCYTIC TRAFFICKING REGULATION IN ARABIDOPSIS THALIANA. Claudia González-Ramírez, Lorena Pizarro and Lorena Norambuena.

17:00 Hacia la Institucionalización de la Biología Vegetal en Chile.

Dr. Ariel Orellana

Organizing Committee Meeting

18:00 - 18:30 Coffee Break

18:45 – 19:45 Closing Ceremony, Best Oral and Poster Presentation Awards – Araucanía Room

Chair: Boris Sagredo

20:00 - 21:30 Dinner – Gran Hotel Pucón Calafquén Restaurant

22:30 After Dinner

Thursday – December 5, 2013

8:00 - 11:00 Breakfast – Gran Hotel Pucón Calafquén Restaurant

Hotel Check-out before 12:00 PM- Gran Hotel Pucón Reception

Plenary Lectures

PL1

**THE STRUCTURE AND EVOLUTION OF COMPLEX GENOMES:
THE *FRAGARIA* (STRAWBERRY) OCTOPLOIDS AND DECAPLOIDS**

Thomas M. Davis¹, Bo Liu¹, Kim E. Hummer², Qian Zhang¹, David Wood¹, Lise Mahoney¹,
Yilong Yang¹, Laura DiMeglio¹, Melanie Shields¹, Magnus Lundberg¹

tom.davis@unh.edu

¹Department of Biological Sciences, University of New Hampshire,
Durham, New Hampshire, 03824, USA. ²USDA ARS National Clonal Germplasm Repository,
Corvallis, Oregon.

The genus *Fragaria* consists of about 23 species, ranging in ploidy from diploid ($2n=2x=14$) to decaploid ($2n=10x=70$). Surprisingly, a new 10x species was recently discovered in the Cascade Mountains of Oregon, giving encouragement to further collection and characterization of wild *Fragaria* germplasm within the Americas as well as in Asian centers of species diversity. At the University of New Hampshire, we are utilizing multiple approaches to study genome evolution and structure in *Fragaria*. The goals of our research program are to 1) better understand the genomic compositions and evolutionary histories of the *Fragaria* polyploid genomes; and 2) encourage and participate in the implementation of marker-assisted breeding (MAB) in the octoploid (8x), cultivated strawberry, *Fragaria x ananassa*. Our research approaches include: collection, characterization, and enhancement of wild germplasm; flow cytometric determination of C value; molecular cytogenetics; high throughput comparative genomic and amplicon sequencing; SNP genotyping, linkage mapping, and pedigree analysis. We have contributed to the development of important genomic resources, including the *Fragaria vesca* 'Hawaii 4' reference genome and, as members of the International RosBREED project, the newly released Axiom IStraw90 SNP array. Our data suggest that localized regions of ploidy reduction occur throughout the 8x genomes - a finding with important implications for basic research and MAB - and that the *Fragaria* 10x species are of recent hybrid origin.

Acknowledgments: This work was funded in part by USDA-CSREES NRI Plant Genome Grants 2008-35300-04411 and 2005-35300-15467, USDA-NIFA-SCRI grant number 2009-51181-05808, and by the New Hampshire Agricultural Experiment Station. T.M.D acknowledges current support from CONICYT Fellowship N° MEC-80122010.

PL2

**HORMONAL CROSSTALK IN ROOTS OF PLANTS SUBJECTED TO
WATER-STRESS CONDITIONS**

Carlos de Ollas, Vicent Arbona, **Aurelio Gómez-Cadenas**,

aurelio.gomez@uji.es

Departamento de Ciencias Agrarias y del Medio Natural. Universitat Jaume I, Campus Riu Sec,
E-12071 Castelló de la Plana, Spain

Phytohormones are central players in sensing and signaling numerous environmental conditions like drought. In this work, an experimental system based on severe plant dehydration was established and hormone profiling together with gene expression of key enzymes involved in abscisic acid (ABA) and jasmonic acid (JA) biosynthesis was studied in roots of citrus and Arabidopsis plants. JA concentration transiently increased after stress imposition whereas a more progressive ABA and JA-Ile accumulation was detected, at the same time or right after the JA burst. Molecular data suggested that, at least, part of the hormonal regulation takes place at the biosynthetic level. These observations also pointed to a possible involvement of JA on ABA biosynthesis under stress. To test this hypothesis two systems of study were established: in citrus, JA and ABA biosynthesis were chemically inhibited and subsequently phenotypes rescued by the addition of exogenous hormones; in Arabidopsis, mutants defective in biosynthesis of JA or insensitive to JA-dependent signaling were employed to characterize JA involvement in ABA accumulation under water-stress conditions. Results showed that the early JA accumulation leading to JA-Ile build up was necessary for the subsequent ABA increase in roots under stress whereas the opposite could not be stated. The model includes a burst of JA and a subsequent JA-Ile accumulation in roots of both citrus and Arabidopsis under severe water-stress conditions that lead to a more progressive ABA accumulation that will induce later downstream responses. However, ABA accumulation did not have any influence on JA levels. The present work adds a new level of interaction between JA and ABA at the biosynthetic level that together with the previously described interaction between their signal transduction cascades would allow plants to fine-tune specific responses to different stimuli.

PL3

Avances en el Proyecto CONICYT para el fomento de las Aplicaciones de Secuenciación Masiva (NGS) en Investigación y Desarrollo en Chile

Romilio Espejo

www.omics-solutions.cl

Centro Nacional de Genómica y Bioinformática.

El Centro Nacional de Genómica y Bioinformática (OMICS-Solutions) es el resultado de un proyecto CONICYT de Investigación asociativa, equipamiento mayor, cuyo objetivo es promover el uso de la genómica y bioinformática. En esta reunión revisaremos los logros del Centro en secuenciación masiva y bioinformática con ejemplos concretos de trabajos realizados. El Centro entrega servicios con siete plataformas de secuenciación masiva, capaces de entregar lecturas de la secuencia problema por un total desde 20 Mbp hasta 120.000 Mbp. Estas son 454Jr y FLX+ de ROCHE, Ion Torrent, Ion Proton, SOLID 5500 y SOLID 4 de LIFE Technologies, y MiSeq de Illumina. El Centro mantiene un equipo estable formado por tres ingenieros biotecnólogos y dos ingenieros bioinformáticos, que han recibido entrenamiento en cada uno de los métodos y plataformas utilizadas. A la fecha OMICS Solutions ha efectuado más de 50 servicios de secuenciación masiva, aplicando muy diferentes metodologías y plataformas. Se revisará brevemente la metodología, plataforma y los resultados obtenidos en los siguientes servicios: a) la identificación y diseño de marcadores (microsatélites) en diferentes vegetales, b) la secuenciación de un genoma humano, c) la expresión diferencial en peces. La identificación de marcadores se ha realizado utilizando secuenciación desde un extremo "single end" de fragmentos de DNA de aproximadamente 500 bp. Cuatro diferentes especies pudieron analizarse en una misma corrida del pirosecuenciador ROCHE Jr., generando suficientes lecturas para identificar al menos aproximadamente 1000 marcadores por especie, con diseño efectivo de partidores para amplificación por PCR y posterior validación. La secuenciación del genoma humano se realizó preparando una genoteca "mate paired ends" que luego fue secuenciada en la plataforma SOLID 5500 xl, capaz de rendir lecturas que sumen hasta 120 Giga bases totales. La comparación de los resultados obtenidos con un genoma de referencia permitió identificar 261.110 SNPs y 10.344 variantes estructurales, asociadas a grandes inserciones y deleciones, algunos relacionados con ciertos fenotipos, ancestría y patologías. La expresión diferencial en peces se realizó preparando una biblioteca "single end" desde mRNA obtenido de los peces en diferentes condiciones, que se secuenció en PGM ion Torrent y en Ion Proton. Los resultados permitieron observar la expresión diferencial de varios genes relacionado con la condición original.

Proyecto CONICYT ECM01

PL4

**THE FASCINATING WORLD OF BERRY VIRUSES – MIXED INFECTIONS
ARE THE NORM**

Robert R. Martin¹, Diego F. Quito-Avila², Karen E. Keller¹, Nola J. Mosier¹, Alfredo Diaz Lara;
Kara E. Sarver¹, Jake E. Dittrich¹, Ioannis E. Tzanetakis³

¹USDA-ARS Horticultural Crops Research Unit, Corvallis, Oregon, USA; ²Centro de
Investigaciones Biotecnológicas del Ecuador, Guayaquil-Ecuador; Department Plant
Pathology, University of Arkansas, Fayetteville, USA

With the application of molecular tools for characterization of viruses in berry crops, it has become clear that many diseases previously attributed to a virus are actually caused by virus complexes. As a group, berry crops including; *Fragaria*, *Rubus* and *Vaccinium*, are known hosts of at least 30 genera of plant viruses. These crops are propagated vegetatively and grown in nurseries and/or fruiting fields for multiple years and are often infected with virus complexes. In addition, a single 'disease' may be caused by multiple complexes with different viruses involved in various geographical regions. The complexity of many virus diseases in berry crops can be looked at as a positive or negative in terms of disease management. With a negative outlook one might ask, 'How could we possibly control all these viruses, and look at it as a hopeless situation'. On the positive side one might say, 'Since multiple viruses are necessary to get disease, which is the easiest of the viruses to control? Since virus complexes are common, we need to consider two types of control. For nurseries, virus control is important to prevent movement of viruses with propagation material. For fruit growers, disease control should be emphasized and controlling one or more viruses that are critical for disease development can often control the disease even though one or more viruses may be present in the field. Characterization of virus complexes, production of clean planting stocks and disease management will be discussed.

PL5

CHEMICAL BIOLOGY REVEALS INSIGHTS INTO ENDOMEMBRANE TRAFFICKING

Glenn R. Hicks, Chunhua Zhang, Michelle Brown, Michael C. Young, Wilhelmina Van de Ven, Richard Hooley, and Natasha V. Raikhel

ghicks@ucr.edu

Center for Plant Cell Biology and Institute for Integrative Genome Biology,
University of California, Riverside

Endomembrane trafficking is essential for coordinated growth and development in plants and response to environment. In particular, key plasma membrane proteins such as PIN auxin transporters and the BRI1 brassinosteroid receptor translocate between the endosomes and plasma membrane. The discovery of the important roles of the endomembrane system in hormone signaling in the last decade has contributed greatly to the understanding of signal transduction pathways and their integration during plant growth and development. Due to genetic redundancy and the highly dynamic properties of the endomembrane system, it is challenging to use traditional knock-out mutants alone to study trafficking. Chemical genomics provides a valuable tool for solving these problems by the rapid action, specificity and reversibility of small molecules. A very successful example is Brefeldin A which has been widely used in to study trafficking and signaling. Utilizing a previously published large-scale chemical library screening approach, we identified groups of small molecules that affect trafficking of PIN auxin transporters and other plasma membrane proteins in Arabidopsis. We are currently combining biochemical and proteomic approaches with genetic screening to identify the target of several bioactive compounds. One of these compounds (ES2) appears to target the exocyst complex in Arabidopsis which is essential for exocystosis, cell division and development. This and other results in dissecting endomembrane trafficking will be discussed.

PL6

HACIA LA INSTITUCIONALIZACIÓN DE LA BIOLOGÍA VEGETAL EN CHILE

Red de Biología Vegetal

Han pasado 8 años desde que nos comenzamos a reunir como comunidad de biólogos vegetales. El objetivo de esta iniciativa era juntarnos para presentar y discutir los trabajos que los distintos grupos desarrollan en Chile. De esta manera dimos forma a la “Red de Biología Vegetal”, la que ha resultado instrumental para conocernos, establecer vínculos, promover colaboraciones de trabajo y aumentar nuestra masa crítica. Hemos crecido y logramos ser reconocidos a nivel nacional e internacional. Nuestra actividad crece y se nos abren nuevos desafíos. Hace un tiempo fuimos convocados a integrarnos a The Global Plant Council (www.globalplantcouncil.org), organización que convoca a 25 importantes sociedades científicas del mundo en el área de la biología vegetal. Esta entidad busca coordinar estrategias y proyectos que tengan impacto en problemas de orden global, así como transformarse en una voz que transmita propuesta frente a instituciones como la ONU o la FAO. Situaciones como esta nos llevan a considerar darle una nueva estructura a nuestra Red, de manera de enfrentar estos desafíos de una manera mas coordinada, formal y que responda al conjunto de las inquietudes de quienes forman parte de la comunidad de biólogos vegetales.

Suscriben:

Ariel Orellana, Rodrigo Gutierrez, Boris Sagredo, Patricio Arce, Liliana Cardemil, Raúl Herrera, Xavier Jordana, Lorena Norambuena, Francisca Blanco, Wendy González, Lida Fuentes, Cristian Ibáñez, Mauricio González A., Erwin Krauskopf.

Oral Sessions

OS1

**QTL ANALYSIS FOR FRUIT QUALITY TRAITS IN PEACH
(*PRUNUS PERSICA* (L.) BATSCH)**

¹Gerardo Núñez, ^{1,2}Alejandra Cifuentes, ¹Reinaldo Campos-Vargas, ²Gamaliel Lemus,
³Rodrigo Infante, ¹Ariel Orellana, ¹Claudio Meneses

gera.nunez.lillo@gmail.com

¹Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello. República 217, Santiago. ² Centro de Frutales de Carozo. Instituto de Investigaciones Agropecuarias (INIA-Rayentué), Rengo. ³Universidad de Chile, Facultad de Ciencias Agronómicas, Santiago.

Chile is the first peach exporter in the South Hemisphere and the fifth producer in the world. Chilean initiatives of peach breeding have been carried out, where the fruit quality and postharvest performance are the main goals for the new peach varieties. Breeding is a time consuming and costly process. For this reason, the development of genomic tools to support the early selection of genotypes is quite relevant to improve the efficiency of breeding programs. Thus, the aim of this work was to identify genome regions associated with fruit quality traits in peach using a QTL analysis. The segregating population of 151 individuals obtained from Venus x Venus was evaluated in four periods: Harvest, harvest + shelf life, 21 days at 0 °C and 21 days at 0 °C + shelf life. Firmness, weight, color, soluble solids, titratable acidity, ripening time, browning and mealiness were evaluated. The segregating population was genotyped with microsatellite markers (SSRs) and SNPs using 9K SNP array for peach. A linkage map was built with 1820 markers, which were mapped in 8 linkage groups. The resulting map spanned a total distance of 382.9 cM with an average of 0,21 cM between adjacent markers. A QTL for titratable acidity was associated to chromosome 1 (5,2 Mbp with 42% variance explained) and in chromosome 4, QTLs for weight (2 Mbp with 15% variance explained), soluble solids contents (1,9 Mbp with 32% variance explained) and two for mealiness (6,53 Mbp and 0,5 Mbp with 35% and 41% variance explained respectively) were detected. Currently, additional work has being done in order to get new molecular markers related to fruit quality traits.

Acknowledgements: Fondecyt 11121396, Fondecyt 1110954, FONDAP CRG 15070009; Núcleo Milenio P10-062-F and Basal PFB-16.

OS2

IDENTIFICATION AND CHARACTERIZATION OF PUTATIVE MARKERS DERIVED FROM A TRANSCRIPTOMIC APPROACH (RNA-SEQ) ASSOCIATED TO BERRY SIZE IN TABLE GRAPES

Claudia Muñoz^{1,4}, Alicia Sanchez³, Alex Di Génova^{2,4}, Alejandro Maass^{2,4}, Mauricio González-Aguero³, Ariel Orellana^{1,4}, Patricio Hinrichsen³

phinrichsen@inia.cl

¹Núcleo Milenio en Genómica Funcional de Plantas, Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello, Santiago. ²Centro de Modelamiento Matemático, Universidad de Chile, Santiago. ³INIA La Platina, Santiago.

⁴FONDAP Center for Genome Regulation, Santiago.

Development and maturation of grape berries has been intensely studied, nevertheless its molecular regulation is still poorly understood. Our aim is to identify genetic factors associated with berry size, which can be used as markers for the selection of new genotypes from a table grape breeding program. We have analyzed data from a massive transcriptomic experiment using RNA-Seq Illumina sequencing technology. Forty-seven samples corresponding to early phenological stages (anthesis, post-setting and 6-8 mm berry diameter) of 12 segregants from a 'Ruby' x 'Sultanina' crossing plus both parents were sequenced. The segregants represented contrasting phenotypes for berry size and seeds content. A total of 477 million reads of 47 bp average length were aligned onto the 12X PN40024 reference genome. In order to identify differentially expressed (DE) genes at each phenological stage, total reads from genotypes contrasting for berry size were compared. Also, single nucleotide polymorphisms (SNPs) and insertions/deletions (INDELs) were detected using GATK structural variant calling. With this approach, 190 DE genes were detected ($p\text{-value} < 1 \times 10^{-5}$), from which eight candidates genes were selected and validated by Real Time PCR. After the global identification of SNPs and INDELs, biallelic polymorphisms were selected and filtered by thin (100bp) and MAF (20%) using VCF pipeline. Then, 177 non-synonymous and intragenic SNPs and 15 INDELs were manually analyzed, considering their coverage. Twenty-one SNPs and five INDELs have been confirmed by sequencing and are currently being validated by qPCR-HRM in the RxS population and in a nuclear collection of *Vitis vinifera* L, in order to evaluate their transferability.

Acknowledgements: Genoma-Chile, FONDEF G07I-1002 y G09I-1007, Basal-CMM, Fondecyt 1110954, FONDAP CRG 15070009, Núcleo Milenio P10-062-F, Basal PFB-16 and Programa Mece-sup.

OS3

STUDY OF RIPENING AND CHILLING INJURY IN EARLY AND LATE VARIETIES OF PEACHES AND NECTARINES (*PRUNUS PERSICA*) USING RNA-SEQ

Dayan Sanhueza, Claudio Meneses, Paula Vizoso, Christian Silva, Iván Balic,
Ariel Orellana, Reinaldo Campos-Vargas.

dayansanhueza@gmail.com

Universidad Andrés Bello, Facultad Cs. Biológicas, Centro de Biotecnología Vegetal.

Peaches or nectarines are affected by chilling injury (CI) problems, as consequence of long term cold storage. In *P. persica*, CI includes abnormal cell wall disassembly with development of a dry and mealy mesocarp. However, this occurs mainly in cultivars that are harvested in late season but not in early season ones. Hereby, our main objective was to find differential expression of genes that could be related with processes that happen during mealiness development in late varieties compare to sound early ones. We analyzed the transcriptomes of a early nectarine (VET2, juicy), a late nectarine (VET3, mealy) and a late peach (QCC8, mealy) in four conditions, mature, ripe, 21 days at 4 °C and 21 days 4 °C plus shelf life. Twenty four (3 varieties x 4 conditions x 2 replicated) RNA libraries were constructed and sequenced using HiSeq 2000. We obtained between 19-34 millions reads in the different libraries with 101 bp average length. The reads were trimmed and mapped against peach genome reference. To carried out the data analyses the different conditions were grouped in pairs (VET2 vs. VET3, VET2 vs. QCC8 and VET3 vs. QCC8) and the transcripts were filtered by p-value < 0.01. Using hierarchical clustering we selected the genes with highest increase or decrease expression levels and categorized using GO database. As mealiness has been related with cell wall, we took special consideration of sequences that codify for enzymes that participate in cell wall metabolism. In order to accomplish this we filtered those genes that are common between VET2 vs VET3 and VET2 vs QCC8 in 21 days at 4 °C and shelf life. For those samples that are firm, among the sequences with highest increase we found XET and PME1. Otherwise, for mealy fruits we observed an important increase in expansin and ACC oxidase, meanwhile considerable repression was detected for UDP-glycosyltransferase. Currently we are doing other analyses in order to get more information about genes and CI development.

Acknowledgements: DS and IB CONICYT-Doctoral Fellowship; UNAB DI-64-12/1; DS CONICYT Doctoral thesis; PFB-16; FONDAP-CRG 15090007.

OS4

LONG-DISTANCE GENE SILENCING BY ARTIFICIAL MIRNAS

Daniela Quiroz^{1,2}, Catalina Alvarez², Álvaro Castro^{2,3}, Humberto Prieto²

danielaquirozlarraín@gmail.com

¹Universidad Andrés Bello. ²Instituto de Investigaciones Agropecuarias.

³Universidad de Santiago de Chile.

MicroRNAs (miRNAs) study has been subject of interest since its discovery in *C. elegans* in 1993. In plants, microRNAs play important roles in developmental processes, metabolic pathways and stress responses. miRNAs are 21 or 22 nt molecules produced by non-coding transcripts synthesized by RNA polymerase II. An important aspect in small RNAs (sRNA) research is to understand their movement in plants, including cell-to-cell and long-distance movement. Although many studies provide evidence of miRNA's traffic, there's no agreement about the direction of the movement, or which are the mobile signals and the mechanisms that allow the transportation. We designed two artificial pre-miRNA (MIR-A-GFP and MIR-B-GFP) based on the modification of *V. vinifera* endogenous pre-miRNA vvi-MIR319e for targeting the gene that encodes the green fluorescent protein (GFP). The artificial miRNA activity to silence GFP gene was verified in *V. vinifera* and *N. benthamiana* through transient transformation by agroinfiltration. We generated transgenic lines of *N. benthamiana* expressing the artificial pre-miRNAs constitutively. In parallel, a double transgenic line was made by re-transforming self-pollinated F₂ constitutive GFP lines with MIR-B-GFP. With these transgenic lines, 4 sRNAs libraries were generated. Illumina sRNAs sequences confirmed the existence of 21 and 22 nt miRNAs and also 21-24 nt secondary siRNA derived from GFP messenger processing. To provide new evidence on the traffic of sRNAs, we designed a grafting assay to evaluate long-distance movement of the 21-24 nt molecules. Transgenic lines expressing MIR-A-GFP and MIR-B-GFP were grafted with plants that expressed GFP. Both lines were used as scion or rootstock to test directionality of the sRNA movement, also to elucidate which specific species generated are able to move long distance. Results showed a bidirectional movement of the signals, being faster the silencing of GFP when the sRNAs are produced in the shoot.

Acknowledgements: FONDEF G09i1007 and BIOFRUTALES.

OS5

**PHYSIOLOGICAL AND PRODUCTIVE RESPONSES MEDIATED BY NITROGEN AVAILABILITY
IN *COLOBANTHUS QUITENSIS* AND *DESCHAMPSIA ANTARCTICA* EVALUATED
IN GLASSHOUSE**

Claudia Rabert^{1,2}, Mario Díaz¹, Claudia Mella¹, Carla Alvear^{1,3}, Miren Alberdi^{2,3},
Marjorie Reyes^{2,3}, León Bravo^{1,2}

claudia.rabert@ufrontera.cl

¹Laboratorio de Fisiología y Biología Molecular Vegetal, Facultad de Ciencias Agropecuarias y Forestales, UFRO, Temuco. ²Center of Plant, Soil Interaction and Natural Resources Biotechnology, Scientific and Technological Bioresource Nucleus, UFRO, Temuco.

³Departamento de Ciencias Químicas y Recursos Naturales, UFRO, Temuco.

The Antarctic Peninsula is characterized by limited growing conditions for establishment of plants, such as: strong winds, low temperatures, short growing season, available land, and water and nutrients availability. Only two vascular plants have been able to grow under these conditions; *D. antarctica* (*Poaceae*) and *C. quitensis* (*Caryophyllaceae*). Several reports consider that the populations' increase of these species during the last decades is caused mainly by climate warming. Low temperatures limit biological nutrient cycling in Antarctic and hence nutrient availability is scarce and usually associated to birds and/or seals colonies. The average air temperature has increased 2.6°C over the last 50 years which may explain a higher nutrient cycling and higher available resources (macro and micronutrients); in field experiments using OTC showed 2 to 3 fold increase in nitrate and ammonium soil availability in warmed sites. It is not established how these two antarctic species respond to increased fertilization. This investigation relates nutrients availability with plant growth and photosynthetic yield (Fv/Fm) responses. Two sources of nitrogen (ammonium or nitrate) and five doses (0, 4, 8, 16 and 32 mM) in both species were assayed. Both species responded better to ammonium source, increasing aerial biomass at lower concentration than nitrate source. The optimal ammonium concentration is around 8 mM for both species while no distinctive optimum was observed with nitrate source, instead a sort of saturation behavior. Fv/Fm decrease only under high concentration of ammonium (32 mM) this is associated with toxicity signals observed in plants. Therefore, a putative explanation for population expansion for these species in Antarctic, is the increased availability of nutrients generated by current environmental conditions imposed by global warming.

Acknowledgments: project ART1102.

OS6

STUDIES OF THE EXPRESSION OF ADAPTIVE DIFFERENTIAL MECHANISMS IN TWO GENOTYPES OF *SOLANUM PERUVIANUM* L. DURING WATER STRESS

Oscar Arrey^{1,3}, Yazmina Stappung², Gerardo Tapia³

oarrey@udec.cl

¹Unidad de Recursos Genéticos, Instituto de Investigaciones Agropecuarias, INIA Quilamapu, Chillán. ²Centro de Bioinformática y Simulación Molecular, Universidad de Talca, Talca.

³Departamento de Ciencias y Tecnología Vegetal, Escuela de Ciencias y Tecnologías, Campus Los Ángeles, Universidad de Concepción, Los Ángeles.

Solanum peruvianum L. is a wild tomato species inhabiting the desert and semi-desert areas of northern Chile and southern Peru from the sea level to approximately 1500 m above the sea level. This specie has a high degree of allogamy and genetic variability within and among populations. As part of this research we have selected two genotypes of *S. peruvianum* from two populations, one located near the coast (Lp108) and the other at 1100 m above the sea level. (Lp134) and are subject to different environmental conditions in terms of temperature and humidity. In order to study the characteristics of tolerance to water stress associated with these different environments, both genotypes were evaluated for traits related to water stress tolerance including anatomical, physiological and biochemical aspects. In addition, a transcriptome analysis has been made using a whole genome chip microarray. The results were grouped by hierarchical cluster complemented by gene expression studies performed by qRT-PCR. The assay consisted of two water treatments, one full and one deficit. The Lp134 genotype showed a greater development expressed in plant growth compared to Lp108, both in the full and deficit treatments developed under controlled conditions. Lp134 presented a greater increase in the osmotic potential than Lp108, which is possibly associated with an increased accumulation of compatible osmolytes. Genotypes also showed differences associated with stomatal conductance and stomatal density and the activity of antioxidant enzymes. These results suggest that *S. peruvianum* has different mechanisms of abiotic stress tolerance, which can be directly associated with specific micro-habitats formed by climatic and edaphic conditions.

Acknowledgments: Proyect FONTAGRO FTG-8071/08.

OS7

**COLOBANTHUS QUITENSIS, A MODEL SPECIE OF GENOMIC RESPONSE
TO EXTREME ENVIRONMENT**

Marely Cuba-Díaz¹

mcuba@udec.cl

¹Laboratorio de Biotecnología y Estudios Ambientales. Depto. de Ciencias y Tecnología Vegetal, Campus Los Ángeles, Universidad de Concepción.

Terrestrial Antarctic ecosystem has scientific value based on the combination of a set of environmental stressors, extreme low temperatures, desiccating winds, freezing, osmotic stress, low nutrients availability and elevated UV radiation. In addition, the global warming generates a more complex situation in Antarctic, e.g. the air temperature of the Peninsula has increased by 2°C over the last 40–50 years, and the retreat of glaciers and permanent ice will offer opportunities for colonization of pristine habitats in newly exposed areas. Only two vascular plants naturally inhabit in Antarctic, *Deschampsia antarctica* Desv. (Poaceae) and *Colobanthus quitensis* (Kunth) Bartl. (Caryophyllaceae) both have become in recent times in models for understanding biochemistry and molecular mechanism that enable them to survive in these harsh conditions. Besides this, *C. quitensis* has a wide geographical distribution, from Mexico to the north of the Antarctic Peninsula, which also involved stressful environmental habitats. At the southern end of its colonization range occurs at sea level, but towards the north it is growing at high altitudes reaching up to 4200 m.a.s.l., showing a considerable morphologic variability along its wide distribution range, providing an interesting model for ecotypic differentiation and for intra-specie genetic diversity. Physiological, molecular, morphological, genetic properties as well as some aspects of the molecular ecology of *C. quitensis* will be shown as a model of genomic responses to environmental extremes elsewhere on the planet.

Acknowledgements. DIUC-208.112.044-1.0 e INACH T_03-09

OS8

ASSESSMENT BY MICROSATELLITES OF GENETIC DIVERSITY OF CHILEAN POPULATIONS OF ALGARROBO (*PROSOPIS CHILENSIS*) IN RESPONSE TO SALINE STRESS

Claus Westphal¹, Cristian Ibáñez¹, Antonio Gonzalez²

cwestphal@alumnosul.cl

¹Departamento de Biología. Facultad de Ciencias. Universidad de La Serena. La Serena.

²Centro de Investigación en Ecosistemas (CIEco). Facultad de Ciencias. Universidad Nacional Autónoma de México (UNAM). Morelia. México.

Algarrobo (*Prosopis chilensis*) is a tree capable of tolerating several biotic and abiotic stresses. Previously, we had evaluated the salinity tolerance of several trees of *P. chilensis* distributed along Coquimbo, Valparaíso and Metropolitan Regions. What we found were trees, mainly located in the Choapa Valley (Coquimbo Region), which in response to several astringent and very astringent concentrations of NaCl, consistently showed the best performance at germination and physiological levels. In contrast, the poorest response was obtained with trees located in the most North and South limit of its distribution (Elqui Province and Chacabuco Province, respectively). Intrigued by this different geographic response to saline stress, we studied three universal chloroplasts of plants and six nuclear markers specifically designed in *P. chilensis*. Analysis of chloroplast markers showed that genetic differences at intra and inter population level was observed, and trees coming from north, central and south of distribution were effectively different at chloroplast level. Moreover, nuclear microsatellites confirmed that genetic diversity was also occurring among populations and also confirmed a genetic structure throughout the natural range of this tree. Therefore, molecular marker analysis correlates well with those obtained previously at germination and physiological level, corroborating that higher saline tolerance observed in the trees of Choapa Valley were owing to a genetic diversity. These results generate important information in order to strengthen future works related to breeding programs and conservation initiatives over marginal areas where salinity and other stresses like drought are or might be a constraint.

Acknowledgements: Fondecyt 1110831; CONICYT for a Doctoral fellowship (grant n° 21120460) and MECESUP FIAC-2, Universidad Católica del Norte.

OS9

**GENE EXPRESSION CHANGES IN NEEDLES AND STEMS OF *PINUS RADIATA*
ROOTSTOCK PLANTS OF TWO AGES WITH DIFFERENT ROOTING CAPABILITY**

Carolina Álvarez¹, Luis Valledor², Patricia Sáez¹, Manuel Sánchez-Olate¹ Darcy Ríos¹

caalvarez@udec.cl

¹Laboratorio de Cultivos de Tejidos Vegetales, Facultad de Ciencias Forestales, Centro de Biotecnología y Departamento de Silvicultura, Facultad de Ciencias Forestales. Universidad de Concepción. ²Laboratory of Adaption Biotechnologies. Global Change Research Centre, Academy of Sciences of the Czech Republic, Brno, Czech Republic.

A major problem in forest clonal productivity is the loss of rooting capability with the increasing age of rootstock plants. However, despite of the importance of loss of morphogenetic competence, very little research has been done about the underlying mechanisms involved in this process. For this reason, a gene expression analysis using dot blot technique was performed in needles and stems of 1- and 3-year old *Pinus radiata* rootstock plants with a proved decrease in rooting capability. Genes selected for the expression analysis were previously associated to the process of decrease in morphogenetic competence in *P. radiata* tissues. Also, quality control of dot blot experiments was achieved through quantitative RT-PCR (qRT-PCR). Juvenile 1-year-old rootstock plants showed a higher number of up-regulated genes mainly corresponding to photosynthesis, protein synthesis, degradation and modification and genes related to activation of transcription and translation. On the contrary, aged 3-year-old rootstock plants were characterized by a low amount of genes accumulated on needles and stems. Specifically in stems of aged 3-year-old rootstock plants, an increase in the expression of an ethylene response factor (ERF) gene was found. This could indicate that genes involved in organ senescence and ethylene pathways may regulate the loss of morphogenetic competence. Finally, the analyses clearly showed that the selected genes were capable of differentiate between *P. radiata* rootstock plants with high (1-year-old) and low (3-year-old) morphogenetic capability in needles and stems, which is also correlated with age related changes in the anatomy of both types of tissues.

Acknowledgments: DIUC 211.142.031-1.0 research projects, MECESUP scholarship and Proplantas S.A nursery.

OS10

**CAN FCSEPLIKE REGULATES THE EXPRESSION OF CELL WALL DISSEMBLING GENES
DURING RIPENING OF *FRAGARIA CHILOENSIS* FRUIT?**

Luis Morales-Quintana^{1,3}, Daniela Urbina¹, Carlos Figueroa², Raúl Herrera¹,
María Alejandra Moya-León¹

lumorales@alumnos.utalca.cl

¹Laboratorio de Fisiología Vegetal y Genética Molecular, Instituto de Biología Vegetal y Biotecnología, Universidad de Talca. ²Facultad de Ciencias Forestales, Universidad de Concepción. ³Programa de Doctorado en Ciencias Mención Ingeniería Genética Vegetal, Universidad de Talca.

MADS-box transcription factors (TF) have been described as master switches during floral and fruit development in angiosperms. In particular, SEPALLATA like subfamily seems to play a key role in fruit development and ripening. Chilean strawberry (*Fragaria chiloensis*) is a non-climacteric fruit that soften very fast during ripening. Previously, we identified a MADS-box TF that is induced during ripening of Chilean strawberry fruit. This TF was classified as SEPALLATA and named as FcSEPlike. FcSEPlike was cloned and its ectopic expression in tobacco epidermal cells revealed a nuclear subcellular distribution. On the other hand, three genes involved in cell wall dissembling are induced during *F. chiloensis* softening: *polygalacturonase1* (*FcPG1*), *pectate lyase 1* (*FcPL1*) and *expansin 2* (*FcExpA2*). Their promoters were isolated and bioinformatic analysis indicated the presence of MADS-box responsive elements (CARG-box) on each one. As a way to explain if FcSEPlike TF plays a role in the regulation of *FcPG1*, *FcPL1* and *FcExpA2*, bioinformatic analyses were performed. Through molecular modeling the structural model of FcSEPlike was generated, displaying a structure consisting in 5 β -sheets and 2 α -helices. By molecular docking and molecular dynamics simulation the interaction of FcSEPlike TF with a canonical set of cis-regulatory elements of MADS-type and specific cis-regulatory elements found in the promoter regions of the genes under study was explored. The results showed that all interactions are favourable, although cis-regulatory elements from *FcPG1* and *FcExpA2* promoters have better interaction with FcSEPlike. The data suggests that FcSEPlike could regulate the expression of *FcPG1* and *FcExpA2* during ripening of strawberry fruit. This in silico prediction is waiting for experimental validation.

Acknowledgements: CONICYT for Doctoral fellowship and D.U. thanks to PBCT PSD-61. Research supported by Anillo ACT-1110 and Fondecyt 1110792.

OS11

**STRUCTURAL CHARACTERIZATION OF TWO MADS TRANSCRIPTION FACTORS
THAT BIND TO THE PROMOTER OF PRXTH, A GENE INVOLVED IN THE
INCLINATION RESPONSE OF *PINUS RADIATA* D. DON**

Verónica Latapiat¹, Luis Morales-Quintana^{1,2}, María Alejandra Moya-León, Raúl Herrera¹

vlatapiatg@alumnos.utalca.cl

¹Laboratorio de Fisiología Vegetal y Genética Molecular, Instituto de Biología Vegetal
y Biotecnología, Universidad de Talca. ²Programa de Doctorado en Ciencias
Mención Ingeniería Genética Vegetal, Universidad de Talca.

Pinus radiata D. Don is the most important forest species for the Chilean economy. The growth of trees can be affected by abiotic stresses, causing the loss of their verticality. Inclined trunks reorient its vertical growth leading to compression wood formation. This type of wood is characterized by high levels of lignin, less cellulose content and greater micro-fiber angle, and therefore provides a low wood quality. A xyloglucan endotransglucosylase/hydrolase gene (*PrXTH*), involved in wood formation, has shown to be induced in response to inclination in the lower side of inclined radiata pine stems. On the other hand, the activation of two MADS transcription factors (TF) (PrMADBT and PrMADBJ) at early stages of tilting response has been demonstrated. The analysis of the promoter region of *PrXTH* indicated that it contains sequences that could be recognized by MADS TFs. Using bioinformatics tools the interaction of MADS TFs and *PrXTH* promoter was explored. The structure of PrMADBT and PrMADBJ was obtained by molecular modeling. Molecular docking and molecular dynamics simulation were used to determine if these MADS TFs could bind to cis-regulatory elements. Several DNA sequences from the promoter and canonical binding sequences were tested, and among them, CARG4 a sequence from the promoter showed the best interaction with homo-dimers of PrMADS. The residues K23, R26, K30 and K31 located within the MADS domain of each TF explains the binding specificity to the CarG box sequence from the promoter of *PrXTH*.

Acknowledgements: CONICYT for a Doctoral fellowship. Research supported by Anillo ACT-1110 and Fondecyt 1120635 project.

OS12

**EFFECT OF MODIFIED ATMOSPHERE PACKAGING (MAP) ON RACHIS QUALITY
OF 'RED GLOBE' AND 'FLAME SEEDLESS' VARIETIES**

Christian Silva-Sanzana¹, Iván Balic¹, Adrián Moreno¹, Dayan Sanhueza¹,
Patricio Olmedo¹, Bruno Defilippi², Reinaldo Campos-Vargas¹

c.silva.sanzana@gmail.com

¹Universidad Andrés Bello, Facultad Ciencias Biológicas, Centro de Biotecnología Vegetal, República 217, Santiago. ²Instituto de Investigaciones Agropecuarias, INIA La Platina, Santiago.

Rachis browning corresponds to a postharvest disorder that drastically reduces table grape quality. This problem has been mainly associated to water loss, but is feasible the possibility to involved other factors. Modified atmosphere packaging (MAP) is a technology used to improve table grape postharvest quality. The objective of this study was to analyze the MAP effect on table grape rachis browning during cold storage at 0 °C in 'Red Globe' and 'Flame Seedless' varieties. MAP stored bunches of both varieties showed higher green color percentage than conventional storage, this situation was concomitant with a lower chlorophyll-*a* content and higher amount of pheophytin-*a*. To understand these results, we carried out a transcriptional analysis of chlorophyll breakdown pathway genes. We observed that compared to harvest, conventional packaging at 0°C induced a decrease in transcript amount of all analyzed genes in both varieties. However, MAP storage caused different responses between varieties. 'Red Globe' showed a significant transcript reduction of Metal Chelating Substance (MCS), Pheophytinase (PPH) and Red Chlorophyll Catabolite Reductase (RCCR) compared with conventional storage, except pheophorbide-*a* Oxygenase (PaO) that showed no differences between both storage strategies. On the other hand, in 'Flame Seedless' was found an increase in transcript abundance of MCS, PaO and RCCR, while PPH showed no changes respect to conventional storage. Moreover, histological assay revealed that browned rachises showed more suberized and lignified cells layers than green rachises. Our results suggest that MAP represent a suitable storage strategy to reduce rachis browning without affecting berries quality, and that rachis browning correspond to a multifactorial process that can be explained by a green color loss related to chlorophyll breakdown and a brown color gain due to periderm formation.

Acknowledgements: Fondecyt 1085025.

OS13

**ROLES OF ANTIOXIDANT SYSTEMS ON APPLE SKIN BROWNING DISORDERS
POSTHARVEST ASSOCIATED TO HIGH SOLAR IRRADIATION**

Carolina Torres¹, Omar Hernández², Iván Razmilic³, Alejandra Moya⁴

cartorres@utalca.cl

¹Facultad de Ciencias Agrarias, Universidad de Talca. ²Centro de Pomáceas, Universidad de Talca. ³Instituto de Química y de Recursos Naturales, Universidad de Talca.

⁴Instituto de Biología Vegetal y Biotecnología, Universidad de Talca.

Postharvest peel-browning disorders (sunscald and staining) on apples derived from high solar irradiation cause important economic losses. Symptoms develop only on sun-exposed sections of the fruit with or without visible sun-injury. In order to determine the role of different antioxidant systems in 'sunscald' (cv. Granny Smith) and 'stain' (cv. Fuji) development, fruit with different sun exposures and sun-injury levels on the tree were harvested and stored at 0°C for up to 4 months. Antioxidant metabolites (ascorbic acid, AsA, glutathione, GSH), AsA-GSH recycling enzymes activities and transcripts levels, and quercetin concentrations were monitored monthly on fruit peel. Unexposed fruit did not develop sunscald or stain. In general, in both cvs. total ascorbic acid (AsA) was the highest on fruit with no sunburn symptoms, with over 90% of it in its oxidized form. AsA-GSH recycling enzyme activities or transcription levels did not differ between sun-exposures during cold storage. In both cvs., all quercetin glycosides were higher in fruit with visible sun-injury compared to those with no symptoms (sun-exposed or shaded) throughout the storage period. In severe sunburned fruit quercetin glycosides represented around 70% of total phenolics compared to 10% in shaded fruit tissue. These results indicate that in both cvs. the AsA-GSH cycle does not play a direct role in 'sunscald' or 'stain' development. Instead, quercetin glycosides appear to be directly related with sunscald expression postharvest, i.e. the higher their concentration in the tissue, the faster the accumulation of their brown derivatives products.

Acknowledgements: This work was supported by Fondecyt Regular 1100013.

OS14

FLAVONOLS: MOLECULES THAT PLAY AN IMPORTANT ROLE IN THE GROWTH OF PINE

Patricio Ramos¹, María Alejandra Moya-León¹, Raúl Herrera¹

pramos@utalca.cl

¹Laboratorio de Fisiología Vegetal y Genética Molecular, Instituto de Biología Vegetal y Biotecnología, Universidad De Talca.

Plants are continuously exposed to gravity force that eventually could affect their vertical growth. In trees, the inclination response is a widely studied biological phenomenon; however, the molecular mechanism is still unknown. Based on a Suppressive Subtractive Hybridization (SSH) transcriptomic approach, the bioinformatic analysis revealed that many categories of genes showed differential expression. One of the most interesting groups of genes was the related to the phenylpropanoid compounds. The biosynthetic genes involved in this pathway were analyzed by qPCR and showed a significant induction after inclination, in a temporal and spatial differential manner along the stem. In addition to the analysis of expression profile, an auxin response gene, which is repressed in presence of the hormone, therefore was used as an auxin distribution sensor, showed a differential expression pattern along and across the stem. Additionally, flavonols (quercetin and kaempferol) showed an accumulation according to the expression profile observed for their biosynthetic genes. The results agreed with molecular function purposed to flavonols of blocking auxin transport and, suggest that an auxin distribution could be involved in the inclination response in pine. These observations are according to the Cholodny-Went hypothesis, indicating a concerted activation of genes that generates a misbalance in local auxin distribution that finally induces stem reorientation. The results obtained leading to a greater understanding of the molecular mechanism that governs the biological process.

Acknowledgements: This project was supported by FONDECYT projects 11121170, 1120635 and PBCT anillo ACT-41.

OS15

EFFECT OF APPLICATION OF PECTIN DERIVED OLIGOSACCHARIDES ON THE ACCUMULATION OF ANTHOCYANIN IN BERRIES OF CABERNET SAUVIGNON

Daniel Villegas¹, Michael Handford², Alonso Perez-Donoso¹

dlvilleg@uc.cl

¹Laboratorio de Fisiología Frutal, Facultad Agronomía, Universidad Católica de Chile.

²Laboratorio de Biología Molecular Vegetal, Facultad de Ciencias, Universidad de Chile.

During the 2012-2013 season, full clusters of Cabernet Sauvignon grapes were treated before veraison with a 1.5 mg/ml solution of pectin derived oligosaccharides (PDOs) obtained through partial enzymatic hydrolysis of poly-galacturonic acid. After the application date, berry samples were taken at periodic intervals in order to determine growth and maturity parameters, total anthocyanin content as well as the anthocyanin profile and relative expression of key genes in the phenylpropanoid pathway. The result show that PDO application significantly increase total anthocyanin content at 16 days after treatment (d.a.t.) compared to untreated samples, without generating significant effect in growth and maturity parameters measured in this assay. Beside the effect on total anthocyanin content, PDO treatment also modified anthocyanin profile (percentage distribution of the different anthocyanin present in grapes berries). In this way, PDO treated berries possess significant increases in malvidin percentage as well as significant changes in tri-hydroxylated versus di-hydroxylated forms and methylated versus non-methylated forms. Relative expression of *PAL*, *UFGT*, *F-3'-H* and *F-3'5'-H* genes was determined by quantitative RT-PCR. Of the studied genes, only *UFGT* present significant increase in relative expression when compared to untreated samples. This increase was evident at 9 d.a.t., which might be related to the increase in total anthocyanin content observed at 16 d.a.t. However, this increased expression was transient and any difference disappeared in subsequent samplings. In conclusion, the application of PDOs leads to increased anthocyanin content and change in the anthocyanin profile, accompanied by specific changes in the expression of key genes of the phenylpropanoid pathway.

Acknowledgments: This work was funded by FONDECYT 3120064.

OS16

***PRUNUS* ROOTSTOCK BREEDING PROGRAM FOR THE NEW CHILEAN
AGRICULTURAL REQUIREMENTS**

Felipe Gainza-Cortés¹, Verónica Guajardo¹, Ismael Opazo¹, Mauricio Ortiz¹, Rubén Almada¹,
Paula Pimentel¹, Boris Sagredo¹, Manuel Pinto¹, José San Martín^{1,2}, Gamalier Lemus^{1,3},
Jorge Pinochet¹, Carlos Muñoz⁴

fgainza@ceaf.cl

¹Centro de Estudios Avanzados en Fruticultura, Rengo. ²INIA, CRI Quilamapu, Chillán.

³INIA, CRI Rayentue, Rengo. ⁴Universidad de Chile, Facultad de Ciencias Agronómicas,
La Pintana, Santiago.

The current global agricultural challenge implies the need to generate new technologies and farming systems. In this sense, the creation of new rootstocks offers an alternative to stone fruit crop, particularly in Chile where there are site-specific problems. The implementation of Molecular Assisted Breeding (MAB) in order to give support to the phenotypic evaluation of plant breeding has great potential assisting the selection of new genotypes of rootstocks. Marker-Assisted Selection (MAS) can shorten the time required to obtain new cultivars and can make the process more cost-effective than selection based exclusively on phenotype, selecting genotypes at the seedling stage. In this way, the Center for Advanced Studies in Fruits (CEAF) have been implemented a long-term rootstock breeding program supported by three complementary programs: Physiology of the stress focusing on the study of critical physiological parameters associated to tolerance to abiotic stresses (hypoxia, drought and salinity); Genomics, focusing on the study of the molecular basis of *Prunus* spp. rootstocks under root hypoxia stress; and Agronomy focusing on optimizing water and soil management, and the interaction of grafted rootstock in *Prunus* spp. under limiting conditions of soil and water. With the development of early screening tools, the plant material generated in this program and those from co-obtention agreements with other private programs (AGROMILLORA and CSIC, Spain) are being subjected to rigorous selection for higher agronomic performance, as well as for traits of resistance and/or tolerance to abiotic stresses, multiple resistance to major pests and soil diseases, compatibility and vigor reduction features to implement high density production systems. These traits are considered strategic to the Chilean fruit sector, especially for the export-oriented industry.

Acknowledgments: CONICYT-REGIONAL/GORE O'HIGGINS/CEAF/R08I1001.

OS17

CONSTRUCTION OF A HIGHLY DENSE MAP OF SWEET CHERRY AND ITS COMPARATIVE ANALYSES WITH OTHERS PRUNUS LINKAGE MAPS

Carolina Klagges^{1,4}, Alejandra Guzmán², Levi Mansur², Eduardo Gratacós²,
Herman Silva³, Lee A. Meisel⁴

carolako@gmail.com

¹Doctorado en Biotecnología del Universidad Andrés Bello, Facultad de Ciencias Biológicas, República 217, Santiago. ²Estación Experimental La Palma, Facultad de Agronomía, Pontificia Universidad Católica de Valparaíso, Quillota. ³Laboratorio de Genómica Funcional y Bioinformática, Departamento de Producción Agrícola, Facultad de Ciencias Agronómicas, Universidad de Chile, Santiago. ⁴INTA-Universidad de Chile, Santiago.

Sweet cherry (*Prunus avium*) is an important fruit tree crop. The construction of a sweet cherry genetic linkage map is important to facilitate quantitative trait locus analyses and gene tagging for marker assisted breeding programs. In this work, high-density genetic map for two parents and a F1 population obtained from an intra-specific cross between 'Black Tartarian' × 'Kordia' (BT×K) was constructed using the recently designed RosBREED 6K Sweet Cherry SNP chip. A total of 5,696 SNP markers were tested in each progeny, mapping 723 SNPs in the BT×K linkage map. The resulting map spanned 752.9 cM with an average distance of 1.1 cM between adjacent markers. The map displayed high synteny and co-linearity compared with others linkage maps and with the peach genome v1.0 for all eight linkage groups (LG1-LG8). However, we identified inversions in sweet cherry genome when compared to the peach genome at LG1, LG5 and LG7. The highly dense linkage map developed from this segregating population of contrasting cherry cultivars has been incorporated into the Genome Database for Rosaceae (GDR) in the Comparative Map Viewer for Rosaceae (GDR-CMap) such that these maps may be used by the international community to develop markers associated with economically important traits.

Acknowledgements: 07CN13PBT-167 INNOVA Chile CORFO project, CONICYT doctoral fellowship (N°21120115 and N°24121484). UNAB DI-78-12/I project, Fondecyt N°1120261 and N°1121021.

OS18

**OVERVIEW: IMPROVING SEMOLINA YELLOWNESS OF DURUM WHEAT
(*TRITICUM TURGIDUM* L. VAR. *DURUM*) IN CHILE**

Albert Schulthess¹, Karen Campos¹, Nicolás Jiménez¹, Iván Matus², Andrés Schwember¹

awschult@uc.cl

¹Pontificia Universidad Católica de Chile. Facultad de Agronomía e Ingeniería Forestal, Santiago. ²Instituto de Investigaciones Agropecuarias, Centro Regional de Investigación Quilamapu, Chillán.

Semolina yellowness of durum wheat (*Triticum turgidum* L. var. *durum*) is considered an important trait which determines the organoleptic quality of pasta. In most studies semolina yellowness is described as a complex inherited trait, although it has a strong genotypic component, facilitating the breeding for high semolina yellowness internationally. However, in Chile this quality trait has not got much attention until recently. Under the same environmental conditions, a group of commercial Chilean cultivars showed intermediate to low levels of semolina yellowness relative to elite international genotypes. This reflects the current need of plant breeding efforts for improving semolina color in Chile. The standard variety field trials of INIA grown across different environments along the Chilean production zone of durum wheat have shown that semolina yellowness is mainly determined by the genotypic factor, which could facilitate future breeding efforts for improving this trait. There is no significant correlation between semolina yellowness and grain yield, suggesting that breeding for high semolina yellowness / high yielding varieties would be possible. Additionally, some genotypes from these trials, when compared to the current INIA commercial varieties, stood out in terms of semolina yellowness. A population of genotypes grown in two locations in Chile showed high variability for semolina yellowness due to a strong genotypic component. However, *PSY1A* and *PSY1B* loci, candidate genes for semolina yellowness, were almost fixed for this population, which precluded the search of marker-trait associations but also confirmed the complex inheritance of semolina yellowness. Finally, the expression patterns of *PSY1* homolog genes during grain development are currently being studied to search for their possible association with semolina yellowness. Keywords: durum wheat, genotype, semolina yellowness, *PSY1A*, *PSY1B*.

Acknowledgements: Fondecyt de Iniciación 11110066, 2011-2014 and INIA-Quilamapu.

OS19

**CHARACTERIZATION OF CHOCLERO MAIZE (*ZEA MAYS* L.) POPULATIONS
ASSESSED BY MORPHOLOGICAL TRAITS AND SSR MOLECULAR MARKERS**

Erika Salazar^{1,3}, José Correa¹, María José Araya, Marco Méndez², Carolina Araya¹,
Boris Sagredo⁴, Basilio Carrasco³

esalazar@inia.cl

¹Instituto de Investigaciones Agropecuarias, CRI La Platina, Santiago. ²Universidad de Chile, Facultad de Ciencias, Santiago. ³Pontificia Universidad Católica de Chile, Facultad de Agronomía e Ingeniería Forestal, Santiago. ⁴Instituto de Investigaciones Agropecuarias, CRI Rayentué, Rengo.

Diversity characterization allows for the understanding of genetic-environmental relationships between populations, facilitating their use by breeders. This study evaluates the morphological and molecular variability of 34 accessions of Choclero maize landraces collected between the III and VIII region. For morphological characterization, trials were conducted on 3 localities. The experimental design was a randomized complete block with 3 replicates, measuring a total of 41 traits. All quantitative characters, except tillering index and number of ears by tiller showed significant differences. Based on the correlation matrix, a principal component analysis (PCA) was conducted. A cluster analysis after standardization of the variables was also constructed. Dendrogram showed four main groups, which had some correspondence with the clustering observed in the first two axes of the PCA. The description of the groups was complemented with relevant qualitative characters descriptions for each group. The results showed the existence of morphological inter population variability within Choclero race, evidenced by the formation of clearly distinguished groups, in part explained by the diversity of environments of the collection. To validate grouping, a discriminant analysis was performed with the formed groups at two different distances in the cluster analysis. At the molecular level, at least 16 individuals per population were characterized with 10 SSR widely distributed in the genome. A preliminary analysis of the molecular data showed genetic differences among populations indicating the possibility of existence of population structure at the genetic level.

Acknowledgements: CONICYT scholarship Doctorado en Chile 2008 (21080026).

OS20

**LIPOXYGENASE AND APPLE AROMA: GENE EXPRESSION OF LIPOXYGENASE
FAMILY MEMBERS IN APPLE PEEL**

Carolina Contreras and Randolph Beaudry

contre33@msu.edu

Department of Horticulture, Michigan State University, East Lansing, MI 48824, USA.

Many aroma volatiles in fresh apple are produced via cellular disruption due to cutting or mastication. Six-carbon (C_6) volatiles, including the aldehydes trans-2-hexenal, hexanal and cis-3-hexenal, as well as their corresponding alcohols, are produced from action of the lipoxygenase (LOX) pathway on substrates released by tissue disruption. LOX genes are classified based on function and are grouped into 13-LOX and the 9-LOX groups, which generate C_6 and C_9 aldehydes, respectively. Another classification system is based on structure with those having a putative chloroplast transit peptide called *type-2* LOXs, and those that do not are called *type-1* LOXs. All 13 LOXs are thought to be *type-2* LOXs. It has been proposed that a 13-LOX gene with a chloroplast transit peptide may be involved in apple aroma. In our work, twenty-two lipoxygenase gene sequences were retrieved from the apple genome to identify possible LOX gene candidates that might participate in the aroma production in apple. We isolated RNA from apple skin for 8 time points throughout ripening (immature to senescent stage), made cDNA and performed semi-quantitative RT-PCR for all 22 LOXs. The expression of most of genes exhibited no discernable pattern during ripening; however, at least 6 LOXs were highly expressed and ripening-dependent. qRT-PCR was performed on these 6 LOX candidates. Of these, 4 LOX genes were down-regulated during ripening, and 2 LOX genes were up-regulated as ripening progressed. Changes in the lipid profile and C_6 aldehyde and alcohol production correlated (positively and negatively) with changes in gene expression data for the 6 LOX genes, suggesting they are good candidates for further investigation into their involvement in the biosynthesis of disruption-dependent aroma volatiles. Confocal microscopy analysis and biochemical characterization of apple LOX proteins was performed to interpret gene expression data and to more fully understand the role of LOX in aroma formation.

OS21

EXPRESSION ANALYSIS OF GENES OF THE GIBBERELLIN METABOLIC AND SIGNALLING PATHWAYS OF SEGREGANTS OF A 'RUBY SEEDLESS' X 'SULTANINA' POPULATION

Gonzalo Ravest^{1,2}, Sebastián Silva¹, Maribel Mamani^{1,2}, Patricio Hinrichsen¹

¹INIA CRI-La Platina, and ²Facultad de Agronomía, Universidad de Chile. Santiago, Chile.

phinrichsen@inia.cl

Chile is the first world exporter of table grapes. However, this industry is until now almost entirely based on imported genetics. Because of this, a breeding program was initiated ca. 20 years ago, focused in traits that included berry size as one of the main targets to improve. Gibberellins (GAs) are phytohormones thought to have a crucial role in cluster and berry development and growing on table grapes, and so the genes involved in their metabolism and signal transduction are evident candidates for research on the genetic determinism of berry size. However, little is known about the signaling processes that trigger these changes in the fruit. Reports of GA signaling in model plants indicate the interaction between a GA receptor (GID1) and a GA repressor (DELLA genes) that regulate the expression of GA-responsive genes. In this work, we searched for GID1 and DELLA orthologues in the *Vitis vinifera* L. reference genome (PN40024), performing transcriptional analyses in segregants of a 'Ruby Seedless' x 'Sultanina' population with contrasting phenotypes for fruit size. We found two GID1 and three DELLA orthologues, which expression is currently under evaluation by qPCR. Also, we looked at the series of gibberellin oxidase genic isoforms participating in the synthesis (GA20ox and GA2ox) and degradation (GA3ox) of GA, and the relation of their expression with the phenotypes of the studied segregants. We found that GA20ox4 and GA20ox2 have higher expression levels on fruits with larger sizes at 50% flowering and fruit-setting stages. In the case of GA3ox4, larger fruits had higher expression than smaller ones; GA2ox2, instead, showed higher expression levels at 6-8 mm in segregants of smaller berry sizes. This information gives insight on the metabolic processes involved in determining the fruit size and shows the importance of a better understanding of the signaling process of gibberellins.

Acknowledgements: Genoma-Chile grant FONDEF G071-1002.

OS22

**PHOSPHATE SOLUBILIZING BACTERIA NATIVE PLANT GROWTH PROMOTING
IN THE AREA OF LA SERENA, IV REGION OF COQUIMBO**

Andrés Rodríguez-Aguilera^{1,2}, Stefanie Maldonado², Alexandra Stoll², Jaime Bravo³

androdriguez@utalca.cl

¹Programa de Magister en Horticultura, Facultad de Ciencias Agrarias, Universidad de Talca.

²Centro de Estudios Avanzados en Zonas Áridas (CEAZA), La Serena. ³Microbiología, Universidad Politécnica de Pénjamo, México.

Ecosystems productivity is based on fine balances that operate at different associative levels. In this context, plant-microorganisms interactions constitute an important pillar to maintain the richness of biotic resources. Part of these interactions occurs in an area known as rhizosphere. This area is defined as a dynamic environment where the exudates, produced by the plants roots, determine the identity and structure of microbial community. This work is focused on characterizing native phosphate solubilizing bacteria isolated from the rhizosphere of the fourth region of Chile agricultural soils. The results suggest that these bacterial isolates are capable of solubilizing the inorganic phosphorus efficiently generating compounds that support the plant growth such as plant hormones, siderophores. Also we propose using 1-aminocyclopropane-1-carboxylic acid (ACC) as the only nitrogen source, intervening in the ethylene synthesis, which influences strongly in plant stress responses. This improves plant germination and enhances growth. Besides, the bacteria strain was able to induce changes in root patterns, and improve biomass in lettuce varieties that are commonly used for productive agricultural areas of Northern Chile. Finally, we propose to design a bio formula adapted at conditions that will have a real contribution to the demands and sustainability in favor of people's life quality.

Acknowledgements: This work was funded by the project GORE Coquimbo FIC 2012 BIP 30127532-0.

OS23

**FUNCTIONAL CHARACTERIZATION OF TWO NUCLEOTIDE SUGAR
TRANSPORTERS IN VITIS VINIFERA**

Daniella Utz, Michael Handford

daniutz@ug.uchile.cl

Laboratorio de Biología Molecular Vegetal, Facultad de Ciencias,
Universidad de Chile, Santiago.

The plant cell wall is mainly composed of polysaccharides, of which the non-cellulosic polysaccharides are synthesised in the Golgi apparatus. Here, glycosylation reactions are catalysed by glycosyltransferases whose substrates are sugars activated by the addition of a nucleotide, most of which are made in the cytosol. To transfer nucleotide-sugars into the Golgi lumen, a family of nucleotide-sugar transporters (NSTs) are localised in the membrane of the Golgi apparatus, although several are also localised in the endoplasmatic reticulum. Some NSTs in *A. thaliana* have been functionally characterised; the GONST1-5 family is specific for GDP-sugars and localised in the Golgi. In *Vitis vinifera* L., it has been determined that polysaccharides contain sugars derived from GDP-sugars. To determine the conservation of the mechanism involved in the synthesis of polysaccharides, we analysed bioinformatically the grapevine genome (Pinot noir PN40024) for the presence of GONST orthologues. We found two sequences with $\geq 78\%$ identity at the amino acid level, both of which possess the molecular characteristics of the GONST1-5 family. We have called these orthologues VvGONST-A and VvGONST-B. The cloning of both NSTs from the Thompson Seedless variety was performed successfully, and both are expressed throughout berry development and mature leaves as determined by qRT-PCR. By transient transformation of tobacco leaves with VvGONST-A::GFP and VvGONST-B::GFP, we determined that both are localised in the Golgi, as demonstrated by their sensitivity to brefeldin A and their colocalisation with a marker of the Golgi apparatus. Finally, assays are underway to determine the transport specificity of VvGONST-A and VvGONST-B using radiolabelled nucleotide-sugars.

Acknowledgements: This work was supported by CONICYT 21090418 and 24120980.

**bZIP25 A NOVEL TRANSCRIPTIONAL REGULATOR OF PROTEIN TRAFFICKING
INVOLVED IN ENDOCYTOSIS REGULATION IDENTIFIED BY A SYSTEM BIOLOGY
APPROACH IN *ARABIDOPSIS THALIANA***

Lorena Pizarro¹, Claudio Osorio¹, Alexander Vergara, Marcela Rojas-Pierce², Rodrigo Gutiérrez³,
Glenn R. Hicks⁴, Lorena Norambuena¹.

lorepizaro@gmail.com

¹Plant Molecular Biology Laboratory. Faculty of Science. University of Chile. ²Department of Plant Biology. North Carolina State University. ³Plant Systems Biology Lab. Molecular Genetics and Microbiology Department. Faculty of Biological Sciences. Pontificia Universidad Católica de Chile. ⁴Institute for Integrative Genome Biology & Center for Plant Cell Biology and Department of Botany and Plant Sciences. University of California.

Protein trafficking through endomembrane system is essential for accurate protein localization, function and turn-over. Thus, many physiological processes such as development, and salt resistance depend on protein trafficking. Over the past few years, transcriptional regulation has been described as an important component in trafficking regulation. However, no transcription factor (TF) related to this process has been reported in plants. We have used a systems biology approach in *Arabidopsis* for searching TFs involved in trafficking regulation. We built a co-expression network that connects TFs and trafficking protein-encoding genes, hereinafter called trafficking-genes, obtaining the Trafficking Regulatory Network. In this network, an interaction is established when a TF is co-expressed with a trafficking-gene (co-expression interaction) and the TF binding-site is over-represented in the trafficking-gene promoter (binding-site interaction). The Trafficking Regulatory Network has 744 interactions established between 276 TFs and 124 trafficking-genes. In order to select the best TFs candidates to be trafficking regulators, the TFs were ranked according to topological network analysis, co-expression interaction and binding-site interaction degree. The TF bZIP25 was ranked on the top. Transcript analysis of a bZIP25 defective mutant, *bzip25-2*, suggests that this TF is a negative regulator of trafficking-genes related to endocytosis such as *SYP122* and *ARA6*. Consistently, this transcriptional regulation is tightly correlated with higher rate of endocytosis in *bzip25-2* compared to wild-type, validating our systems approach. Significantly, *bzip25-2* shows alterations in phenotypes related to trafficking, such as root development and salt resistance that correlate with its endocytosis rate. Overall our evidence supports a role for bZIP25 in regulating trafficking, particularly endocytosis, which results in impacts on physiological processes. These results identify bZIP25 as a novel transcriptional regulator of endocytosis, and the first report of a TF involved in protein trafficking regulation in *Arabidopsis*.

Acknowledgements: FONDECYT 1120289 and 11080240. MECESUP short-term and CONICYT Graduate-Student Fellowship. CONICYT PhD-thesis Grant.

OS25

THE NAC FAMILY TRANSCRIPTION FACTORS NTL9 AND NAC017 PARTICIPATE IN ENDOCYTIC TRAFFICKING REGULATION IN ARABIDOPSIS THALIANA.

Claudia González-Ramírez, Lorena Pizarro, Lorena Norambuena

escarlata228@gmail.com

Plant Molecular Biology Laboratory. Faculty of Sciences. University of Chile.

Cell protein trafficking relies upon a set of membranous structures referred as the Endomembrane System (ES), which in plants include the plasma membrane, early and late endosomes, trans-Golgi network, Golgi apparatus, endoplasmic reticulum and vacuole. Trafficking among these compartments comprises the endocytic route, where plasma membrane components are internalized to endosomes and eventually reach the vacuole; and the secretory route, where newly synthesized proteins at the endoplasmic reticulum are targeted to ES compartments. Although protein trafficking is important in different processes, little is known about the involvement of transcription factors (TFs) in regulating either ES components or trafficking pathways throughout cellular compartments. In our laboratory, the TFs NTL9 and NAC017 have been identified as putative ES regulators. To test their functional role, we have investigated endocytic trafficking of the tracer FM4-64 in *Arabidopsis thaliana* insertional mutants for these TFs. FM4-64 internalization indicates that two allelic mutants of *NTL9* have accelerated rate of endocytosis from plasma membrane to endosomes, suggesting that NTL9 is a negative regulator of this process. Interestingly, vesicle trafficking from endosomes towards the vacuole is not accelerated. On the other hand, NAC017 is a regulator of endocytic trafficking towards the vacuole since *NAC017* mutants have alterations on this pathway. As Sortin2 increases lateral roots formation in wild-type plants by a mechanism that depends on trafficking from late endosomes towards the vacuole, we investigated Sortin2 effect on NTL9 and NAC017 mutants. Both the loss of function, *ntl9-1*, and the likely gain of function, *nac017-4*, mutants exhibit decreased Sortin2-lateral root induction regarding wild-type plants. Consistently, neither *NTL9* nor *NAC017* mutants endocytosis rate towards the vacuole is increased by Sortin2 as in the wild-type plants. Overall, our results indicate that NTL9 and NAC017 participate in endocytic trafficking regulation involving endocytosis from plasma membrane and endosomal trafficking towards the vacuole.

Acknowledgements: This work was supported by Fondecyt n°1120289

Panel Sessions

PS1

**THE ACQUISITION OF SECONDARY STRUCTURE OF COLD RESPONSIVE (COR)
PROTEINS IN PLANTS RELATED WITH THE RESISTANCE TO STRESS CONDITIONS:
A MOLECULAR DYNAMICS STUDY OF THIS EVENT**

Carlos Navarro-Retamal¹, Julio Caballero¹, Jans Alzate-Morales¹, Anne Bremer²,
Lukasz E. Bucki², Anja Thalhammer¹, Dirk Hinch¹, Wendy González²

carlos.navarro87@gmail.com

¹Center for Bioinformatics and Molecular Simulations, University of Talca, Chile;

²Max Planck Institute of Molecular Plant Physiology, Germany.

Freezing and low temperatures are one of the most important environmental factors that affect plant growth. Some plant species are able to gain freezing tolerance by different changes that involved the expression of a large number of cold responsive (COR) genes, many of which encode LEA proteins. Dehydration stress-related late embryogenesis abundant (LEA) proteins have been found in plants (in the chloroplast and the cytosol), invertebrates and bacteria. Most LEA proteins are unstructured in solution but can form amphipathic alpha helix during drying. In the model plant *Arabidopsis thaliana* the LEA proteins are divided into nine phylogenetically unrelated groups according to their amino acid sequence similarity. The function of the LEA protein is presumed to confer desiccation and freezing tolerance to the plant, but the specific (functional) role is still unknown. *In vitro* studies suggest that LEA protein may have different roles: water binding, ion sequestration, stabilization of DNA/RNA, proteins, membranes, etc. We aim to clarify the functional role of two cold responsive proteins: cor15a (At2g42540) and cor15b (At2g42530) of *Arabidopsis thaliana* using molecular dynamics simulations in 1) full-water conditions, 2) full-glycerol conditions, 3) interacting with membrane and a desiccated system (in vacuo). We will try to elucidate the behavior of cor15 proteins on different solvents and how do they interact and protect macromolecules (like the lipids on the membrane) in stress conditions.

Acknowledgements: CNR thanks to doctoral fellowship awarded by Government of Chile through CONICYT N° 21120691.

PS2

PEACH (*PRUNUS PERSICA* (L) BATSCH VAR. O'HENRY) PROTEOME BIOINFORMATIC ANALYSIS FOR THE METABOLITE SEARCH OF QUALITY PREDICTORS

Gianfranco Benedetto¹, Carol Moraga¹, Ricardo Nilo¹, Ariel Orellana¹,
Andréa Miyasaka Almeida¹.

gianfrancobenedetto@gmail.com

¹FONDAP Center for Genome Regulation, Núcleo Milenio en Genómica Funcional de Plantas, Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello, Santiago, Chile.

Peach (*Prunus persica*) is a nutritional, much appreciated fruit and widely commercialized. Chile is the main producer and exporter of the southern hemisphere, hence is of vital importance that the product improves its quality for continuing attracting the markets. In this work, data from the analysis of the proteome of peach variety O'Henry in two developmental stages (mature and ripe fruit) was analyzed to look for metabolic pathways related to quality traits. Broad proteome analysis allowed the identification of 4105 proteins, of which 2615 were present in both developmental stages, and only 322 were differentially expressed. Metabolic model reconstruction was done using the bioinformatics tool, Paintomics. This method allows the integration of transcriptomics and metabolomics for the visualization of data, using the KEGG (Kyoto Encyclopedia of Genes and Genomes). In this way, mapping of the proteins allows the visualization of enriched metabolic pathways. In this work, 3620 proteins were mapped in different metabolic pathways in two developmental stages, where the most prominent pathways were: starch and sucrose metabolism, mannose and fructose metabolism, amino sugar and nucleotide sugar metabolism and glycolysis / gluconeogenesis. This analysis will help in expanding the knowledge of fruit quality and the developmental process of the fruit.

Acknowledgements: FONDECYT N°1130197; FONDAP CRG 15070009; Núcleo Milenio P10-062-F and Basal PFB-16.

PS3

**BINDING PREDICTION OF *PRXTH* ENZYME WITH PUTATIVE SUBSTRATES
BY MOLECULAR DOCKING, MOLECULAR DYNAMICS AND MM-GBSA**

Cristian Carrasco¹, Claudio Valenzuela¹, Patricio Ramos², María Alejandra Moya-León²,
Raúl Herrera².

cristiancarrascoarellana@gmail.com

¹Programa de Doctorado en Ciencias Mención Ingeniería Genética Vegetal,
Universidad de Talca; ²Laboratorio de Fisiología Vegetal y Genética Molecular,
Instituto de Biología Vegetal y Biotecnología, Universidad de Talca, Talca, Chile.

Different reports demonstrate that xyloglucan endotransglucosylase/hydrolase (XTH) enzyme participates in cell wall remodeling. XTH interacts with the hemicellulose fraction of plant cell wall, providing plasticity for cell elongation and development. Previously, a *PrXTH* gene was isolated from radiata pine and shown to be induced in response to tree bending, suggesting an active participation in compression wood formation. Xyloglucans are the main hemicelluloses in radiata pine, and they consist of a central cellulosic chain with substitutions of different types. The structural model of PrXTH protein was generated through comparative modeling methodology using 1UMZ (Ptt-xet16a) as template. The model is composed of 1 α helix, 4 α helices 3₁₀, 15 β -sheets and 19 loops, and presents a β -sandwich structure, which is a curvature generated by antiparallel β sheets, placing the conserved catalytic domain in the middle area of this slit. By using bioinformatic tools the predicted binding and complex stability of PrXTH protein with different xyloglucans (XXXGXXXG, XXFGXXFG, XLFGXLFG) and cellulose (GGGGGGGG) was evaluated. Molecular dockings predicted the conformation of protein-substrates interaction. Then, molecular dynamics was used to evaluate the protein-substrate stability. Finally, MM-GBSA was used to estimate the total energy of protein-substrate complexes ($\Delta\Delta G$) and also to determine individual energies that participate in the binding. The results indicate that PrXTH has the best stability with XXXGXXXG substrate, and the substrate locates in the concave face of PrXTH model, in parallel to the β -sandwich structure. In addition, Van der Waals forces are the most relevant energies for protein-substrate interaction.

Acknowledgements: Fondecyt N°1120635.

PS4

**IDENTIFICATION OF HORMONAL RESPONSIVE ELEMENTS IN THE PROMOTER
REGIONS OF PYR/PYL GENES WITHIN THE FRAGARIA VESCA GENOME**

Yazmina Stappung, Rodrigo Lizana, Raúl Herrera, María Alejandra Moya-León.

ystappung@alumnos.utalca.cl

Laboratorio Fisiología Vegetal y Genética Molecular, Instituto de Biología Vegetal
y Biotecnología, Universidad de Talca.

The woodland strawberry, *Fragaria vesca*, is the first genome sequenced of the *Fragaria* genus. Due to its substantial sequence identity it serves as a reference model for species such as the cultivated strawberry (*Fragaria x ananassa*) and the Chilean strawberry (*Fragaria chiloensis*). Strawberry is a non-climacteric fruit, and its ripening seems to be coordinated by the action of some hormones like auxins (AUX) and abscisic acid (ABA). However, there is still scarce genetic/molecular evidence. To understand the hormone regulatory mechanism, it could be useful to identify hormone recognition sites in the promoter regions of target genes. With this objective, we designed a bioinformatic workflow in order to identify ABA and AUX responsive elements in genes involved in strawberry ripening development. We develop a strategy to get in an automatic way a list of candidate genes and their promoter regions (2000 bp upstream from 5'UTR). With this information, a search in Plantpan database was performed to identify different responsive elements. Then a redundant filter was performed and a detailed image of the localization of each hormone responsive element was built. To test this methodology, three different ABA receptor genes in *F. vesca* (*FvPYR1*, *FvPYL3* and *FvPYL7*) were analyzed. From the visual chart of each promoter region the abundance and distribution of different hormone responsive elements can be appreciated. This allows us to identify the potential hormonal regulation of these genes. The three of them have ABA responsive elements; *FvPYL3* and *FvPYL7* also have AUX responsive elements, although *FvPYR1* does not have them. This indicates that these genes might have differential hormonal regulation.

Acknowledgments: ANILLO ACT 1110 project.

PS5

**UNDERSTANDING THE ROLE OF SER370 OF ALCOHOL ACYLTRANSFERASE
FROM MOUNTAIN PAPAYA FRUIT**

Maryela Saens¹, Luis Morales-Quintana^{1,2}, María Alejandra Moya-León¹, Raúl Herrera¹

msaens@alumnos.otalca.cl

¹Laboratorio de Fisiología Vegetal y Genética Molecular, Instituto de Biología Vegetal y Biotecnología, Universidad de Talca; ²Programa de Doctorado en Ciencias Mención Ingeniería Genética Vegetal, Universidad de Talca.

In mountain papaya fruit (*Vasconcellea pubescens*) the aroma is mainly determined by esters which are produced by esterification reactions catalyzed by the enzyme alcohol acyltransferase (VpAAT1). This enzyme can use a variety of alcohols and acyl-CoAs as substrates. Previous studies have shown that VpAAT1 protein has a solvent channel in the center of the structure formed between two major domains, with residues H166 and D170 from the HxxxD motif exposed to the solvent channel. Through *in silico* studies a serine residue (S363) has been highlighted to be important for substrate interaction and to stabilize the alcohols within the solvent channel of CmAAT1 enzyme from *Cucumis melo*, a phylogenetic related enzyme to VpAAT1. The equivalent residue in VpAAT1 corresponds to S370 that is located in the center of the solvent channel in front of the catalytic residues. In order to clarify the role of S370 in ester biosynthesis, *in silico* VpAAT1 mutants were designed: S370A and S370T. Molecular dockings, molecular dynamics and MM-GBSA methodologies provided a detailed characterization of the protein structure and its interaction with several substrates. Enzyme-substrate interaction energies were less favorable for the mutants compared to the native protein indicating that serine substitution affects the interaction of VpAAT1 with the substrates. Using molecular dynamics the position of the substrates was explored and revealed that serine substitutions by alanine or threonine avoid the adequate interaction of substrates with the residues of the active site. Finally, the results obtained by MM-GBSA confirm the same findings. The data presented provided further evidence of the importance of Ser370 residue for VpAAT1 activity.

Acknowledgments: LMQ acknowledges CONICYT for a Doctoral fellowship. This research was supported by Anillo ACT-1110 project.

**MOLECULAR MODELLING AND DOCKING SIMULATIONS OF FCEXPA2
FROM *FRAGARIA CHILOENSIS***

Felipe Valenzuela¹, Luis Morales-Quintana¹, Carlos Gaete-Eastman², Raúl Herrera²,
María Alejandra Moya-León².

fvalenzuelar@alumnos.utalca.cl

¹Programa de Doctorado en Ciencias Mención Ingeniería Genética Vegetal,
Universidad de Talca; Laboratorio de Fisiología Vegetal y Genética Molecular,
Instituto de Biología Vegetal y Biotecnología, Universidad de Talca, Talca, Chile.

Chilean strawberry (*Fragaria chiloensis*) fruit shows a very short shelf life due to its fast softening rate. Fruit softening has been related to cell wall remodeling. Changes in the cellulose-hemicellulose fraction have been reported during ripening of *Fragaria chiloensis* fruit, in addition to the accumulation of FcEXPA2 transcripts. To gain insight about the protein structure of FcEXPA2 and its mechanism of action at the molecular level, the structure of the protein was built by comparative modeling methodology. The protein structure was validated and refined through molecular dynamics simulation. The model obtained for FcEXPA2 comprises two domains (D1 and D2), with 12 β -sheets and 4 α -helices. D1 is a β -barrel and resembles the structure of family-45 glycoside hydrolases (GH45), conserving almost all the residues of the catalytic site. D2 is a β -sandwich with aromatic residues forming the potential polysaccharide binding surface. On the other hand, the interaction of FcEXPA2 with a set of putative substrates such as xyloglucans (XGs) and cellulose was explored using molecular docking simulations. The results of the interaction between FcEXPA2 protein with these potential substrates suggest that stable conformational complexes are formed, with favorable affinity energies for the binding of XGs and cellulose. The data is congruent with a probable role of FcEXPA2 protein in the disassembling of the cellulose-XG matrix during ripening of strawberry fruit.

Acknowledgements: LMQ grants to CONICYT for a Doctoral fellowship. This research was supported by Anillo ACT-1110 and Fondecyt N° 1110792 projects.

PS7

EXPRESSION, CLONING AND IN SILICO STRUCTURAL ANALYSIS OF NON SYMBIOTIC HEMOGLOBINS FROM STONE FRUIT ROOTSTOCKS (*PRUNUS SP. L.*)

Matías Troncoso¹, Manuel Acuña¹, Luis Morales-Quintana², Raúl Herrera²,
Paula Pimentel¹, Boris Sagredo¹, Rubén Almada¹

mtroncosoa@alumnos.otalca.cl

¹Centro de Estudios Avanzados en Fruticultura, INIA CRI Rayentué, Rengo, Chile;

²Laboratorio de Fisiología Vegetal y Genética Molecular, Instituto de Biología Vegetal
y Biotecnología, Universidad de Talca, Talca, Chile.

Root hypoxia limits stone fruit tree development. To overcome this problem, stone fruit trees are usually grafted on rootstocks (species of the *Prunus* L. genus) with different degrees of tolerance to root hypoxia. However, the molecular base of such variability is largely unknown. In a number of crops and tree species, non-symbiotic hemoglobin (Hb)-like genes stand out among hypoxia-related genes, but no such studies have been done with the *Prunus* species used as rootstocks. In this study, we analyzed the expression pattern of class 1 non-symbiotic Hb-like (*nsHb*) in roots of *Prunus* rootstock with different responses to this stress. We observed that the putative *Prunus nsHb* was induced by root hypoxia in all analyzed *Prunus* genotypes. However, *Prunus nsHb* had higher expression levels in roots of tolerant rootstocks. On the other hand, elucidating the structure of nsHbs is of interest because this information could be essential to fully understanding the function of these proteins in rootstock responses to root hypoxia. Therefore, five previously uncharacterized class 1 *nsHb* cDNAs were isolated from M2624 (*P. cerasifera* x *P. munsoniana*), Cab6P (*P. cerasus*), GxN (*P. dulcis* x *P. persica*), Colt (*P. avium* x *P. pseudocerasus*) and F12 (*P. avium*) rootstocks which are highly tolerant, tolerant, sensitive, moderately sensitive and extremely sensitive to root hypoxia, respectively. Class 1 *nsHb* genes are predicted to encode a protein of 157 amino acid residues. When *Prunus nsHbs* were compared to each other a higher degree of amino acid identity (97%- 99%) was observed. Then, we modeled the tertiary structure of M2624's and F12's *nsHb* proteins by comparative modeling. The predicted *Prunus nsHbs* folded into the globin fold with 3/3 alpha-helical organization and heme-Fe hexacoordination. Molecular docking simulations will be employed to understand the interaction between the nsHbs and putative ligands such nitric oxide and oxygen.

Acknowledgments: CEAF_R08I1001 and FONDECYT N°11110079. Rootstock plants were gently provided by Agromillora Sur S.A.

PS8

TRANSCRIPTOMICS IN *HYMENOGLOSSUM CRUENTUM* DURING A DESICCATION-REHYDRATION CYCLE

Giovanni Larama^{1,2}, **Graciela Berrios**¹, Ana Gutierrez^{1,3}, Patricio Manque⁴ León Bravo^{1,3}

gberr001@gmail.com

¹ Laboratorio de Fisiología y Biología Molecular Vegetal. Facultad de Ciencias Agropecuarias y Forestales. Departamento de Producción Agropecuaria. UFRO; Centro de Modelación y Computación Científica, Universidad de La Frontera. Temuco, Chile; ³ Center of Plant, Soil Interaction and Natural Resources Biotechnology, Scientific and Technological Bioresource Nucleus. UFRO; Centro de Genómica y Bioinformática, Universidad Mayor, Santiago Chile.

Hymenoglossum cruentum is an epiphyte filmy fern from *Hymenophyllaceae* family, this is a poikilohydric desiccation tolerant species, capable of survive for about a month in a dehydrated state without suffering significant cellular damage and it recovers full functionality after rehydration. Therefore, it is a very interesting model to study the molecular mechanisms involved in desiccation tolerance and discover new genes associated to cell stability and fast recovery from almost dryness. To achieve this goal, RNA from three hydration states of *Hymenoglossum cruentum* during desiccation-rehydration cycle was sequenced by Illumina MiSeq platform. The transcriptome was assembled de novo and analyzed in order to identify differentially expressed genes related to desiccation tolerance. The results from statistical analysis showed 297 genes that significantly ($P < 0.05$) change their expression level upon dehydration and rehydration. Identification was carried out using Blast+ software with UniProt database, being able to identify four interesting groups of genes. First, a group of highly hydrophilic proteins named LEA (ASR1, LEA-D113 and Dehydrins) related to cellular components stabilization, this group increases their relative abundance from 13.2% to 28%. Second, up regulation of highly hydrophobic peptides named RCI (Rare Cold-Inducible) that has been shown to be localized in membranes performing a protective role. Third, a group of cytoskeleton components, the expression of dynein light chain and formin-like protein were altered, suggesting changes in cytoskeleton during desiccation-rehydration cycle. Fourth, a group of stress related protein, up regulation of oxidative stress response (Ferritin, glutathione S-Transferase and Catalase) and finally two transcription factors (DREB and DOG1). There is also a high percentage (25%) of sequences that were not identified by similarity using BLAST in public databases. These genes are currently being validated by qRT-PCR. Results showed a variety of mechanism related to desiccation tolerance in *H. cruentum* contributing to improve the molecular knowledge in resurrection plants.

Acknowledgements: FONDECYT N° 1120964.

PS9

**TRANSCRIPTOME ANALYSIS BY RNA-SEQ DURING DEVELOPMENT AND RIPENING
OF RASPBERRY FRUIT (*RUBUS IDAEUS* HERITAGE).**

Aníbal Ayala ^{1,2}, Dante Travisany ³, Alex Di Genova ³, Liliam Monsalve ^{1,5},
Juan Pablo Martínez ^{2,1}, Bruno Defilippi ⁴, Alejandro Maass ³, Lida Fuentes ^{1,2}.

lfuentes@creas.cl

¹Centro Regional de Estudio en Alimentos y Salud (CREAS), Valparaíso, Chile, INIA La Cruz,
La Cruz, Valparaíso, Chile ², Centro de Modelamiento Matemático (CMM), Facultad de
Ciencias Físicas y Matemáticas, Universidad de Chile, ³Centro de Regulación del Genoma,
FONDAP 15090007. ⁴Unidad de Postcosecha, INIA La Platina, Santiago, Chile,
⁵Universidad Católica de Valparaíso, Valparaíso, Chile

Raspberry (*Rubus idaeus*) is an economic important fruit, with a rapid ripening rate and a very short shelf life. The softening progress is a consequence of changes in the composition and architecture of the cell wall. These changes in cell are due to enzymes that modify and degrade the cell wall. With the aim to screen differentially expressed genes during development and ripening of red raspberry fruit, we use an RNA-Seq approach, using RNA libraries generated at flower (BBFL), developing (BBVE) and ripening fruit stages (BBSM) and sequenced in an Illumina HiSeq 2000 platform generating approximately 373 Million paired end reads (101-bp in size). Then using 298 million high-quality reads from all libraries we assembly and generate the fruit transcriptome of red raspberry fruit. A set of 41,650 high quality transcripts were generated and 24,509 CDS were identified using a mix of *ab-initio* and comparative methods. A total of 18,171 CDS were successful characterized using several databases including UniprotKB, NCBI Non-Redundant, KEGG, Gene Ontology among others also including InterPRO-Scan in order to search for conserved domains. Using a pipeline for differential expression analysis a total of 2409 transcripts were identified as differentially expressed between the three libraries. The data can be accessed at: <http://rubus.cmm.uchile.cl>. Transcripts associated to cell-wall such as polygalacturonase, expansin and pectate lyase were detected during all stages of fruit development, but increasing during ripening stage. In addition, transcript levels for cell wall related genes were evaluated by qRT-PCR.

Acknowledgements: FONDECYT 11110438, "Centro Regional de Estudios en Alimentos Saludables, CONICYT-REGIONAL, GORE Región de Valparaíso, R12C1001", CIRIC INRIA-Chile, Powered@NLHPC(ECM-02) supercomputing infrastructure.

PS10

**DEVELOPMENT OF EST SSR BASED ON THE FIRST TRANSCRIPTOME
OF *PRUNUS SALICINA***

Máximo González¹, Erika Salazar², Paloma Morales¹, Isidora Mura¹,
Jonathan Maldonado³, Herman silva³, Basilio Carrasco¹.

mpgonza2@uc.cl

¹Laboratorio de Genética Molecular, Facultad de Agronomía e Ingeniería Forestal, Pontificia Universidad Católica de Chile, Santiago, Chile. ². Unidad de Recursos Genéticos, CRI La Platina, Instituto de Investigaciones Agropecuarias, Santiago, Chile. ³. Laboratorio de Genómica funcional y bioinformática, Facultad de Agronomía, Universidad de Chile.

Chile is the largest exporter of Japanese plum (*Prunus salicina*) in the world. In order to respond to the needs of the productive sector, especially in regard to the supply of new varieties with better agronomic and quality characteristics, Chile kicked off a new breeding program for this species. In many fruit trees the development of molecular markers has allowed the construction of saturated genetic maps, to which genes controlling important traits are assigned. Markers flanking these genes play an important role in selecting parental combinations and seedlings with positive traits, but also are important in detecting recessive traits, enable pre-selection for polygenic quantitative traits and contribute to the understanding of biochemical and physiological processes involved in the pathway of important traits. Despite this, the availability of molecular markers for *Prunus salicina* is limited. In order to understand some of the molecular mechanisms involved in the Japanese plum fruits, we did two total RNA libraries by using Illumina technology and therefore, the first transcriptome of this specie was developed. We generate ~55,000 contigs with 2.728 repeated sequences with potential to be used as SSR molecular markers. Thirty seven potential markers associated with the metabolic pathways of sugar, acidity and color were identified and all of them were analyzed in 29 commercial varieties.

Acknowledgments: This work was funded by the CONICYT scholarships "Doctorado en Chile 2009 (N° 21090118)", "Apoyo a la Tesis Doctoral 2011 (24110179)" and "Tesis en la Industria 2013 (N° 781211008)". Consorcio tecnológico de la Fruta.

PS11

**GENOMIC ORGANIZATION OF MYRCENE SINTETASE IN SCENTED LINES
OF *ALSTROEMERIA***

Gerardo Núñez¹, Daniela Elizondo¹, Danilo Aros², Claudio Meneses¹.

gera.nunez.lillo@gmail.com

¹Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello, República 217, Santiago, Chile; ²Universidad de Chile, Facultad de Ciencias Agronómicas, La Pintana, Santiago, Chile.

Alstroemeria species are geophytes, perennials and native to South America, with Chile and Brazil as major centers of biodiversity. *Alstroemeria* has become a major product in the global industry of cut flower and the new varieties developed are not scented, so the study of floral scent in this species is very interesting. A gene responsible for the biosynthesis of myrcene (*AlstroTPS*), a terpene present in the floral scent of *Alstroemeria* cv. 'Sweet Laura' has been previously identified and characterized. The aim of this study was to confirm the presence of myrcene synthase in 5 scented lines of *Alstroemeria* (selfing of *A. caryophyllaea*: DANCAR001, DANCAR003, DANCAR004, DANCAR006 and DANCAR017) and then characterize its genomic organization. As a negative control a Brazilian native species (*A. psittacina*) was also evaluated. For high quality DNA extraction the commercial kit DNeasy Plant Mini Kit (QIAGEN) was used. *AlstroTPS* was amplified and sequenced in Macrogen. The results were aligned and analyzed using the program Geneious 3.6.2. For all the samples evaluated the presence of an insertion of 105 bp was observed, suggesting that this may correspond to an intron that would alter the classification of this gene. Besides this variation, a deletion of 60 bp was identified only in lines DANCAR001, DANCAR004 and DANCAR006. This may explain segregation in the aroma of these individuals to produce proteins with reduced activity. Finally, we identified an insertion of 4 bp in *A. psittacina* that change the reading frame of the gene, generating a stop codon, which would explain the absence of aroma in this species. In conclusion, variations in the genomic structure of *AlstroTPS* identified through sequence analysis may explain the segregation of scent of *Alstroemeria* flowers. This data could be used to design selection markers for supporting breeding programs focused on the character of scent.

PS12

**THE t-SNARE SYP123 IS INVOLVED IN THE POLARIZED SECRETION OF PROTEINS
AT THE TIP OF GROWING ROOT HAIRS AND CONTRIBUTES TO PLANT
RESISTANCE AGAINST PATHOGENS.**

Begoña Galilea, Cecilia Rodriguez-Furlán, Camilo Recabarren, Ariel Orellana,
Francisca Blanco

i.galilea@uandresbello.edu

FONDAP Center for Genome Regulation, Núcleo Milenio en Genómica Funcional de Plantas,
Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello.

Intracellular membrane trafficking along with endocytic and secretory transport pathways plays an essential role in multiple cellular functions. Briefly, proteins destined for transport to distinct locations are collectively assembled into vesicles and delivered to their target site by vesicular fusion. SNARE (soluble N-ethylmaleimide-sensitive factor-attachment protein receptor) proteins are required for these events, during which v-SNAREs (vesicle SNAREs) interact with t-SNAREs (target SNAREs) to allow the specific transfer of cargo from donor vesicle to target membrane. Several t-SNAREs, members of the syntaxins subfamily (SYP), have also been related to the plant response against pathogens. Recently, SYP-123 has been shown to be specifically expressed in the root hairs and accumulated at the growing tip region of root hairs. In this work we analyzed if the polarized trafficking activity is related to the plant-defense response. Using a SYP123 knockout insertional mutant and the complemented plant (*syp123-/-/35S::SYP123-GFP*), we evaluated the role of SYP123 during pathogen defense response. For this purpose, we analyzed the effect of virulent bacterias in roots phenotype and their susceptibility to infection. To further investigate the role of SYP123 in pathogen defense we study the capacity of plants defective on SYP123 to established systemic acquired resistance. Since protein secretion may be altered, we measured extracellular PR1 as an indicator of antimicrobial protein secretion capability. These results indicate that SYP123 is involved in the polarized secretion of protein in growing root hairs and that this activity also contributes to the establishment of plant defense against pathogens.

Acknowledgments: Proyecto Regular UNAB N° 274, Fondecyt 11121387 and Núcleo Milenio P10-062-F.

PS13

**ANALYSIS OF THE REGULATED IRE1-DEPENDENT DECAY (RIDD) PATHWAY USING
UDP-GLUCOSE:GLYCOPROTEIN GLUCOSYLTRANSFERASE (UGGT) MUTANTS**

Sebastián Chávez, Adrian Moreno, Ariel Orellana y Francisca Blanco.

seb.chavez@uandresbello.edu

FONDAP Center for Genome Regulation, Millennium Nucleus for Plant Functional Genomics,
Plant Biotechnology Centre, School of Biological Sciences, Universidad Andrés Bello.

The mechanism of endoplasmic reticulum quality control (ER-QC), allows the correct synthesis and folding of proteins associated to the secretory pathway. This mechanism depends on the N-linked glycosylation of proteins that are clients of this process. One key player of this process is Glucosidase II (GII), because it is in charge of delivering the mono-glucosylated proteins (clients) to the calnexin/calreticulin (CNX/CRT) cycle. It also acts at the end of the cycle in order to get the proteins from the ER to its final destination. If correct folding by ER-QC is not accomplished, UDP-glucose:glycoprotein glucosyltransferase (UGGT) returns the proteins to the CNX/CRT cycle, to obtain the correct conformation. The GII mutant (*rsw3-1*) shows an abnormal development under stress conditions (heat), displaying an abnormal growth of the root tip, this phenomenon in particular has been denominated *radially swollen root*. This mutant also shows a lower production of cellulose. We propose that this reduction on the cellulose production, thus the *swelling*, is caused by a decreased expression of cellulose synthesis related genes (*CESA1* and *CESA3*), due to a degradation of mRNA by *IRE1* (*regulated IRE1-dependent decay: RIDD*), mechanism related to the *unfolded protein response* (UPR). We analyzed the expression levels of some genes associated to cellulose production: *CESA1*, *CESA3*, *BGAL10* and *BXL2*, in two UGGT mutants (*uggt1-1* and *uggt1-2*) using q-PCR. Also, we evaluated the presence of *root swelling* on those mutants under heat stress. Our results suggest a decreased expression of *CESA1* and *CESA3*. Additionally, *uggt1-2* shows a *root swelling*-like phenotype. These results give more information about the *RIDD* pathway under stress conditions, including some target genes.

Acknowledgments: FONDAP CRG 15070009, Fondecyt 1110954, Núcleo Milenio P10-062-F, Basal PFB-16 and Fondecyt 11121387.

PS14

PRODUCTION OF TRANS-RESVERATROL IN CELL SUSPENSION CULTURES OF BLUEBERRIES (*VACCINIUM CORYMBOSUM* L.) AND MURTILLA (*UGNI MOLINAE* TURCZ.)

Carolina Salazar¹, Evelyn Bustos², Matilde Uribe³, Claudia Pérez²

carolinasalazar@udec.cl

¹Departamento de Producción Vegetal, Facultad de Agronomía, Universidad de Concepción, Vicente Méndez 545, Chillán, Chile, ²Laboratorio de Química de Productos Naturales, Departamento de Botánica, Facultad de Ciencias Naturales y Oceanográficas, Universidad de Concepción, Concepción, Chile, ³Facultad de Ciencias Forestales-Laboratorio de Cultivo de Tejidos, Centro de Biotecnología, Universidad de Concepción, Concepción, Chile.

Trans-Resveratrol (3,5,4'-trihydroxystilbene) is a stilbene present naturally in plants with various health benefits, especially for their antioxidant properties, anti-inflammatory, anti-cancer and anti-diabetic, which allow prolonging the cell longevity. The present study reports for the first time, the quantification of *t*-resveratrol and *t*-piceid by HPLC in elicited cell suspensions of three ecotypes of *Ugni molinae* and two cultivars of *Vaccinium corymbosum*, endemic and cultivated species of Chile. For both species, leaves and fruit were established in vitro for the induction of callus and three elicitors were used in suspension; 10 mM β -cyclodextrin, 100 μ M of salicylic acid and 1% (v/v) of *Botrytis cinerea*. Stilbene quantification was performed in fruits, leaves, callus, cell aggregates and elicited medium. The response of the explants showed significant differences in contamination and callus formation. There were also differences between the genotypes in the production of resveratrol, with the best cell lines obtained in callus from immature fruits ecotype 3 of murtilla, with 1 mg L⁻¹ 2, 4-D, 0.5 mg L⁻¹ KIN and 1 mg L⁻¹ and from leaves of blueberries 'Legacy' with 5 mg L⁻¹ 2, 4-D, 1 mg L⁻¹ KIN and 1 mg L⁻¹ of ANA as growth regulators. The higher concentrations of *t*-resveratrol were obtained in ripe fruit of murtilla and blueberries, with values of 5100 g g⁻¹ and 558,4 mg g⁻¹ PF respectively and a total of 553,5 mg g⁻¹ FP and 441,6 mg g⁻¹ PF in murtilla and blueberries callus. Using the optimal culture conditions, these results may be scalable to an industrial level, being a viable alternative for obtaining resveratrol, enhancing these species agronomically to be highly competitive.

Acknowledgements: This thesis project was conducted with funding from Innova Bío-Bío, through the support line of Higher Education Graduate thesis, project number 12.247.

PS15

**bZIP25 TRANSCRIPTION FACTOR PARTICIPATES IN LATERAL ROOT DEVELOPMENT
REGULATING CLATHRIN-MEDIATED ENDOCYTOSIS**

Claudio Osorio, Lorena Pizarro, Lorena Norambuena.

anduin@ug.uchile.cl

Plant Molecular Biology Laboratory. Faculty of Science. University of Chile

Protein and membrane trafficking in plant cells is highly regulated through a dynamical system of membranous organelles. Functionally, the SE is divided into two trafficking routes: the secretory and the endocytic pathway. The endocytic pathway allows the internalization of extracellular components and plasma membrane into the cell. Endocytosis is essential for plant development processes, such as the multiple developmental stages during lateral root formation that includes initiation, patterning and emergence. Our laboratory has characterized the nuclear transcription factor bZIP25 which modulates protein and membrane trafficking in *Arabidopsis thaliana*. *bzip25-2*, a bZIP25 loss of function mutant, has an accelerated endocytosis and a greater number of lateral roots, supporting the connection between these two processes. Our goal is to study whether bZIP25 participates in lateral root development by regulating endocytosis. Lateral root development is induced by auxin, which inhibits endocytic trafficking. *bzip25-2* is sensitive to endocytosis auxin-inhibition of the tracer FM4-64. However, under auxin treatment *bzip25-2* has a drastic increase endocytic rate compared to wild-type. *bzip25-2* lateral roots are induced under auxin treatments, indicating its sensitivity to auxin also at this level. On the other hand the disruptor of clathrin-mediated endocytosis, TyrfostinA23 (TyrA23), blocks endocytosis and alters lateral roots development in wild-type plants. The *bzip25-2* endocytosis rate is unaffected by TyrA23 and, consistently, lateral roots emergence is not affected under treatment with this drug. TyrA23 *bzip25-2* resistance could be due to increments of proteins involved in the endocytosis, such as CLC2, overcoming the inhibition caused by the drug. Indeed *bzip25-2* has higher transcript level of CLC2. These results strongly suggest that bZIP25 participates in the lateral roots development, particularly in the emergence of lateral roots by modulating clathrin-mediated endocytosis independently of auxin signaling. These results provide more evidence for the endocytic trafficking role in postembryonic development of roots.

Acknowledgements: FONDECYT1120289

PS16

VISUALIZATION OF THE TISSUE SPECIFIC JASMONATE RESPONSE TO SALINE STRESS IN ROOTS OF TRANSGENIC *ARABIDOPSIS THALIANA* BY USING POSITIVE- AND NEGATIVE-REPORTER SYSTEMS

Giovanna Miranda, Mirko Pavicic, Pablo Figueroa.

gmiranda.flores3.0@gmail.com

Escuela de Biotecnología, Facultad de Ciencias, Universidad Santo Tomás, Santiago, Chile

Jasmonate (JA) is a critical hormone in plant responses to biotic and abiotic stresses. The build-up of salts on the surface of soil is a widespread agricultural problem affecting world's irrigated cropland. Understanding plant tolerance to salinity is therefore relevant. Microarray gene expression data sets available through public repositories show that JA early responsive genes are up-regulated by salt stress in *Arabidopsis thaliana* roots. However, reporter systems for studying the tissue and cell specific jasmonate response have not been well developed and validated as with other hormones. To test our hypothesis that JA signaling pathway is activated by salt stress, we analyzed the effect of salinity on the JA response by three different experimental approaches: 1. Plants containing a positive reporter system (*JAZ1pro::GUS*) where *GUS* expression is controlled by a JA-responsive promoter. 2. Plants containing a negative reporter system (*35Spro::JAZ1-GUS*) where the *JAZ1-GUS* chimeric protein is destabilized by JA and its expression is controlled by a constitutive promoter. 3. Measuring the relative transcript levels of several early JA responsive genes by RT-qPCR assays in roots. We observed that salt treatment (150 mM NaCl for 3 h) induces reporter accumulation and reduction, in the primary root of *JAZ1pro::GUS* and *35Spro::JAZ1-GUS* seedlings, respectively. The root tip was the zone where highest JA response was visualized in *35Spro::JAZ1-GUS* seedlings treated with salt. These results are in agreement with the observed up-regulation of transcripts for nine *JAZ* genes encoding for JA signaling regulators and five JA biosynthetic pathway genes in roots under salt stress. Together, these results show that salt stress triggers JA signaling pathway activation in *Arabidopsis* roots. Moreover, the described reporter systems are complementary and useful to visualize the status of JA response in a spatio-temporal fashion in transgenic roots.

Acknowledgements: FONDECYT N° 1120086.

PS17

THE GRAPEVINE GENES *VVFSK* AND *VVFTK* CODES FOR PUTATIVE RECEPTOR KINASES INVOLVED IN THE REPRODUCTIVE DEVELOPMENT

Sebastián González, Simón Ruiz-Lara, **Enrique González**

egonzale@utalca.cl

Instituto de Biología Vegetal y Biotecnología, Universidad de Talca

Vitis vinífera cv. Carménère has high tendency to develop parthenocarpic fruits yielding seeded and seedless berries in the same cluster. Even though the origin of this condition has not been fully elucidated, parthenocarpy has been associated to deficiencies in pollen germination capability which uncouples pollination and fertilization events. As a first approach to the study of the molecular/genetic events involved in this problem, expression profiles of approximately 4,800 genes were analyzed in both, seeded and seedless berries. Since a complex network involving several receptor kinase proteins are involved in the regulation of the fertilization event, we focused on the EST VVCCGS2117F10.b, which is highly repressed in seedless berries and shows homology to *ScORK17* gene from *Solanum chacoense* which codes for a receptor kinase involved in the regulation of ovule development. When the EST sequence was screened against the grapevine genome, two genes, *VvFSK* and *VvFTK*, were identified. In silico structural analysis revealed that both encoded proteins correspond to putative receptor kinases. Although both genes are transcribed in flowers, *VvFSK* is strongly expressed in seeds of berries at pre-veraison stage, while *VvFTK* is actively transcribed in tendrils. The *in silico* analysis of their promoter regions revealed the presence of cis regulatory sequences involved in the response to plant hormones and other elicitor molecules as sucrose induction as well as to phloem-specific expression. In agreement with their promoter structure, both genes are induced by sucrose and ABA. The function of *VvFSK* and *VvFTK* in grapevine reproductive development remains unknown, additional experiments are being conducted to assess such role.

Acknowledgements: Fondef G071003

PS18

**IDENTIFICATION AND CHARACTERIZATION OF GRAPEVINE BOR-TYPE
TRANSPORTERS ENCODING GENES**

Germán Saavedra, Rosa Roa, Mónica Yáñez, Simón Ruiz-Lara, Enrique González

gsaavedra@utalca.cl

Instituto de Biología Vegetal y Biotecnología, Universidad de Talca

Due to its role in the rhamnogalacturonan II structure, a cell wall polysaccharide, boron (B) is an essential micronutrient for vegetative and reproductive development in plants. A narrow range of B concentrations is favorable for plant growth. Outside this range, boron could be either toxic or can trigger deficiency symptoms. Transporters belonging to the BOR family are essential for maintaining boron homeostasis. In *Arabidopsis thaliana* grown under B limiting condition, this microelement is incorporated from the soil as boride by the AtNIP5;1 uptake transporter and then relocated to aerial tissues through xylem loading by the efflux transporter AtBOR1. On the other hand, when plants are exposed to toxic B concentrations, tolerance is mediated by boron exclusion due to AtBOR4 activity. Six putative BOR-type transporters encoding genes have been identified in the grapevine genome. A phylogenetical analysis of predicted protein sequences shows that three of them were grouped in the same clade with AtBOR1 while the other three were grouped together with AtBOR4. Four grapevine genes (*VvBOR1*, *VvBOR2*, *VvBOR4.4* and *VvBOR4.9*) were further analyzed. Their transcriptional pattern in different tissues and in response to exogenous treatments was assessed by qRT-PCR. A differential expression profile was determined for these genes in both, vegetative and reproductive tissues at different developmental stages, as well as in response to exogenous treatments with hormones, elicitor compounds and sodium borate, suggesting different roles for the encoded transporters in B assimilation and distribution in grapevine plants.

Acknowledgements: Fondecyt N° 1120871

PS19

**CHARACTERIZATION OF PSY1A AND PSY1B LOCI IN A POPULATION OF DURUM
WHEAT (*TRITICUM TURGIDUM* L. VAR. *DURUM*) WITH HIGH GENOTYPIC
VARIATION FOR SEMOLINA YELLOWNESS**

Albert Schulthess¹, Karen Campos¹, Nicolás Jiménez¹, Ivan Matus², Andrés R. Schwember¹

awschult@uc.cl

¹Pontificia Universidad Católica de Chile. Facultad de Agronomía e Ingeniería Forestal, Santiago, Chile; ²Instituto de Investigaciones Agropecuarias, Centro Regional de Investigación Quilamapu, Chillán, Chile.

Semolina yellowness of durum wheat (*Triticum turgidum* L. var. *durum*) is an important quality trait highly associated to the organoleptic quality of pasta. The main objective of this study was to characterize *PSY1A* and *PSY1B*, candidate genes for semolina yellowness, in one population of durum wheat comprising 100 genotypes. Regardless of the determination method for semolina yellowness, the phenotypic data of the population grown in two locations showed high variability due to the strong genotypic effect controlling this trait. Most genotypes of the population studied carried the *PSY1A*I and *PSY1B*a alleles, associated in past studies to intermediate and low levels of grain yellow pigment content, respectively. The low rates of polymorphism for these loci precluded the search of marker-trait associations but also confirmed the complex inheritance of semolina yellowness mentioned in past studies.

Acknowledgements: This work was financially supported by Conicyt (grant Fondecyt N°11110066, 2011-2014) and INIA-Quilamapu.

PS20

**GENES RELATED WITH SOLUBLE SUGARS ACCUMULATION IN TABLE GRAPE
(*VITIS VINIFERA* L.) BERRIES ARE RELATED TO BERRY SIZE**

Romina Silva¹, Pablo Laiz¹, Maribel Mamani^{1,2}, Denisse Laborie¹, José Correa^{1,2},
Manuel Pinto¹, Patricio Hinrichsen¹

phinrichsen@inia.cl

¹INIA La Platina, Santiago, Chile; ²Universidad de Chile, Santiago, Chile.

In table grapes, as in many others fruit crops, efficiency and coordinated transport of photosynthetes are essential processes for plant growth and also for their development and productivity. In this context sugar content (monosaccharides derived from sucrose) is an important quality factor and could be related to berry size. We selected three genes involved in sugar metabolism in grape berry, *VvGIN1* (*Vitis vinifera* vacuolar invertase 1), *VvTMT1* (*V. vinifera* tonoplast monosaccharide transporter1) and *VvSK1* (*V. vinifera* sugar inducible protein kinase), in order to test this hypothesis. The behaviour of these genes was assessed by qPCR in phenotypes contrasting for berry size and sugar content in four developmental stages (6-8 mm, pre-veraison, post-veraison and maturity). Transcripts for each gene were expressed in all developmental stages analyzed. Expression of *VvGIN1* was higher in early stages, which precedes massive sugar accumulation and only in these stages phenotype contrasting for berry size showed differential expression. *VvSK1* and *VvTMT1* were expressed mostly in more advanced developmental stages, after veraison. *VvSK1* was differentially expressed in all stages, with larger expression levels in genotypes harboring smaller berries; *VvTMT1* showed a similar behaviour, but only in advanced developmental stages. None of these genes showed a correlation between expression level and sugar content. These results suggest that these genes involved in sugar metabolism could also be related to berry size determination.

Acknowledgement: Programa Genoma-FONDEF grants G07I-1002 and G09I-1007, and FONDECYT N° 1120888.

PS21

THE TRANSCRIPTION FACTORS WRKY7, 11 AND 17 HAVE A ROLE IN THE TRANSCRIPTIONAL REPRESSION OF UPR-RESPONDING GENES DURING THE ESTABLISHMENT OF PLANT IMMUNITY IN *ARABIDOPSIS THALIANA*

Paulina Arraño, Begoña Galilea, Adrián Moreno, Francisca Blanco.

p.arrano@uandresbello.edu

¹FONDAP Center for Genome Regulation, Núcleo Milenio en Genómica Funcional de Plantas. Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello, Santiago, Chile.

The endoplasmic reticulum is the organelle where one-third of proteins are synthesized and folded in their final conformation. An increase in the number of proteins that will be synthesized generates a decrease in the capacity of folding, triggering of the Unfolded Protein Response (UPR). The pathogen attack is an event that triggers the UPR, a pathway that is Salicylic Acid-dependent and leads to the establishment of plant immunity. A fine tuning of the UPR-responding genes is necessary for mitigate the damage and to avoid the programmed cell death generated in this response. Recently, it has been described the role of WRKY family of transcription factors in biotic stress, specifically WRKY7, -11 and -17 that negatively controls the UPR-responding genes and plant immunity. To evaluate the role of those factors in the regulation of UPR-responding genes during the defense response, we analyzed the expression of those genes by q-PCR in wild type and WRKY-mutant plants. We compared the kinetics of expression after treatments with Salicylic Acid (SA), or after the challenge with *Pst* DC3000. We observed a positive regulation of the UPR-responding genes in the mutant plants suggesting that those factors negatively regulate the expression of UPR-responding genes. To get insights about the mechanism that regulate the function of these WRKY TF we analyzed the *in vivo* interaction between the factors WRKY7, -11 and -17 with CaM by experiments of Bimolecular Fluorescence Complementation (BiFC) in leaves of tobacco plants, before and after treatments with SA since it has been reported the *in vitro* interaction of WRKY7 with Calmodulin (CaM). CaM acts as a primary Ca²⁺ sensor and an interesting scenario involving calcium as activator of these transcription factors during the establishment of plant immunity is strongly suggested.

Acknowledgements: Fondecyt 11121387 and Nucleo Milenio P-10-062-F.

PS22

**IS LIGNIFICATION RESPONSIBLE FOR HIGHER FRUIT FIRMNESS IN SUN-EXPOSED
APPLE TISSUES COMPARED TO UNEXPOSED ONES?**

Constanza Azócar^{1,2}, Carolina Torres³, Patricio Ramos⁴, María Alejandra Moya-León⁴

co.azocar@gmail.com

¹Escuela de Ingeniería en Biotecnología, Universidad Andrés Bello, Santiago, Chile;

²Centro de Pomáceas, Universidad de Talca; ³Facultad de Ciencias Agrarias - Universidad de Talca; ⁴Instituto de Biología Vegetal y Biotecnología, Universidad de Talca, Talca, Chile.

Sun-injury or sunburn is a physiological disorder that affects various fruit crops, vegetables and ornamental species causing great economic losses in semi-arid growing regions. This disorder is caused by high solar irradiance and elevated temperatures during fruit growing season. Although symptoms appear on the fruit surface (skin), sun-exposure has also profound effects on their internal quality. Sun-exposed fruit has higher soluble solids (sugars), mineral content and firmness. In order to test the hypothesis that lignification is responsible for the higher firmness of the sun-exposed side of the fruit, a series of experiment were conducted during 2012-2013 season. Fuji and Royal Gala apples at different growth stage were manually exposed to direct sunlight. Tissue sampling (peel) was carried out after 1, 3, 6, 14, and 21 days of exposure. Prior peel removal fruit was classified as healthy (non-sun-injured), with mild, moderate or severe sun-injury. Quantitative RT-PCR was performed to determine the expression of genes encoding for the enzymes of the phenylpropanoid pathway (*PAL*, *CHS*, *F3H*, *CAD*, *CCR* and *COMT1*). Lignin was analyzed by ultraviolet light spectroscopy and phloroglucinol staining. Lignin was observed and detected only after 29 days of exposure in both cvs. Moderate and severe sun-injured tissue showed the highest lignin content. *PAL* transcripts appeared as early as 1 day after exposure and steadily increased until 21 days after. The expression of *CHS*, *F3H*, *CAD*, *CCR* and *COMT1* varied upon sun-injury level in the fruit, and throughout the sampling period.

Acknowledgements: Fondecyt Regular N°1100013.

PS23

**ACIDITY IN CHERIMOYA (*ANNONA CHERIMOLA* MILL.): HOW DOES THE FRUIT
PREPARE THEIR MACHINERY DURING DEVELOPMENT TO INCREASE ORGANIC
ACIDS DURING RIPENING?**

María Sofia Zamudio¹, Luis Tejerina^{1,2}, Rosa Molina¹, Nilo Mejia¹, Pedro Undurraga³, Bruno G. Defilippi¹, Mauricio González-Agüero¹.

maugonzalez@inia.cl

¹Instituto de Investigaciones Agropecuarias, INIA-La Platina, Santiago, Chile;

²Universidad Andrés Bello, Santiago, Chile, ³Pontificia Universidad Católica de Valparaíso, Facultad de Agronomía, La Palma, Chile.

Acidity is an organoleptic characteristic that helps to determine quality and consumer acceptability for many fruits. Cherimoya is a subtropical fruit with a unique and delicate flavor characterized by an unusual acidity behavior, i.e. showing an increase of organic acid contents during ripening. This characteristic make it an excellent fruit model to study the influence of acidity on fruit flavor and further understanding fruit ripening processes. However, little is known about the pathways responsible for synthesis, regulation and 'abnormal' accumulation of organic acids during development and ripening in this fruit. We believe that this different acid behavior during ripening comes from differential composition of organic acids during development and a different balance between enzymatic activity and/or expression of genes encoding key enzymes involved in synthesis and degradation of these metabolites. In this work, we identified and characterized several key genes encoding putative proteins related to citric, malic and tartaric acids metabolism, such as: NADP-isocitrate dehydrogenase, NAD- and NADP-dependent malic enzymes, phosphoenolpyruvate carboxylase, aconitate hydratase 1 and 3, citrate synthase and cytosolic malate dehydrogenase, among others. Their expression profile was characterized by real-time PCR assays in several developmental stages from two cherimoya varieties ('Concha lisa' and 'Bronceada'), and correlated with accumulation of citric, malic and tartaric acids. As cherimoya development progressed, significant changes in transcript levels were observed for some of the analyzed genes. The significance of these changes during cherimola development and the integration of further global gene expression analysis will provide valuable and novel information about the importance and influence of acidity on overall flavor and fruit quality

Acknowledgements: Fondecyt N°1130630.

CHARACTERISATION OF GENES INVOLVED IN H₂S METABOLISM IN STRAWBERRY

Sebastian Molinett*, Yazmina Stappung, Raúl Herrera, María Alejandra Moya-León

smolinett@utalca.cl

Laboratorio Fisiología Vegetal y Genética Molecular, Instituto de Biología Vegetal
y Biotecnología, Universidad de Talca.

Hydrogen sulfide (H₂S) has traditionally been considered as a phytotoxin, however recent evidences show that H₂S plays several physiological roles in plants, such as seed germination, root organogenesis, biotic/abiotic stress tolerance, senescence of cut flowers, regulation of ripening, etc. Furthermore, it is now recognized that plants have enzymes which generate H₂S (cysteine desulfhydrase, β -cyanoalanine synthase) and remove it (O-acetylserine lyase). Despite this evidence, the physiological roles that H₂S plays in strawberry remain unclear. In addition, the genes involved in H₂S metabolism are still not annotated or characterised. In this work, the genes that encode the enzymes involved in H₂S metabolism were identified within the genome of *Fragaria vesca*, a model strawberry species, and then characterised. We used the translated nucleotide sequences of *Arabidopsis thaliana* genes for data mining in *F. vesca* genome, performing local alignments by blastx. Our results indicate that woodland strawberry genome have 3 putative genes that encode for cysteine desulfhydrase, 1 putative gene for β -cyanoalanine synthase and 2 putative genes for O-acetylserine lyase. Furthermore, we obtained their respective genomic locations, amino acid sequence of proteins, full-length mRNAs and promoter regions. We analysed the promoter sequences of these genes in *F. vesca*. Our results suggest the presence of several physiological and growth regulators responsive elements within their promoter regions, such as MYBs, MYCs, Agamous, ABREs, WRKYs, etc. Finally, the expression level of these genes during *Fragaria chiloensis* fruit development was determined. RT-qPCR analyses indicate that the genes involved in H₂S synthesis and degradation are strongly expressed at early stages of fruit development, decreasing after that. This suggests that H₂S metabolism is mainly active during early developmental stages in strawberry fruit.

Acknowledgments: ANILLO ACT 1110 project.

PS25

**COMPOUND ACCUMULATION AND TRANSCRIPTIONAL PROFILE OF FLAVONOID
PATHWAY IN FLESH AND SKIN TISSUES AT DIFFERENT FRUIT DEVELOPMENT
STAGES OF JAPANESE PLUM (*PRUNUS SALICINA*)**

Máximo González¹, Soledad Cabrera¹, Pilar Olea¹, Erika Salazar², Basilio Carrasco¹.

mpgonza2@uc.cl

¹Facultad de Agronomía e Ingeniería Forestal, Pontificia Universidad Católica de Chile.
Av. Vicuña Mackenna 4860, Macul, Santiago, Chile. ²Unidad de Recursos Genéticos,
CRI La Platina, Instituto de Investigaciones Agropecuarias. Av. Santa Rosa 11610,
La Pintana, Santiago, Chile.

Varieties of Japanese plum (*Prunus salicina*) are diverse in terms of skin and flesh phenotypes. Previous studies have described different patterns of flavonoids accumulation associated with fruit phenotypes. In order to identify possible points of regulation associated with the color character, skin and flesh tissues in four contrasting varieties and considering four stages of fruit development (from pit hardening to full color stage) were characterized. Accumulation pattern of total phenols, tannins and anthocyanins were analyzed. Moreover, partial sequences from structural genes of the flavonoid pathway were isolated and specific primers were designed for each gene. Subsequently, the transcriptional expression analysis done by qPCR allowed us the identification of expression patterns associated to different developmental stages and tissues for each of the analyzed varieties. The structural genes *CHS*, *CHI*, *F3H* and *DFR* showed consistent expression patterns as described previously, however, different expression patterns were observed in *ANS/LDOX* and *UGT* genes. These genes were expressed in the presence of anthocyanins and strongly repressed in absence of it on the tissues, opening the hypothesis of a new regulatory mechanism not described for the *Prunus* genera.

Acknowledgments: CONICYT scholarships "Doctorado en Chile 2009 (N° 21090118)", "Apoyo a la Tesis Doctoral 2011 (24110179)" and "Tesis en la Industria 2013 (N° 781211008)". Consorcio tecnológico de la Fruta.

PS26

**MS-RAPD APLICATION FOR IDENTIFYING DIFFERENTIALLY METHYLATED
REGIONS RELATED WITH HETEROBLASTY IN *EUCALYPTUS GLOBULUS***

Macarena Arellano, Paulina Rivas, Rodrigo Hasbún

macaarellano@udec.cl

Facultad de Ciencias Forestales y Centro de Biotecnología, Universidad de Concepción, Chile.

DNA methylation involves the addition of a methyl group to the 5 position of the cytosine pyrimidine ring. In plants these methylation occur in CpG, CpHpH, CpHpG genomic sites where H represents A, C or T. DNA methylation is an epigenetic modification (heritable changes in gene expression that will not accompanied by alterations in the DNA), which plays an essential role in modulating the development of plants. One process mainly controlled by epigenetic modifications and especially DNA methylation is the vegetative phase change (also called heteroblastic change). Heteroblasty corresponds to a sequence of progressive changes observed in the leaves of the plants during ontogeny. These changes are verifiable at the morphologic, physiologic and biochemical level. A model plant for the study of this change is *Eucalyptus globulus*, species in which it has significant ecological effects. The purpose of this work is the use of MS-RAPD technique to detect differentially methylated regions (DMRs) between juvenile and adult leaves. DMRs are genomic regions with different methylation status among multiple samples (tissues, cells, individuals or others) and are regarded as possible functional regions involved in gene transcriptional regulation. MS-RAPD involves the digestion of DNA with a restriction enzyme sensitive to methylation (MspI) and methylation insensitive (HpaII). Following digestion, PCR is performed using RAPD primers of 10 to 14 bp for detecting differential fragments corresponding to DMRs. We show the results obtained after applying and optimize the technique for the experimental system under study.

Acknowledgments: FONDECYT N°11110505.

PS27

QUALITATIVE DETERMINATION OF AUXIN AND FLAVONOIDS BY CHROMATOGRAPHIC METHODS (HPLC AND TLC) IN *EUCALYPTUS GLOBULUS*

Fabián Zúñiga^{1,2}, Lissette Figueroa², Julian C. Verdonk¹, Claudia Sandoval²,
Andrea Miyasaka Almeida¹

fabian0110@gmail.com

¹FONDAP Center for Genome Regulation, Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello, Santiago, Chile. ²Center for Bioinformatics and Integrative Biology, Facultad de Ciencias Biológicas, Universidad Andrés Bello, Santiago, Chile

Eucalyptus is the second most widely planted multipurpose woody tree, and the most interesting one from a forestry perspective. *Eucalyptus globulus* wood has short fibers, which makes it very suitable for pulp production and the manufacturing of paper. In Chile, they can be found mainly in the central-south of the Bío-Bío region. For industry purposes, the main way to grow trees is by planting cuttings of several families of clones. However, many eucalyptus varieties have difficulties for rooting when propagated in this manner. This can eventually lead to a lower production of timber. In higher plants, a proper balance of the plant hormones auxin and cytokinin is important for root differentiation. Auxin is synthesized from tryptophan, an aromatic amino acid (AAA), which is synthesized from precursors from the shikimate pathway. Flavonoid biosynthesis also uses these precursors, and wounding of tissues as a result of the cutting process can cause an increase in the levels of flavonoids. We hypothesize that this might interfere with rooting of the cuttings. To test the hypothesis, flavonoids were extracted from different sections of *Eucalyptus globulus* seedlings and analyzed by thin layer chromatography (TLC). Flavonoids were extracted from leaves, stem and roots of *Eucalyptus globulus* seedlings in normal and stress conditions, and separated by TLC using a solution of chloroform, methanol and formic acid in a ratio of 90:9:1. Flavonoid contents in both leaves and shoots seem to be the same in stressed and non-stressed seedlings. However the roots from stressed seedlings showed at least double accumulation of flavonoids. In conclusion, we show that flavonoid production is increased in response to stress. The accumulation of flavonoids in roots could be the inhibiting factor for root growth through inhibition of auxin transport.

Acknowledgements: FONDAP CRG 15070009; Núcleo Milenio P10-062-F, Basal PFB-16 and Proyecto Anillo Científico ACT1107.

**SUGAR TRANSPORTERS: GENE EXPRESSION RESPONSES TO DIFFERENT
WATER STRESS LEVELS IN LEAVES AND GRAPES OF *VITIS VINIFERA* CV.
CARMENERE DURING RIPENING**

Luis Villalobos, Nicolás Ríos, Nicolás Franck, Claudio Pastenes

cpastene@uchile.cl

Facultad de Ciencias Agronómicas, Universidad de Chile.

In grapevines, water stress promotes a positive impact on berry sugar and phenolics, increasing fruit quality for winemaking. Nevertheless, the combined effect of drought, high temperature and high evaporative demand during summer could limit carbon distribution from source to sink tissues. Also, sugars are nutrients and signaling molecules distributed through the plant via sugar transporters, which are crucial for carbon allocation. The aim of this study was to determine the effects of different water stress levels on sugar transporters in leaves and grapes of Carménère vines during ripening. Gene expression levels of sucrose transporter (*VvSUC11*, *VvSUC12* and *VvSUC27*) and hexose transporters (*VvHT1*, *VvHT3* and *VvHT5*) were assayed by qPCR. Grapevines were exposed to 4 different irrigation regimes reaching an average midday stem water potential of -1.1MPa (T1), -1.0MPa (T2), -0.9MPa (T3) and -0.83MPa (T4), along the season. In leaves, T1 and T2 showed higher expression levels of *VvSUC11* and *VvSUC12* than T3 and T4 at 20 DAV. Such effects could be a response to the need for efficient sugar loading to the phloem, upon lower assimilates availability, due to the reduced assimilation rates in leaves of plants with higher water stress levels, concomitant to the increasing demand for sugar unloading at the berry sites. As for grapes, *VvSUC27* -a low affinity, high capacity transporter-, *VvHT1* and *VvHT3* reached higher expression levels at 5 days after veraison (DAV) decreasing later throughout the season. These transporters seem to be closely related to the increase of sugar unloading early after veraison. Interestingly, expression levels were increased in T2, the treatment with the higher sugar content at that time. Nevertheless, *VvHT1*, *VvHT3*, *VvHT5*, *VvSUC11* and *VvSUC12* transcripts, were increased in T3 and T4 at 20 DAV, treatments with highest sugar content through the season.

Acknowledgements: FONDECYT N° 1110193.

PS29

APPLYING CISGENESIS TO ENHANCE *VENTURIA INAEQUALIS* RESISTANCE IN APPLE

Javier Chilian, Karen Lisboa, Viviana Becerra, Mario Paredes

jchilian@inia.cl

INIA Quilamapu, Chillán

Apple scab caused by the ascomycete *Venturia inaequalis*, is the most important disease in all apple-growing regions. To overcome this disease, fruits growers may spray fungicides up to 20 times per season, which increase the cost of apple production and the concerns from consumers and environmentalists over pesticides uses. An alternative approach is the development of resistant varieties through classical breeding, but the introgression of traits from wild germplasm into elite cultivars result in a very slow and inefficient process. However, currently it has been proposed another method named cisgenesis as a faster alternative. Cisgenesis is different from transgenesis because is based in the introduction of isolated genes in their natural configuration from crossable or sexually compatible species and the use of the marker-free technologies. The pMF1 vector can be used to obtain marker-free plants by recombinase-based excision of a fragment carrying undesired gene sequences, such as antibiotic-selection marker genes. The aim of this work was to use cisgenesis to improve the *Venturia* tolerance to apple cv. Gala. The *Vf2* gene was isolated from Prima cv. and cloned into the binary vector pMF1. At this moment, apple cv. Gala is being transformed using *Agrobacterium* infiltration. This is the first research work in Chile using cisgenesis to improve apple cultivars.

Acknowledgements: INNOVA BIO BIO and CONICYT PSD-56

PS30

OPTIMIZATION OF THE TOTAL POLYPHENOL EXTRACTION AND QUANTIFICATION OF ANTIOXIDANT ACTIVITY IN OAT GENOTYPES

Paulina Torres^{1,2}, Francisca Acevedo^{1,2}, Eduardo Morales^{1,2}, Monica Rubilar^{1,2},
Monica Mathias³, Haroldo Salvo-Garrido^{1,3}

p.torres01@ufromail.cl

¹Agriaquaculture Nutritional Genomic Center (CGNA), Technology and Processes Unit.
Department of Chemical Engineering, UFRO, Temuco, Chile; ²Scientific and Technological
Bioresource Nucleus, BIOREN – Universidad de La Frontera; Temuco, Chile.

³Agricultural Research Institute (INIA). Camino Cajón Vilcún, Chile

Oat (*Avena sativa* L.) is a rich source of bioactive phytochemicals. Among these components, the polyphenols present a high antioxidant activity and help to reduce the risk of cardiovascular disease and obesity. The established health-beneficial properties of oats have led to an increase in their consumption in recent years. The objective of the present study was to find the optimum extraction conditions for the highest total phenolic content using response surface methodology (MSR), and to determine the polyphenol content and total antioxidant capacity of ten oat genotypes. For this purpose, the effect of the three variables: water/ethanol ratio (X_1), solid/solvent ratio (X_2) and extraction temperature (X_3) on the extraction process of total polyphenols was investigated. The total polyphenol content was determined by Folin-Ciocalteu method, using gallic acid as standard. Data and statistical analysis was carried out using Design-Expert® 6.0.6 package. The optimal conditions ($p < 0.05$) for the higher total polyphenol content were: water/ethanol ratio of 70/30, solid/solvent ratio of 1:6 and extraction temperature of 45°C. Based on these results, the total polyphenols content (TPC) and antioxidant activity (AOA) of ten oat genotypes were determined. The highest TPC was 66.39 ± 4.74 mg equivalents of gallic acid/g oat for genotypes AVE13 and the greatest AOA was 4.75 ± 0.45 mg equivalents of Trolox/g oat for the genotype AVE14. In this study, the best conditions for the total polyphenol extraction process were determined, which is a necessary tool for the optimal quantification of these bioactive compounds in oats. These results will contribute in the genetic improvement of oat to generate healthier foods.

Acknowledgments: INNOVA project 12IDL2-13628.

PS31

**DETERMINATION OF DNA METHYLATION KINETIC IN THE PROMOTER REGION
OF DORMANCY ASSOCIATED MADS-BOX GENE (*DAM6*) IN SWEET CHERRY
(*PRUNUS AVIUM*) DURING BUD DORMANCY RELEASE**

Karin Rothkegel^{1,2}, Soraya Bravo³, Humberto Prieto⁴ and Andréa Miyasaka Almeida^{1,2}.

k.rothkegel@uandresbello.edu

¹Centro de Biotecnología Vegetal, Universidad Andrés Bello. República 217, Santiago, Chile.

²FONDAP Center for Genome Regulation, Santiago, Chile. ³Centro de Biotecnología Gran Concepción, Facultad de Ciencias Biológicas Universidad Andrés Bello. Concepción, Chile.

⁴Instituto de Investigaciones Agropecuarias, INIA La Platina, Santiago, Chile.

The induction of bud dormancy release at the correct moment is important in perennial trees to survive low temperatures and complete reproduction cycle in temperate climates. DORMANCY ASSOCIATED MADS-BOX (*DAM*) genes are negative flowering regulators and strong candidates for the regulation of bud formation and growth cessation in peach. Specifically, *DAM5* and *DAM6* are winter expressed and suppressed by chilling temperatures, suggesting a role in regulating endodormancy and being regulated by chilling accumulation. *DAM6* also showed to be regulated at the chromatin level by histone modifications in a similar way to its orthologous in *Arabidopsis* during vernalization, proposing a similar regulation in dormancy. To better understand this mechanism in cherry (*Prunus avium* var. Bing), we isolated the promoter region of *DAM6* and analyzed its methylation pattern through bisulfite sequencing in floral buds sampled weekly during the dormancy period. Bisulfite sequencing refers to the treatment of DNA with sodium bisulfite to convert unmethylated cytosine to uracil, followed by sequencing to analyze cytosine methylation. We obtained the methylation pattern of a 207 bp region located at approximately 400 pb upstream of the ATG site in sweet cherry. These results suggest a dynamic change and an increase of methylated cytosines at this genomic region, as the chilling requirement (CR) is being fulfilled. In addition, we observed the possible presence, through bioinformatic analysis, of a nucleosome. This could suggest a possible relationship between cytosine methylation and gene regulation during dormancy. However, to better understand how this process affects dormancy and flowering in economically important fruit trees, it is necessary to perform more studies of epigenetic mechanisms in the *DAM* genes, as histone modifications and non-coding regulatory RNAs analysis.

Acknowledgements: FONDEF G09I1008; FONDAP CRG 15070009 and Basal PFB-16.

PS32

**CHROMATIN IMMUNOPRECIPITATION ASSAY (CHIP) IN FRESH SAMPLES OF BUDS
FROM SWEET CHERRY (*PRUNUS AVIUM* L.) AND PEACH (*PRUNUS PERSICA* L.)**

Sebastián Tapia¹, Humberto Prieto¹, Andréa Miyasaka Almeida^{2,3}

setapiag@gmail.com

¹Instituto de Investigaciones Agropecuarias, INIA La Platina, Santiago, Chile.

²Centro de Biotecnología Vegetal, Universidad Andrés Bello, Santiago, Chile.

³FONDAP Center for Genome Regulation, Santiago, Chile.

Molecular knowledge of important physiological processes for perenne plants, like blooming and dormancy is fundamental for fruit development. There is evidence that the DAM (DORMANCY ASSOCIATED MADS-BOX) gene family is involved in the process of dormancy in peach (*Prunus persica* L.). This family is comprised of 6 genes in tandem that are potential candidates for maintaining the bud in dormant state, while the environmental conditions are unfavorable for plant growth. When a winter passes, the bud accumulate chilling hours and its DNA at DAM loci suffer modifications in the chromatin level, which could allow a differential expression of these genes. The degree of chromatin compaction depends on post-translational modification of histones such as methylation or acetylation. These modifications alter the interaction between the histone proteins and DNA, affecting DNA packing. The chromatin immunoprecipitation assay (ChIP) is a powerful tool that allows the analysis of the chromatin structure. In this work we standardize a protocol for ChIP assay in floral buds of sweet cherry (var. Bing) and peach (var. Magique) sampled during the period of dormancy, with the aim of studying the post-translational modifications of histones that occur in the promoter region of the DAM5 and DAM6 genes.

Acknowledgements: FONDEF G09I1008; FONDAP CRG 15070009 and Basal PFB-16.

PS33

**RELATIVE EXPRESSION OF GENES RELATED WITH COLD TOLERANCE AT THE SEEDLING
STAGE IN TEMPERATE RICE**

Gabriel Donoso¹, María Leyton², Mario Paredes¹ and Viviana Becerra¹

gabriel.donoso@inia.cl

¹Centro de Biotecnología de los Alimentos (CBA), Centro Regional de Investigación Quilamapu, Instituto de Investigaciones Agropecuarias (INIA), Chillán, Chile. ²Facultad de Agronomía, Universidad de Concepción, Campus Chillán, Chile.

Low temperature is one of the main abiotic stresses affecting rice yield in Chile. Alterations in phenology and physiology of the crop are observed after cold event. By this means, the objective of this work was to study the relative expression of genes related with oxidative stress in Chilean varieties of rice. For this, we analyzed cold tolerance of Diamante-INIA and Zafiro-INIA and one experimental line from INIA Chile, Quila 241701, with high cold tolerance. For analysis, the Spanish variety, Susan, was used as check tolerance and Oryzica 1 as susceptible check. Oxidative stress was measured through Lipid peroxidation. To find mechanisms of cold tolerance in Chilean varieties, we determine the relative expression of genes related with oxidative stress, like superoxide dismutase (SOD), glutathione reductase (GR), and catalase (CAT). Lipid peroxidation allowed differentiating the physiological stress level of the genotypes under study, finding that in Oryzica 1 was higher than genotypes Diamante-INIA, Zafiro-INIA, Quila 241701 and Susan. Also, significant increases in relative expression of the gene encoding CAT in Oryzica 1 were observed.

Acknowledgements: FONDECYT 1110405

PS34

**STUDY OF COLD TOLERANCE IN TEMPERATE RICE AT GERMINATION STAGE
(*ORYZA SATIVA* L.) USING PROTEOMIC TOOLS**

José González², Uri Aceituno¹, Mario Paredes¹, Viviana Becerra¹, Jesús Jorrín³, **Gabriel Donoso¹**

gabriel.donoso@inia.cl

¹Centro de Biotecnología de los Alimentos (CBA), Centro Regional de Investigación Quilamapu, Instituto de Investigaciones Agropecuarias (INIA), Chillán, Chile.

²Facultad de Agronomía, Universidad de Concepción, Campus Chillán, Chile. ³Department of Biochemistry and Molecular Biology, University of Córdoba, Córdoba, Spain.

Low temperatures at germination stages are the principal causes of low emergence of coleoptile after sowing. Rice Breeding Program from INIA Chile has varieties with high adaptation to cold stress at germination stage. However the mechanisms of cold tolerance in these varieties are still unknown. Therefore, the aim of this study is to find proteins related to cold tolerance at germination stage in Chilean varieties. For this, embryo proteins from Chilean rice varieties, Diamante-INIA and Zafiro-INIA, and the cold susceptible variety Oryzica 1, were extracted using TCA/Phenol protocol. Two-dimensional gel electrophoresis of protein extracts were made with isoelectrofocusing in the 5-8 pH range and SDS-PAGE with 12% polyacrylamide gels. Gels were stained with Coomassie Blue and scanned using Image Scanner III (GE Healthcare). The analysis was performed with PDquest 2-D analysis software (BIO-RAD). Non-difference in protein yield between varieties was observed. Quantitative and qualitative differences between Chilean varieties and Oryzica 1 were observed. Proteins related to these differences will be related to cold tolerance at germination stage in Chilean rice varieties.

Acknowledgements: FONDECYT 1110405

PS35

PHYLOGENETIC AND TRANSCRIPTIONAL ANALYSIS OF NIPS (NODULIN 26-LIKE INTRINSIC PROTEINS) GENES IN *PRUNUS* ROOTSTOCKS UNDER HYPOXIA STRESS.

Francisco Correa¹, Ariel Salvatierra¹, Rubén Almada¹, Manuel Pinto²,
Boris Sagredo¹, Paula Pimentel¹

mike_pancho@hotmail.com

¹Centro de Estudios Avanzados en Fruticultura (CEAF_R08I1001), INIA CRI Rayentué, Rengo, Chile. ²INIA CRI La Platina, Santiago, Chile.

Under field condition hypoxia radical events occur due to problems of flooding, soil compaction, heavy soils with poor drainage or irrigation problems. The stone fruit trees (*Prunus sp.* L.) are grafted onto rootstocks, which determine the tolerance of the varieties to various stresses, e.g. root hypoxia. Although most *Prunus* rootstocks have been classified as sensitive to hypoxia, different responses have been found between genotypes. Aquaporins are structural membrane proteins that have been characterized as carriers of water and/or solutes and involved in the response of plants to various stresses. NIPs (Nodulin 26-like intrinsic proteins) represent a large, diverse family of aquaglyceroporins, with multiple members found in every sequenced plant genome. Functional analyses indicate that Nod26 is an aquaglyceroporin with low intrinsic water permeability and the ability to transport uncharged metabolites. In *Arabidopsis thaliana* under oxygen deprivation, a marked induction of *NIP2;1* gene expression has been reported. Based on structure–function studies, NIP aquaporin family can be subdivided into three subgroups (I, II and III) regarding the identity of the amino acids in the selectivity-determining filter (ar/R region) of the transport pore. Nine members of NIP have been reported in the genome of *Prunus persica* and bioinformatic analysis classified them as three genes of NIP I, five of the NIP II, and one of the NIP III. Gene expression of NIPs members were evaluated in two contrasting genotypes to root hypoxia, ‘Mazzard F12/1’ (sensitive) and ‘Mariana 2624’ (tolerant), under 15 days of hypoxia treatment. qPCR analysis revealed different expression pattern under hypoxia treatment. These results suggest that NIPs aquaporin expression is part of the mechanism to cope the root hypoxia stress in *Prunus*.

Acknowledgments: FONDECYT N°11110080, CEAF_R08I1001. Plants were provided by Agromillora Sur S.A.

PS36

**MUTATION OF ACETOHYDROXY ACID SYNTHASE (AHAS) GENE ENDOWING
NICOSULPHURON RESISTANCE IN SORGHUM HALEPENSE**

Rocío León , María José Hernández, Rodrigo Figueroa, Marlene Gebauer

rleonn@um.uchile.cl

Departamento de Ciencias Vegetales, Facultad de Agronomía e Ingeniería Forestal, Pontificia
Universidad Católica de Chile.

One of the mechanisms responsible for AHAS inhibitors resistance in plants is the presence of specific mutations in the AHAS gene producing a conformational change in the target site and preventing AHAS coupling to this herbicide. In *Sorghum halepense* (Johnsongrass) a polyploid species present in the Chilean cornfields, resistance to AHAS-inhibiting herbicides has been detected, specifically to sulphonylurea (SU) and imidazolinone (IMI). To elucidate the mechanism of nicosulphuron resistance in *S. halepense* biotypes, one population of the Metropolitan Region and two of the Region VI were analyzed. Resistant (R) and susceptible (S) biotypes were identified by whole-plant dose response assays. *In vitro* enzyme activity assays were performed and gene regions where mutations endowing herbicide-resistance have previously been found were amplified using six biotypes per population. Resistance levels detected by AHAS activity assays and mutations found in the AHAS gene, generating amino acid substitution Trp574-Leu suggest a resistance mechanism mediated by target site.

Acknowledgments: CONICYT, FONDECYT N°110535.

PS37

TONOPLAST INTRINSIC PROTEINS (TIPs) SUBFAMILY IN *PRUNUS* SP: ABIOTIC AND HORMONAL REGULATION

Paula Pimentel¹, Ariel Salvatierra¹, Rubén Almada¹, Manuel Pinto^{1,2}, Boris Sagredo¹

ppimentel@inia.cl

¹Centro de Estudios Avanzados en Fruticultura (CEAF), INIA CRI Rayentué, Rengo, Chile.

²INIA CRI La Platina, Santiago, Chile.

Among the problems that restrict the cultivation of species of the genus *Prunus*, are those associated with poor aeration of rhizosphere in clay and/or compacted soils and incorrect irrigation practices. In such soils, heavy rain or excessive irrigation cause a waterlogged effect. In fruit trees, the tolerance to oxygen deficiency in roots is mediated by the characteristics of the rootstock. The *Prunus* rootstocks are classified as anoxia-sensitive, although differences among genotypes have been reported. Tonoplast intrinsic protein (TIP) is a subfamily of the aquaporins (AQPs), also known as major intrinsic protein (MIP) family, which regulate water movement across vacuolar membranes. Aquaporins play a key role in root water uptake capacity, which would provide a molecular basis for fast and reversible regulation of transmembrane water transport. TIPs, being localized at the tonoplast, facilitate rapid osmotic equilibration between the vacuole and cytosol. Some reports have implied that *TIP* genes are associated with plant tolerance to some abiotic stresses that cause water loss, such as drought and high salinity. We identified 8 members of TIP subfamily in peach genome. The accumulation of *TIPs* transcripts in different vegetative tissues (roots, leaves, stem and flower) and the effects of hypoxia, salt, water stresses and abscisic acid on the expression of *TIPs* genes were investigated. *TIPs* gene showed different expression pattern in tissues and only *TIP2;3* showed to be root-specific. Many *TIPs* gene were markedly up or down regulated during different stress and differentially hormone-regulated. These results provide new information about the relationship between TIP and plant abiotic stress tolerance in *Prunus* species.

Acknowledgments: FONDECYT 11110080, CEAF_R08I1001 and CONICYT (Proj. N° 79095006 R.A. and P.P). Plants were provided by Agromillora Sur S.A.

PS38

**DIFFERENTIAL EXPRESSION OF SOS (SALT OVERLY SENSITIVE) AND NHXS
GENES REGULATED BY NA⁺ IN *DESCHAMPSIA ANTARCTICA* DESV.**

Daisy Tapia, León Bravo and Ana Gutiérrez.

d.tapia02@ufromail.cl

Doctorado en Ciencias mención Biología Celular y Molecular aplicada. Laboratorio
de Fisiología y Biología Molecular Vegetal. Facultad de Ciencias Agropecuarias y Forestales.
Scientific and Technological Bioresource Nucleus (BIOREN-UFRO).
Universidad de La Frontera, Temuco, Chile

Salinity is one of the most important abiotic stresses, which limit crop production in arid and semi-arid regions. Over 6% of the world's land is affected by salinity which accounts for more than 800 million ha of land. The ability to mobilize long-distance sodium (Na⁺) from the roots to older leaves is a mechanism present in salt-tolerant plants. The salt overly sensitive (SOS) pathway plays an important role in plant salt tolerance, controlling Na⁺ transport over long distances from the root to shoot. SOS3, SOS2 and SOS1 proteins are involved in this pathway. Moreover, the transporters of the NHX family are widely regarded as key players in the sequestration of Na⁺ into vacuoles to avert ion toxicity in the cytosol of plants under salinity stress. *Deschampsia antarctica* DESV. (Poaceae) is one of the two unique angiosperms that inhabit the Antarctic region, which is one of the harshest ecosystems in the world. This plant survives under hostile environmental conditions, such as drought, flooding, variable temperatures and radiation accompanied by frost, frozen ground and sea spray. The objective of our study was to evaluate the expression profile (qRT-PCR) of SOS pathway and NHX genes in new leaves, old leaves and roots of *Deschampsia antarctica* under different NaCl treatments. We exposed *Deschampsia antarctica* plants to different NaCl concentrations (0.5 M 1.0 M) for 21 days. The results show differential expression of SOS and NHXs genes evaluated in different tissues of plants subjected to NaCl treatments. The results suggest that these mechanisms could be involved in the ability to tolerate high NaCl level in soil in *Deschampsia antarctica*.

Acknowledgements: CONICYT doctoral fellowship, code 24121490-2012. Doctorate Fellowship CONICYT Daisy Tapia.

PS39

**TRANSCRIPTIONAL PROFILES OF GLUTAMATE DECARBOXYLASE (GAD) GENE ISOFORMS
IN *PRUNUS* ROOTSTOCKS UNDER ROOT HYPOXIA STRESS BY FLOODING.**

Ariel Salvatierra¹, Paula Pimentel¹, Manuel Pinto^{1,3}, Boris Sagredo^{1,2}, Patricio Hinrichsen^{1,3}.

asalvatierra@ceaf.cl

¹Centro de Estudios Avanzados en Fruticultura (CEAF), Rengo, Chile;

²INIA CRI Rayentué, Rengo, ³INIA CRI La Platina, Santiago, Chile. .

GABA shunt pathway occurs in a wide range of organisms, such as bacteria, yeasts, plants and animals. The first step of this pathway consists in the decarboxylation of the amino acid glutamate into gamma amino butyric acid (GABA) by glutamate decarboxylase (GAD) enzyme. In plants, GABA level is normally low, but it quick and extensively increases in response to stress (e.g., hypoxia). In our work, GABA metabolism was evaluated in two *Prunus* rootstock genotypes with contrasting responses under root hypoxia. An *in silico* search for orthologous genes of GABA shunt in *Prunus persica* genome revealed four *GAD* isoforms. These gene isoforms showed an expression organ-associated. *GAD4* was identified as a root-specific isoform, but its expression was not induced under root hypoxia condition in the tolerant genotype Mariana M2624. The rest of *GADs* showed an increase in their transcript levels triggered by hypoxia; however, the expression of *GAD2* and *GAD3* was induced rapid and transiently in Mariana M2624 and slow and constantly in Mazzard F12/1. Thus, not only different transcript levels were detected between these contrasting genotypes but, in addition, time-dependent transcriptional profiles were described for each genotype in response to root hypoxia by flooding.

Acknowledgements: AS wants to acknowledge to FONDECYT N° 31301384. PP acknowledges the support of FONDECYT N11110080. Plants were provided by Agromillora Sur S.A.

PS40

EXPRESION OF LIPID TRANSFER PROTEIN GENES (NSLTPS) DURING FRUIT DEVELOPMENT AND ACLIMATION TO DROUGHT STRESS IN SOLANUM SPP.

Carlos Morales^{1,2}, Gerardo Tapia¹, Cristián Balbontín²

cmoralesq@udec.cl

¹Unidad de Recursos Genéticos, Instituto de Investigaciones Agropecuarias, INIA-Quilamapu, Av. Vicente Mendez 515, Chillán. ²Facultad de Ciencias biológicas Universidad de Concepción, Victor Lamas 1290, Concepción.

The nsLTPs (non-specific Lipid Transfer Proteins) genes are proteins characterized by transport of lipids through membranes, which are widely spread in the plant kingdom. Several studies associate them with defense activity against fungi and bacterial pathogens and wall loosening of the plant cells. Also they have been linked with the cuticle development in vascular plants, a hydrophobic layer that covers the epidermis of aerial organs such as fruit, stem, leaves, and parts of the flowers. It works as protective barrier against mechanical damage, loss of water and pathogen invasion, among others. Many LTPs are directly related with plant stress response and some other functions associated with the cuticle such as the integrity of waxy skin layer of fruits. In this research we identified three *nsLTP* genes named *SILTP1*, *SILTP2* and *SILTP5*, belong to nsLTP families IV II, and I, respectively. In order to establish their putative role in cuticle formation of tomato fruit var. Money Maker, we analyzed their expressions, by qRT-PCR, at four stages of fruit development. The genes *SILTP1* and *SILTP5* were mainly induced during pink stage, which is one of the periods of cell expansion where the fruit increases its size. This finding supports the idea that these nsLTPs play a role during fruit development, specifically in cuticle formation during cell elongation. The expression of both *SILTP1* and *SILTP5* genes also was analyzed in the wild specie *Solanum peruvianum*. Both genes were overexpressed during hydric stress in vegetative organs of the plants. Nowadays, we are producing mutants, by Artificial microRNAs (amiRNAs) that have negative regulatory function on gene expression, for the three nsLTPs genes previously identified. Their characterization will help to unveil their role in both tomato fruit development and abiotic stress.

Acknowledgements: Fontagro FTG-8071/08; Fondecyt 11100149.

PS41

ALL MEMBERS OF THE *Brassica napus* PSY GENE FAMILY ARE FUNCTIONAL

Ada López-Emparán^{1,2}, Matías Zúñiga², María Laura Federico¹

ada.lopez@cgna.cl

¹Centro de Genómica Nutricional Agroacuícola (CGNA), Unidad de Genómica y Bioinformática, INIA, Temuco. ²Universidad de Talca, Programa de Doctorado en Cs. Mención Ingeniería Genética Vegetal, Talca.

Phytoene synthase (PSY) has been shown to catalyze the first committed step of carotenoid biosynthesis. PSY is encoded by a single copy gene in *Arabidopsis thaliana*, but most plant species contain a small gene family. In a previous study, we have demonstrated that six *PSY* genes have been retained in *Brassica napus* L. Most importantly, we have shown that all six members are expressed. In this context, the question of whether this large *PSY* family actually encodes six functional enzymes remained to be answered. Therefore, the objectives of this study were to: (i) isolate, characterize and compare the complete protein coding sequences (CDS) of the six *B. napus PSY* genes; (ii) model their predicted tridimensional enzyme structures and (iii) test their phytoene synthase activity in a heterologous complementation system. This study further confirmed that all six *B. napus PSY* genes encode proteins of similar length (414-424 aa) with high sequence identity (86,47% - 99,05%) and contain characteristic motifs present in phytoene synthases: a transit peptide (TP), a conserved trans-isoprenyl diphosphate domain (trans-IPP) and a putative phytoene synthase active site (DXXD) with 4 conserved aspartate residues. Protein modeling predicted that all proteins are alpha-helix rich structures. The two conserved aspartate-rich domains, DELVD and DVGED, localize to two alpha helices on opposite walls of the central cavity, conforming the characteristic phytoene synthase active site. Significantly, we have demonstrated that *E. coli* cells harboring pDS1B- Δ crtB and cotransformed with each of the six *B. napus PSY* genes were capable of producing β -carotene. Thus, we were able to confirm that the largest plant *PSY* gene family described to date encodes functional enzymes. This work provides a pool of functional *PSY* genes suitable for metabolic engineering of seed carotenoid content.

Acknowledgements: FONDECYT 1090726 and CONICYT REGIONAL/GORE ARAUCANÍA/CGNA/R10C1001.

PS42

EXTRACTION AND ANALYSIS OF RNA OF *CISTANTHE LONGISCAPA* FOR HIGH THROUGHPUT SEQUENCING TO IDENTIFY GENES INVOLVED IN THE PROCESS OF SEED GERMINATION.

Daniela Elizondo, Francisca Blanco, Claudio Meneses, Ariel Orellana.

dani.elizondo.munoz@gmail.com

FONDAP Center for Genome Regulation, Millennium Nucleus for Plant Functional Genomics, Plant Biotechnology Centre, School of Biological Sciences, University Andrés Bello.

The “Blooming of the desert” is a world renowned phenomenon that develops at the coast of northern Chile (26-31°S). This phenomenon occurs only when the seeds of ephemeral species have accumulated a minimum volume of water between May and August. The ephemeral species that bloom in the Atacama Desert are able to detect the favorable environmental conditions to germinate and complete their life cycles. It is essential to study genes that are involved in the germination of these seeds in the extreme conditions found in the Atacama Desert. This could be an important breakthrough because its potential application in the development of new crop varieties. *Cistanthe longiscapa* is one of the dominant species in the “Blooming of the desert”, and we use it as a model to get insight into the genes that are required for germination under drought stress. Thus we cultivate *C. longiscapa* *in vitro*. Then we optimized a protocol for RNA extraction to obtain high quality nucleic acids from seeds exposed to water for 1, 5 and 12 hours to be used in high-throughput sequencing. This will allow us to identify genes that are expressed in different stages during seed germination. Using the RNeasy mini kit (Qiagen), we isolated RNA of high quality confirmed by capilar electrophoresis. Moreover we synthesized cDNA and confirmed this genetic material amplifying constitutive and conserved genes from *Arabidopsis thaliana*.

Acknowledgements: Plant Biotechnology Centre, Dr. Reinaldo Campos for assistance in the collection of biological material and Dr. Claudio Meneses for help sequencing and bioinformatics analysis. Also thanks to funding provided by the projects FONDAP CRG 15070009, Fondecyt 1110954, Núcleo Milenio P10-062-F and Basal PFB-16.

PS43

**REGULATION OF ACCLIMATION/DEACCLIMATION PROCESS AND ITS RELATION WITH THE
BUD-BREAK RESPONSE OF *VITIS VINIFERA***

Sebastián Rubio, Francisco Pérez

frperez@uchile.cl

Laboratorio Bioquímica Vegetal, Facultad de Ciencias, Universidad de Chile.

The climatic change is a real problem for temperate fruit trees, mainly because of winter warm spells which lead to a loss of cold hardiness in buds, making them susceptible against early spring frosts. In Santiago of Chile during the last two fall-winter seasons (2012-13), cold hardiness (Hc), measured as low temperature exotherm (LTE), was monitored on buds of *V. vinifera* cv Thompson seedless using FRIOSCAN, a differential thermal analyzer (DTA). From these experiments, two results were pointed out, first, LTE follow the air temperature pattern, and second, buds remains cold-acclimated with a low LTE value beyond the end of the endodormancy (ED) period, and just few days before the onset of bud-break, the value of LTE increases. Therefore, the kinetic of bud de-acclimation becomes essential in the timing of bud sprouting. In the laboratory, using single-bud cuttings it was demonstrated that Hc is a reversible process that depends exclusively on air temperature. To begin to understand the relationship between acclimation/de-acclimation of grapevine buds with bud-sprouting at the molecular level, starch, soluble sugars and genes previously related to acclimation were studied on single-bud cuttings exposed to low and room temperature for 7, 14, 21 and 42 days. The results show that low temperatures induces starch degradation, sugar accumulation and the up-regulation of CBF4, a key transcription factor in cold acclimation.

Acknowledgements: Fondecyt 1110056

RNAi-INDUCED RESISTANCE AGAINST INSECTS IN RAPESEED

Peter Krause, Rod Snowdon , Wolfgang Friedt

peter.krause@agrar.uni-giessen.de

Department of Plant Breeding, Research Centre for Biosystems, Land Use and Nutrition (IFZ),
Justus-Liebig-University Giessen, Heinrich-Buff-Ring 26-32, 35392 Giessen

Rapeseed (*Brassica napus*) is the major oilseed plant in Europe and one of the most important crop plants in agriculture worldwide. Heavy yield loss can occur by different biotic and abiotic stresses. The pollen beetle (*Meligethes aeneus*), one of the most important insect pests in Europe, causes yield losses up to 30%. Frequent applications of insecticides often lead to resistances of pest insects. New technologies based on genetic engineering can help to improve plant resistances against pests and diseases. The particular focus of this work is based on RNA interference (RNAi), discovered in late 1990s by the Nobel Prize winner Fire and Mello in the nematode *C. elegans*. RNAi pathways are conserved mechanisms found in many eukaryotic cells including animals, plants and insects. RNAi is commonly used in reverse genetics applications to find putative target sequences, but it also can be used to improve plants in their tolerance to biotic and abiotic stresses. By means of RNAi, plant performance can be changed by knock-down of specific target genes. Potential lethal acting target genes of pollen beetle were identified by *in silico* research on lethal acting genes of *T. castaneum*. For quantification of the lethality, the mode of action and the lethal dose of the candidate genes, highly beetle specific sequences (300-400 bp) of dsRNAs were generated and tested in oral applications (*in vivo*). Subsequently, best candidates were selected cloned into specific RNAi plasmids containing cohesive double-promoter sequences. GM plants have been produced by means of *Agrobacterium*-mediated transformation. Molecular methods were used afterwards to determine and quantify the gene expression level. Selected T1 plants were used for proof of concept confirmation of *in vitro* feeding trials. The aim is to produce a preproduction model plant which shows a highly-specific resistance against pollen beetle while protection of beneficial organisms at the same time.

PS45

**MOLECULAR CHARACTERIZATION OF GROUP VII *ETHYLENE-RESPONSIVE*
TRANSCRIPTIONAL FACTORS (ERF)-LIKE GENES IN *PRUNUS* SPECIES WITH
DIFFERENT DEGREE OF TOLERANCE TO WATERLOGGING**

Rubén Almada¹, Paula Pimentel¹, Manuel Acuña¹, Pamela Rojas¹, Patricio Hinrichsen ^{1,2},
Manuel Pinto^{1,2}, Boris Sagredo¹

ralmada@ceaf.cl

¹Centro de Estudios Avanzados en Fruticultura (CEAF_R08I1001), INIA CRI Rayentué,
Rengo, Chile. ²INIA CRI La Platina, Santiago, Chile.

Root hypoxia produced by flooding or over-irrigation limits stone fruit tree development, particularly in orchards established on soils with restricted drainage. To overcome this problem, stone fruit trees are usually grafted on rootstocks (species or hybrid of the *Prunus* L. genus) with different degrees of tolerance to root hypoxia. However, the molecular base of such variability is largely unknown. In *Arabidopsis thaliana*, as well as in a number of crops species, group VII ERF-like genes stand out among hypoxia-related genes, but no such studies have been done with the *Prunus* species used as rootstocks. In this study, we analyzed the expression of three group VII ERF-like genes (*ERF1*, *ERF2* and *ERF3*) in roots and leaves of two stone fruit rootstocks with different degree of tolerance to root hypoxia (tolerant vs sensitive). When rootstock plants were exposed to waterlogging the expression of group VII ERF-like genes was induced in roots of the genotypes analyzed. However, group VII ERF-like showed different expression patterns in leaves of plants exposed to root hypoxia. On the other hand, our results show that the regulation of group VII ERFs is not exclusive to ethylene, but also includes the plant hormones ABA, NO, BAP, IAA and GA₃ along with other abiotic factors (such as low and high temperatures, drought and saline stresses). The analysis of promoter sequences of group VII ERF-like genes revealed the presence of cis-regulatory elements potentially involved in the control of their expression by hypoxia, abiotic stresses and hormones. Our results indicate that the hypoxia-mediated expression of group VII ERF-like genes would be tightly regulated by myriad of components pointing towards a crucial role in integrating and fine tuning flooding responses in the *Prunus* species studied.

Acknowledgments: CEAF_R08I1001 and FONDECYT N°11110079. Rootstock plants were gently provided by Agromillora Sur S.A.

PS46

FUNCTIONAL CHARACTERIZATION OF NUCLEOTIDE SUGAR TRANSPORTERS (NSTS) BY EXPLOITING THE ARABIDOPSIS SEED MUCILAGE.

Susana Saez-Aguayo, Daniela Doñas, Francisca Reyes and Ariel Orellana

saez.aguayo@gmail.com

Centro de Biotecnología Vegetal, Universidad Andrés Bello. Centro FONDAP de Regulación del Genoma.

After imbibition a mucilage capsule is formed around Arabidopsis seeds. Mucilage is a polysaccharide matrix, which is produced and accumulated in the epidermal cells during seed coat development. The Arabidopsis mucilage is formed of two structurally distinct layers: the water-soluble and the adherent layer. Both are composed mainly of the pectin rhamnogalacturonan 1 (RG1). The mucilage pectin biosynthesis takes place in the lumen of the Golgi apparatus and requires the participation of many glycosyltransferases, which uses nucleotide sugars as substrates. Nucleotide sugars are synthesized in the cytosol and they are transported into the Golgi apparatus by nucleotide sugar transporters (NSTs). Therefore, NSTs seems to be key elements in the biosynthesis of mucilage polysaccharides. In Arabidopsis, NSTs belongs to a gene family constituted by more than 50 members. In this project we use the seed coat mucilage as tool to identify novel nucleotide sugars transporters involved in the biosynthesis of pectin. *In silico* analyses shows that 20 NSTs exhibit high expression in the seed coat during the stages where mucilage is accumulated in the epidermal cells. Here we present 3 of these 20 NSTs mutants: *nst9*, *nst10* and *nst21*, which have a mucilage phenotype. Biochemical and immunological analysis of these 3 mutants reveals a lower content of soluble mucilage and changes in composition of the adherent layer. We have evidence that *nst10* transports UDP-arabinose, a precursor of arabinan chains, during the early stages of mucilage accumulation. This lack of arabinose seems to affect the mucilage release due to the stiffening of the epidermal cell wall.

Acknowledgments: FONDAP CRG 15070009; Núcleo Milenio P10-062-F, Basal PFB-16. Acknowledgements: Fondecyt 1110954, FONDAP CRG 15070009, Núcleo Milenio P10-062-F and Basal PFB-16.

PS47

SIGNAL TRANSDUCTION IN RESPONSE TO ROOT HYPOXIA: CHARACTERIZATION OF WRKY TRANSCRIPTIONAL FACTORS GENES IN *PRUNUS* ROOTSTOCKS

Carlos Poblete¹, Manuel Acuña¹, Paula Pimentel¹, Pamela Rojas¹, Patricio Hinrichsen^{1,2},
Manuel Pinto^{1,2}, Boris Sagredo¹, Rubén Almada¹

carlos.poblete14@gmail.com

¹Centro de Estudios Avanzados en Fruticultura (CEAF_R08I1001), INIA CRI Rayentué, Rengo, Chile; ²INIA CRI La Platina, Santiago, Chile.

In Chile, root hypoxia limits stone fruit tree development, particularly in orchards established on soils with restricted drainage. To overcome this problem, stone fruit trees are usually grafted on rootstocks (species or hybrid of the *Prunus* L. genus) with different degrees of tolerance to root hypoxia. However, the molecular base of such variability is largely unknown. Transcriptional control plays an important role in regulating hypoxia responses in plants and expression of numerous genes encoding transcription factors (TFs) are regulated during this stress. The expression of genes that encode members of the WRKY transcription factor family is rapidly and strongly induced upon submergence in *Arabidopsis thaliana* suggesting that these TFs might be involved in hypoxic stress response(s). However, the potential role of WRKY-like transcription factors in *Prunus* rootstocks response(s) to root hypoxia is largely unknown. In this study, we used available sequence information to identify a minimum number of 61 *P. persica* WRKY transcription factor (*PpaWRKY*) genes. According to their structural features, the *PpaWRKY* factors were classified into the previously defined polyphyletic WRKY subgroups 1 to 3. With the aim of exploring the possible role of WRKY-like genes during stone fruit rootstock responses to root hypoxia, we monitored the expression patterns of WRKY22-like and WRKY29-like genes in roots of the highly tolerant M2624 rootstock and in F12, another rootstock that is highly sensitive to soil waterlogging. The WRKY22-like and WRKY29-like genes were induced by root hypoxia in both genotypes. However, the WRKY22-like gene was expressed at a higher level in the highly tolerant M2624 genotype compared with the sensitive F12 genotype. Our results suggest that changes in WRKY22-like and WRKY29-like expressions could be part of the adaptive mechanisms that have evolved in the *Prunus* species to survive under hypoxia.

Acknowledgments: CEAF_R08I1001 and FONDECYT N°11110079. Rootstock plants were gently provided by Agromillora Sur S.A.

PS48

**GENETIC CHARACTERIZATION, VIRUS ASSESMENT AND VIRUS-FREE PLANT
REGENERATION OF NON-TRADITIONAL PISCO CULTIVARS
(*VITIS VINIFERA* L.).**

María Alejandra Montoya¹, Cristián González², Antonio Ibacache², and Andrés Zurita-Silva²

andres.zurita@inia.cl

¹Centro de Estudios Avanzados en Zonas Áridas (CEAZA), La Serena, Chile.

²Instituto de Investigaciones Agropecuarias (INIA Intihuasi), La Serena, Chile

Pisco industry is one of the traditional grapevine activities in Atacama and Coquimbo regions. Since 1931, current regulations of Appellation of Origin allows its exclusive production and regulates that viticulture and elaboration of pisco should be performed only from grape musts of specific *Vitis* cultivars grown in these Regions. As consequence, in 1979 SAG established a list of thirteen cultivars for this purpose. Despite of this, the industry only grows five of them, and there is no clear data about the others. We started recovering, genetic characterization and virus assessment for unexploited cultivars. A search at field and grapevine collections throughout Chile was performed to identify those cultivars. We found five out of eight cultivars. To determine genetic relationships and variability, and synonyms or homonyms, we performed molecular genetic analysis by microsatellites (SSR) and Single Nucleotide Polymorphisms (SNPs) detection, which were genotyped from gDNA with 11 and 48 nuclear loci respectively. Molecular data was analysed by multiple correspondence analyses, generating a similarity tree using Euclidean distance and average linkage method chain with a correlation of 0.9, and principal component association for SNPs. We also performed detection for 17 RNA viruses using their specific sequences by RT-qPCR. We detected *Grapevine fanleaf virus* (GFLV) in all samples, and *Grapevine rupestris stem pitting-associated virus* (GRSPaV), *Grapevine rootstock stem lesion associated virus* (GRSLaV), *Grapevine fleck virus* (GFkV) and *Grapevine leafroll-associated virus 2* (GLRaV-2) in some samples. We started meristem culture for grapevine virus eradication, with survivals from 34% to 89%, which correlates with field plant quality. Once genetic relationships and similitude are established, a recovery of ancient cultivars will be performed to support industry diversification.

Acknowledgments: Project InnovaChile 11BPC-9959

PS49

IN SILICO ANALYSIS OF MOTIFS PROMOTERS AND EXPRESSION OF ALCOHOL DEHYDROGENASE RELATED GENES (E.C 1.1.1.1) IN *PRUNUS* SP. L. ROOTSTOCKS WITH CONTRASTING RESPONSE TO HYPOXIA

Simón Solis^{1,4}, Rodrigo Contreras¹, Ariel Salvatierra¹, Rubén Almada¹, Paula Pimentel¹, Manuel Pinto^{1,3} and Boris Sagredo^{1,2}

ssolis@ceaf.cl

¹Centro de Estudios Avanzados en Fruticultura (CEAF), Rengo, Chile. ²INIA CRI Rayentué, Rengo, Chile. ³INIA CRI La Platina, La Pintana, Santiago, Chile. ⁴Universidad Andrés Bello, Facultad de Ciencias Biológicas, Santiago, Chile.

Many species of the genus *Prunus* L. are particularly affected by hypoxic stress. Hypoxia is generated by a decrease of the oxygen supply in the rhizosphere; this is recurrently caused by flooding and/or compact soils, which reduces the diffusion of oxygen to the roots. The most affected process in hypoxia is the energy production, where the generation of ATP is between a 65-97% less compared with normoxia. To overcome this, fermentation pathways are activated, where the ethanolic fermentation helps to keep a minimum energy production, mediated the action of the alcohol dehydrogenase (ADH) enzyme, which transforms acetaldehyde into ethanol, restoring NAD⁺ for the glycolysis. Previous RNA-seq studies have shown the importance of the ethanolic fermentation during hypoxia in *Prunus* rootstocks. The differential expression of the *ADH* related genes in the root transcriptome of two rootstock genotypes with contrasting response to hypoxic stress, suggest differences at gene promoter sequences. In order to obtain a better understanding of the response to hypoxic stress, the promoter sequences of these genes are being analyzed. By *in silico* analysis, there were identified 105 ADH related activity genes (PTHR11695) in the peach genome (*P. persica* L.) and there were 17 genes that presented differential expression under hypoxic condition, in the root transcriptome of both rootstocks genotypes with contrasting responses. From these, only four are involved in the ethanolic fermentation. Analysis *in silico* of *P. persica* promoter genes indicated presence of several motifs related to hypoxia stress. In this moment qRT-PCR experiments are being conducted to confirm their differential pattern of expression. Promoter sequences will be obtained in a massive sequencing using Ion Proton™ System, from both genotypes. Promoter cis elements will be analyzed searching for differences that might explain their differential response to hypoxia conditions.

Acknowledgments: FONDECYT N°1121117 and CEAF_R08I1001.

PS50

**DIFFERENTIAL EXPRESSION OF THE CYTOKININ RESPONSE PATHWAY GENE DURING
FRUIT DEVELOPMENT IN *PRUNUS PERSICA***

Karen Mujica^{1,3}, Fernanda Rodríguez³, Rodrigo Infante², and Lee A. Meisel¹

karen.mujica.i@gmail.com

¹Programa Magister de Bioquímica de la Universidad Andrés Bello, Av. República 217,
837-0146 Santiago. ²Universidad de Chile, Facultad de Ciencias Agronómicas, Chile.

³Plant Molecular Genetics Laboratory, Institute of Nutrition and Food Technology,
University of Chile, Av. El Líbano 5524, 783-0490 Santiago

The availability of the peach genome may be used to facilitate molecular analyses of the cytokinin response pathway during the development of stone-fruits. High levels of cytokinin have been reported previously in early stages of peach fruit development (i.e. "pre-lignification"). However, cytokinin levels decrease at later stages of development (i.e. "post-lignification"). In order to determine the effects cytokinin fluctuations may play in peach fruit development, the expression levels of "cytokinin response pathway orthologs" were analyzed at different developmental stages using qPCR and RNA-seq analyses. To confirm that these orthologs are responsive to cytokinin, transcript levels were also analyzed in fruits at different developmental stages (pre-lignification, lignification and post-lignification) one hour post cytokinin treatment in the laboratory (t-zeatin) and in the field (Cylex ®: benziladenina). These results demonstrate that the "cytokinin response pathway orthologs" are expressed differentially during peach fruit development and that these genes respond differentially to exogenous application of cytokinin in a stage specific manner.

Acknowledgments: FONDECYT 1121021

PS51

**IDENTIFICATION OF SNP-TYPE POLYMORPHISMS ASSOCIATED TO BERRY SIZE
IN TABLE GRAPES (*VITIS VINIFERA* L.)**

Alicia Sánchez¹, Claudia Muñoz^{2,3}, Ariel Orellana^{2,3}, Mauricio González-Aguero¹
and Patricio Hinrichsen¹

phinrichsen@inia.cl

¹INIA La Platina, Santiago, Chile; ²Centro de Biotecnología Vegetal, Universidad Andrés Bello, Santiago, Chile; and ³FONDAP Center for Genome Regulation, Santiago, Chile.

Table grape is the most important fruit crop cultivated in Chile. Berry size is a valuable quality trait for table grape consumers, resulting from cell division and expansion during early phenological stages. Single nucleotide polymorphisms (SNPs) have been widely used as molecular markers in plant breeding programs and linkage maps construction, based on their natural abundance. Also, they can be used as tools for phenotype/genotype association studies. Our aim was to identify SNPs associated to berry size in table grapes, which can be used as selection marker in breeding programs. Using a massive expression experiment (RNA-Seq), seven differentially expressed genes were selected in early stages of berry development, comparing segregants with contrasting phenotypes for berry size derived from the 'Ruby Seedless' x 'Sultanina' crossing. Thirty-one SNPs were identified and 12 of them were analyzed by qPCR-HRM, in a group of six large berry and eight small berry segregants from the RxS crossing. Nine SNPs were confirmed by sequencing and in six of them the association phenotype-genotype was determined. Their transferability was evaluated on 21 table grape varieties and in a *Vitis vinifera* L. core collection (23 genotypes). The polymorphic information content (PIC) was estimated. Considering the allelic frequencies determined for two SNPs, our results suggest a tendency for phenotype-genotype association between large and small berries. The observed PICs values for the group of nine SNPs varied between 0.133 to 0.472, with an average of 0.302. In the case of five of them, high polymorphic levels were observed, supporting their transferability to less related populations. Also, our results suggested the use of these SNPs as tools in plant breeding programs as well as for genetic diversity studies in *Vitis vinifera*.

Acknowledgments: FONDEF-Genoma grants G07I-1002 and G07I-1009; FONDAP CRG 15070009, Núcleo Milenio P10-062-F, Basal PFB-16 and Programa Mecesup.

PS52

**HETEROLOGOUS OVER-EXPRESSION OF TWO RADIATA PINE MADS-BOX GENES
REGULATES PHENYLPROPANOIDS PATHWAY IN *ARABIDOPSIS THALIANA***

Nicolás Cruz¹, Patricio Ramos¹, Claudio Valenzuela¹, Lorena Norambuena²,
Aliosha Figueroa² and Raúl Herrera¹

nicruz@utalca.cl

¹Laboratorio de Fisiología Vegetal y Genética Molecular, Instituto de Biología Vegetal
y Biotecnología, Universidad de Talca. ²Laboratorio de Biología Molecular Vegetal,
Facultad de Ciencias, Universidad de Chile.

In nature, conifer trees develop compression wood in response to inclination at the lower side of the stem. Transcription factors have been described to be involved in lignification processes, but the molecular mechanism underlying the response is still unknown. Total RNA was extracted from radiata pine seedlings after 2.5, 10, 24 h and 1 month of inclination and samples from the upper and lower half of the stem through a longitudinal cut were collected. The search of genes expressed in bent stems was carried out through suppressive subtractive libraries. The analysis revealed that several of these ESTs corresponded to transcription factors with MADS-box and K-box domains. By using 5' and 3' RACE reactions, the full-length sequences of two MADS-box were obtained and named *PrMADBJ* and *PrMADBT*. These genes were introduced into the yeast Y1HGold using the vector pYES2.1. Electrophoretic mobility shift assay (EMSA) was performed in order to determine the specific interaction of these PrMADS-box with DNA elements. These DNA sequences corresponded to the canonic CaGR and those found in the promoter region of a gene involved in cell wall remodeling. Additionally, each sequence was cloned into the binary vector pBI121 and Arabidopsis plants were transformed to generate transgenic plants which over-expressed each *PrMADS-box*. Different transgenic lines were subjected to transcriptomic and metabolomic analyses. The relative expression of several genes involved in the phenylpropanoid biosynthetic pathway was determined by RT-qPCR. Results reveal that the lignin pathway is positively regulated by *PrMADS-box* genes.

Acknowledgments: Fondecyt 1120635, 11121170 and 1120289 projects. N.C. and C.V. thank to CONICYT and U. Talca for doctoral fellowship, respectively.

PS53

**TRANSCRIPTIONAL PROFILE OF SWEET CHERRY FRUITS WITH DIFFERENT
TOLERANCE TO CRACKING.**

Cristián Balbontín¹, Héctor Ayala¹, Joselyn Rubilar¹, Felipe Gainza², Yazmina Stappung³, Jonathan Maldonado⁴, Herman Silva⁴.

cbalbontin@udec.cl

¹*Facultad de Agronomía, Universidad de Concepción, Laboratorio de Biotecnología Vegetal, Chillán, Chile. ² Centro de Estudios Avanzados en Fruticultura, CONICYT-Regional/GORE O'Higgins, Rengo, Chile. ³ Escuela de Bioinformática, Universidad de Talca, Talca, Chile. ⁴ Laboratorio de Genómica Funcional & Bioinformática, Universidad de Chile. Facultad de Ciencias Agronómicas, Departamento de Producción Agrícola, Santiago, Chile.

Rain can induce fruit cracking with a high compromise in the production quality in several cherry-production areas around the world. Recent researches have shown that early cuticle crack development can be correlated with the length of fruit growth phases, which show differences between cultivars with different degrees of cracking tolerance. The development of sweet cherry fruit follows a double sigmoid pattern with two periods of maximum growth, which coincide with the phenological stages of fruit set and fruit color change. In order to better understand of the relationship between cherry fruit cracking and gene expression, we used Illumina paired-end sequencing technology to develop a de novo sweet cherry (*Prunus avium*) fruit transcriptome, identifying highly and differentially expressed transcripts between cultivars and stages of fruit development. The resulting 382 million readings were assembled into 67,383 contigs, with an average length of 894 bp. Overall, 36% of the contigs showed significant similarities to known sequences in GenBank. Non-redundant REFSEQ database and 17,335 contigs could be functionally categorized using Blast2GO. Digital expression analysis allowed us to identify a large set of genes that were differentially expressed between cultivars and phenological stages. These results provides novel insights into sweet cherry fruit biology and a comprehensive resource of candidate genes for future functional analysis as well as their use as molecular marker in sweet cherry breeding programs.

Acknowledgments: CB is supported by CONICYT, FONDECYT/Iniciación N°11100149. HS is supported by CONICYT, FONDECYT/Regular N°1120261

PS54

**TRANSCRIPTIONAL LEVELS OF GENES INVOLVED IN SWEET CHERRY CRACKING
DURING FRUIT DEVELOPMENT.**

Cristián Balbontín¹, Héctor Ayala¹, Joselyn Rubilar¹, Felipe Gainza²

cbalbontin@udec.cl

¹Facultad de Agronomía, Universidad de Concepción, Laboratorio de Biotecnología Vegetal, Chillán, Chile. ²Centro de Estudios Avanzados en Fruticultura (CEAF), CONICYT-Regional/GORE O'Higgins, Rengo, Chile.

The development of cracking in the fruit of sweet cherry (*Prunus avium*) can be induced by rain during fruit ripening or by an abrupt increasing of the cuticle area during fruit growth. Although the difference in the degree of cracking tolerance between different cultivars is very relevant, information about causes of this phenomenon, both physiological and molecular, remains limited. The cuticular membrane is the primary barrier to water transport through aerial parts of the primary plant body and thus their properties can be involved in tolerance to this problem. In our study, we report preliminary results about the evaluation of transcriptional levels of genes involved in the development of the cuticular membrane of cherry fruit. Specifically, we carried out an assessment of three phenological stages during fruit growth, contrasting cultivars with different degrees of cracking tolerance. The expression levels of genes *WINB*, *LTPG1*, *LACS2*, *GPAT4/8* and *CER1* were higher on the cultivar with more tolerance during the different growth stages. These results suggest that the levels of expression of these genes can help to provide a more stable cuticular membrane during the process of fruit growth, preventing the development of cracks before episodes of rain during ripening.

Acknowledgments: CB is supported by CONICYT, FONDECYT/Iniciación N°11100149.

PS55

IDENTIFICATION AND CLONATION OF THE FT/TFL1 FAMILY GENES IN SWEET CHERRY

Michelle Gatica¹, Daniel Almonacid², M^a Angeles Miccono³, Jose G. Vega¹, Fabian Zuñiga¹,
Humberto Prieto³, Andrea Miyasaka Almeida¹.

michelle.unab@gmail.com

¹FONDAP Center for Genome Regulation, Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello, Santiago, Chile. ²Center for Bioinformatics and Integrative Biology, Facultad de Ciencias Biológicas, Universidad Andrés Bello, Santiago, Chile. ³Instituto de Investigaciones Agropecuarias, INIA La Platina, Santiago, Chile

The *FT/TFL1* gene family regulates floral induction in annual and perennial plants. Genes in the FLOWERING LOCUS T (FT) and TERMINAL FLOWER 1 (TFL1) family have been shown to be important in the control of the switch between vegetative and reproductive growth in several plant species. The family members in angiosperms are classified into three clades (MFT, FT, TFL1) well characterized in *Arabidopsis thaliana*. In this species FT/TFL1 gene family is made up of 6 closely related genes; FT, TFL1, ATC (ARABIDOPSIS CENTRORADIALIS HOMOLOGUE), TSF (TWIN SISTER OF FT), BFT (BROTHER OF FT AND TFL1), and MFT (MOTHER OF FT AND TFL1). The aim of this work was to identify and clone the members of the FT/TFL1 family in sweet cherry (*Prunus avium*) due to their important relationship in the control of flowering time and juvenility. Based on the sequence of Arabidopsis for FT/TFL1 family, we performed a BLAST to search for potentials orthologous genes in *Prunus persica* genome. Based on this sequence we designed primers for cloning their *Prunus avium* putative orthologous. Sequences of cloned genes were analyzed with other Rosaceae genes using the program CLUSTALW. We also describe the genomic structure and phylogenetic relationship with other Rosaceae species. The results show that the FT/TFL1 family appears to be highly conserved in different species. Together these results suggest that the identified genes could be regulating the flowering process in sweet cherry. These cloned genes could be applied to future studies and applications regulating the flowering in sweet cherry.

Acknowledgments: FONDEF G09I1008; FONDAP CRG 15070009; and Basal PFB-16.

PS56

**METHYL JASMONATE REGULATES CELL WALL MODIFICATION THROUGH
THE UP-REGULATION OF HEMICELLULOSE MODIFYING GENES DURING RIPENING
OF *FRAGARIA CHILOENSIS* FRUIT.**

Nicolás Figueroa, Cristóbal Concha, Leticia Poblete, Felipe Oñate, Carlos Figueroa.

nicfigueroa@udec.cl

Facultad de Ciencias Forestales y Centro de Biotecnología, Universidad de Concepción,
Concepción, Chile.

The role of methyl jasmonate (MeJA) on cell wall (CW) modification during ripening of non-climacteric fruits is not well understood. We analyzed the effects of 10 and 100 μM MeJA on cell wall composition (CWC) and expression of cell wall-modifying (CWM) genes during ripening of *Fragaria chiloensis* fruits at days 2, 5 and 9 by using an in vitro system. CW was fractionated from alcohol insoluble residue (AIR) into water, CDTA, Na_2CO_3 and KOH soluble fractions (WSF, CSF, NSF and KSF, respectively), and the concentration of uronic acid (UA) and neutral sugar (NS) was colorimetrically determined. The expression of several CWM genes was determined by qPCR. No changes in UA or NS content in the CSF and NSF were detected but at 9d a lower AIR was found in 100 μM MeJA treated fruits, correlating with higher content of UA in the WSF. At 9d, both concentrations of MeJA decreased the content of NS in the KSF, suggesting higher hemicellulose degradation compared to control. The expression of pectate lyase (PL) and polygalacturonase 1 (PG1) was not affected by MeJA, while the expression of pectin methylesterase 1 (PE1) and expansin 2 (EXP2) was decreased at 2d with 100 μM . However, the expression of endoglucanase 1 (EG1) was strongly increased in both treatments at all days, correlating with lower firmness at 5d and lower NS concentration in the KSF at 9d. Furthermore, putative JAs-responsive elements in its promoter region were found. MeJA increased the expression of xyloglucan endotransglycosylase/hydrolase 1 (XTH1) at 5d in both treatments, which could also explain the lower firmness of treated fruits, but decreased its expression at days 2 and 9. In conclusion, MeJA could regulate softening and CWC during ripening of *F. chiloensis* fruit through the up-regulation of hemicellulose modifying genes.

Acknowledgments: FONDECYT 11110171.

PS57

**EXPRESSION PATTERNS OF ABA PERCEPTION GENES (PYR FAMILY) DURING
DEVELOPMENT OF *FRAGARIA CHILOENSIS* FRUIT**

Rodrigo Lizana, Yazmina Stappung, Enzo Viera, Andrea Olivos, Raúl Herrera,
María A. Moya-León

rlizana@utalca.cl

Laboratorio de Fisiología Vegetal y Genética Molecular, Instituto de Biología Vegetal
y Biotecnología, Universidad de Talca.

The native Chilean strawberry fruit (*Fragaria chiloensis* (L.) Mill) has potential as an exotic fruit in the international market. However, its fast softening limits its commercialization. The control of ripening in non-climacteric fruit such as strawberry remains unclear, although auxins and abscisic acid (ABA) may have a role on it. During development of strawberry fruit the level of auxins increases during the first half and decreases after that (with ripening development), while that of ABA increases constantly. Auxins and ABA are synthesized in the achenes of the fruit, and ABA is perceived in the receptacle by different ABA receptors, like the PYR/PYL family. We identified 8 PYR genes within *F. vesca* 's genome, belonging to different phylogenetic groups. Based on their sequence qPCR primers were designed. The transcription level of 8 different PYR genes was determined by qPCR in different *F. chiloensis* fruit stages (C1 to C4) and vegetative tissues (stem, roots, flowers, leaves and runners). Different transcriptional patterns were recorded for *F. chiloensis*'s PYR genes. In particular, FcPYR1 and FcPYR3 displayed a reduction in their transcripts level during fruit development and ripening, while that of FcPYR7 increase specially during ripening. When C2 stage fruits were treated with ABA the expression of FcPYR1, FcPYR3 and FcPYR7 was induced. Treatment of fruit with auxins induced the expression of FcPYR7, and when achenes were removed there was a reduction in transcript level. On the contrary, the expression of FcPYR1 and FcPYR3 was diminished by auxins application. This suggests that PYR7 could be involved in ABA perception during ripening of *F. chiloensis* fruit and it is regulated by auxins and ABA, while FcPYR1 and FcPYR3 are positively regulated by ABA and negatively regulated by auxins.

Acknowledgements: RL to Universidad de Talca for his doctoral fellowship. Research supported by Fondecyt 1110792 and Anillo ACT-1110 projects.

PS58

**OVEREXPRESSION OF *CBF3/DREB1A* GENE IN *CITRUS MACROPHYLLA* W
INCREASE SALINITY TOLERANCE**

Ximena Alvarez-Gerding^{1,2}, Carmen Espinoza², Claudio Inostroza-Blancheteau³,
Patricio Arce-Johnson²

parce@bio.puc.cl.

¹Facultad de Agronomía e Ingeniería Forestal, Pontificia Universidad Católica de Chile, Santiago, Chile. ²Facultad de Ciencias Biológicas, Departamento de Genética Molecular y Microbiología, Pontificia Universidad Católica de Chile, Santiago, Chile.

³Núcleo de Investigación en Producción Alimentaria, Facultad de Recursos Naturales, Escuela de Agronomía, Universidad Católica de Temuco, Temuco, Chile.

Soil salinity is an environmental stress that has increased and agricultural lands may lose their productivity for it cause, especially under irrigation. Citrus species, of tropical origin, are highly sensitive to salts, particularly chloride. Lower growth rate and yield was observed from 1.4 dS m⁻¹, around 15 mM NaCl. The rootstocks comprise the root system of fruit trees and the ability to regulate ion uptake and transport to the shoot rely on them. Therefore, increasing salt tolerance of rootstocks could increase the tolerance of the variety. For this purpose, we genetically transformed *Citrus macrophylla* W. - a rootstock for lemon- with *CBF3/DREB1A* gene from *Arabidopsis thaliana* regulated by the constitutive promoter *CaMV35S*. Transgenic plants showed a normal size, with no evidence of dwarfism, higher growth and stomatal conductance and lower chloride and sodium accumulation in leaves compared to WT plants. This would confirm the higher salt tolerance caused by overexpression *CBF3/DREB1A* gene in the citrus plants. Quantitative real-time (qRT-PCR) analysis showed a high expression of *COR15A*, *LEA 4/5*, *P5CS*, *GOLS*, *LKR/SDH* and *ADC2* genes, only in the transgenic plants under salt treatment, but not under control conditions although driven by a constitutive promoter. This suggests a complementary pathway regulating the expression these *CBF3/DREB1A* target genes.

PS59

**RELATIONSHIPS OF SMALL INTERFERING RNA SPECIES TARGETING
THE *PLUM POX VIRUS* GENOME IN *NICOTIANA BENTHAMIANA* UPON INFECTION
AND RESISTANCE CONDITIONS.**

Montes, C., Castro, Á., Barba, P.2, Rubio, J., Sánchez, E., Carvajal, D.1, Aguirre, C., Tapia, E.,
Dell'Orto, P., Decroocq, V.3, and Prieto, H.

christian.montes.s@gmail.com

Gene silencing approach has become a crucial technique for developing pathogen resistance strategies for *Plum pox virus* (PPV), a high impact viral pathogen in *Prunus* spp. In addition, the use of massive sequence analyses of small RNAs is now offering new possibilities to understand the relevant steps executed during the process of RNA interference (RNAi). In the present work, the design of a silencing vector (pPPViRNA) harboring two regions from the PPV coat protein (CP) gene predicted as highly rich in silencing motifs, was evaluated in *Nicotiana benthamiana* (NB) transgenic plants. NB F₁ plants transformed with pPPViRNA depicted a broad range of susceptibility patterns, including the generation of several fully resistant individuals. Representative individuals for the transgenic-resistant (TG-R) and -susceptible (TG-S) phenotypes were used for massive small RNA analysis under virus challenge regarding siRNAs of known activity (i.e. 21- to 24-nt molecules, siRNAs). Whereas a similar pattern of siRNA species, with almost an exclusive occurrence of 21- and 22-nt siRNAs was seen in TG-S and non-transformed (WT) controls plants, TG-R plants showed the accumulation of the full set of 21- to 24-nt siRNAs, which specifically targeted the CP zones used in the silencing vector. Important associations in terms of sequences were observed between 21-nt and 22-, 23-, and 24-nt molecules, suggesting the generation of siRNA families taking place during RNAi. When the two CP regions used for pPPViRNA were further analyzed, 13 common siRNA families were found to be present in all WT, TG-S, and TG-R plants. The results indicate a potential role of 21-nt species in the biogenesis of 22- to 24-nt species in RNAi in plants.

Acknowledgements: FONDEF-CHILE G09I1008 and ECOS-CONICYT C09B03.

PS60

**STUDY OF THE INTERACTION BETWEEN *PLUM POX VIRUS* - *PRUNUS SPP*
BY GENE SILENCING OF EUKARYOTIC INITIATION TRANSLATION FACTORS,
EIF4F, FROM PEACH (*PRUNUS PERSICA*) IN A HETEROLOGOUS SYSTEM.**

Julia Rubio-Astudillo¹, Christian Montes², Álvaro Castro², Carole Couture³,
Veronique Decroocq³, Humberto Prieto².

jrubioa@gmail.com

¹ Doctorado en Ciencias Silvoagropecuarias y Veterinarias, Universidad de Chile,
Santa Rosa 11315, Santiago de Chile (Chile). ² INIA, La Platina Station, Santa Rosa 11610,
La Pintana, Santiago de Chile (Chile). ³ UMR 1332 BFP INRA, Equipe de Virologie CS20032,
71 Edouard Belin, 33882 Villenave d'Ornon (France)

Plum Pox Virus (PPV) is the most devastating virus disease in stone fruits. Despite the measures taken for its eradication, new outbreaks have been detected recently across the continents. Recent results suggest that virus resistance in plant is mainly achieved by "passive" mechanisms of tolerance. These mechanisms would be determined by the absence or inappropriate presence of "sensitivity" factors in the host. Furthermore, it is known that eukaryotic initiation translation factors (eIFs) exhibit high degrees of similarity between different species, which reinforces their use in a heterologous system for study the interaction between different species and PPV. This project involves the use of highly susceptible sequences of the eIFs gene silencing process, detected in orthologous genes from peach (*Prunus persica*) to develop the respective silencing constructs. The use of this technology in *Arabidopsis thaliana* will determine the role of these factors, eIF4E, eIF4G or its isoforms as determinants of host susceptibility during PPV infection. Recent results indicate that genetic modified *A. thaliana* generated using the silencing construct for eIF(iso)4G10 from peach, do not get infected by the chimeric PPV tagged to GFP, indicating that host expression of this eIF could be indispensable for the interaction between PPV and the orthologous gen in *A. thaliana*.

Acknowledgements: FONDEF-CHILE G09I1008 and ECOS-CONICYT C09B03. JR for Doctoral CONICYT scholarship holder.

PS61

**ANALYSIS OF THE BZIP60 MRNA PROCESSING UNDER DIFFERENT TREATMENTS
IN *ARABIDOPSIS THALIANA* USING CAPILLARY ELECTROPHORESIS**

Juan Pablo Parra, Adrián Moreno, Ariel Orellana

parrajuan24@gmail.com

FONDAP Center for Genome Regulation, Núcleo Milenio en Genómica Funcional de Plantas,
Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello.

The unfolded protein response (UPR) is a signaling pathway associated with the endoplasmic reticulum (ER). Normally, proteins synthesized on the ER that fail to acquire their native conformation are degraded by a mechanism termed ER-associated degradation. Nevertheless, if this mechanism fails to degrade unfolded/misfolded proteins, these are retained in the ER triggering the UPR. During this process a set of genes that can assist the correct folding of proteins are up regulated at transcriptional and translational level. Recent studies in *Arabidopsis thaliana* and rice identified a signaling pathway in which an ER transmembrane protein called IRE1 catalyzed the unconventional cytoplasmic splicing of a transcription factor mRNA leading to the synthesis of an active transcription factor capable of activating the UPR-responding genes. This transcription factor is known as bZIP60 in *Arabidopsis* and bZIP50 in rice. Recent reports have shown that two classical chemicals used to induce the UPR (tunicamycin and DTT) activate the processing of bZIP60. Nevertheless the dynamics and temporality of the unconventional splicing is unknown. Additionally, salicylic acid (SA) can also induce the processing of bZIP60 but the processed product is barely detectable using common agarose electrophoresis as analytical instrument. We have employed the capillary electrophoresis to analyze the non-conventional splicing of bZIP60, due to its high resolution and quantification capacity. Plants treated two hours with tunicamycin and DTT reached twenty five percent of splicing and only five percent in plants treated with SA when compared to control plants. Moreover plants treated with DTT, washed and treated again shown an attenuation of splicing after the washing-out process and similar a bZIP60 splicing percentage after reinduction of the UPR. Our results suggest that bZIP60 processing is a dynamic and stimulus-dependent event and unlike XBP1 in mammals, the bzip60 processing becoming never full.

Acknowledgments: Fondecyt N° 1110954, FONDAP CRG 15070009; Núcleo Milenio P10-062-F and Basal PFB-16.

PS62

PARTICIPATION OF TRANSCRIPTION FACTORS bZIP17 AND bZIP60 IN RESPONSE TO SALT STRESS IN *ARABIDOPSIS THALIANA*

Carlos Henríquez, Adrián Moreno, Irina Mitina, Omar Sandoval, Ariel Orellana

carlos_henriquezva@hotmail.com

FONDAP Center for Genome Regulation, Núcleo Milenio en Genómica Funcional de Plantas, Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello.

In plants, abiotic and biotic stress leads to the accumulation of misfolded proteins in the Endoplasmic Reticulum (ER) triggering the Unfolded Protein Response (UPR). As signal transducer of this response, two transcription factors known as bZIP28 and bZIP60, move to nucleus where they promote up-regulation of ER associated genes such as *BiP* (*Binding Protein*). Another transcription factor associated to the UPR is bZIP17, which is the only ER bound transcription factor described to be activated in response to salt. Recently, bZIP60 has been associated with the response to salt stress and the expression of several stress inducible genes. Nevertheless, the relationship between bZIP17 and bZIP60 in salt stress has not been determined. Analysis of the bZIP60 promoter revealed two putative ABRE elements and a TGACG box. Surprisingly the TGACG box is present in all the genes that are regulated by bZIP17. To get insights of the possible regulation of bZIP60 by bZIP17, we perform RT-qPCR analyses of BiP3, bZIP60 and other stress inducible genes regulated by bZIP60 on bzip17 mutant plants treated with 150 mM NaCl. A significant lack of expression for these genes was observed. Also, the activation of IRE1/bZIP60 branch of the UPR was absent suggesting that this signaling pathway is not associated to the canonical UPR under salt treatment. On the other hand, expression analyses performed on the bzip60 mutants shown that the up-regulation of BiP3 and other bZIP17 target genes was abolished on these plants treated with NaCl. Our results strongly suggest that bZIP17 and bZIP60 participate both together in the transcriptional regulation of a set of stress responsive genes under salt stress conditions.

Acknowledgments: Fondecyt 1110954, FONDAP CRG 15070009; Núcleo Milenio P10-062-F, Basal PFB-16 and CONICYT.

PS63

IDENTIFICATION AND EVALUATION OF DORMANCY RELATED GENES IN SWEET CHERRY BUDS OVER A GROWING SEASON.

José Pablo González¹, Hugo Portillo¹, Michelle Gatica¹, Eduardo Tapia², Evelyn Sanchez²,
Humberto Prieto², Andrea Miyasaka de Almeida¹

jpx14@hotmail.com

¹Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello, FONDAF Center for Genome Regulation, Núcleo Milenio en Genómica Funcional de Plantas, Santiago. ²Instituto de Investigaciones Agropecuarias, INIA La Platina, Santiago.

In winter, perennial trees from temperate zones such as sweet cherry, present a cease of growth. In this season, both floral and vegetative buds stop growing and enter in a latent state called dormancy. To resume growth, the trees need to pass through an extended period of cold denominated as chilling requirement. Annual plants such as *Arabidopsis* passes through a process called vernalization, where they need an extended period of cold, similar of what happens in dormancy. The aim of this study was to identify and follow the expression of dormancy related genes in a growing season and associate them with the chilling requirement in sweet cherry. Due to the similarity between vernalization in annual plants and dormancy in perennial trees, we sought putative orthologous genes involved in vernalization process in the genome of *Prunus persica* (peach). We identified five possible orthologous genes, *VERNALIZATION 5/VIN3-LIKE1 (VRN5/VIL1)*, *VERNALIZATION 2 (VRN2) VIN3-LIKE 1 (VIL2/VEL1)*, *VERNALIZATION 1 (VRN1)* and *FLOWERING LOCUS T (FT)*. *VIL1*, *VEL1* and *VRN2*, are part of the POLYCOMB REPRESSIVE COMPLEX 2 (PRC2), involved in epigenetic repression of *FLOWERING LOCUS C (FLC)* gene, a negative regulator of flowering. The gene expression analysis by qPCR was done in buds from sweet cherry "Bing". We observed that genes *VIL1*, *VEL1*, *VRN1*, *VRN2* and *FT* expression remained low until tree had accumulated about 1300 chilling units, when all the genes showed a peak of expression. The PRC2 genes are necessary for the expression of FT in *Arabidopsis*; we can expect that these genes may be expressed before the induction of FT expression. This was not observed maybe due to the large difference in days between sample collections. This study is the first that identified genes with differential expression in dormant cherry buds, which could be regulating this process.

Acknowledgments: FONDEF G09I1008, FONDAF CRG 15070009; Núcleo Milenio P10-062-F and Basal PFB-16.

PS64

**EFFECT OF SOWING DATE ON SEED YIELD OF CHIA (*SALVIA HISPANICA* L.)
IN THE AZAPA VALLEY**

Leslie Pizarro¹, Hugo Escobar ¹, Cecilia Baginsky², Luis Morales², Herman Silva R²

lesliepizarro@gmail.com

¹ Facultad de Agronomía, Universidad de Tarapacá, Chile; ² Facultad de Ciencias Agronómicas, Universidad de Chile, Santiago.

Chia (*Salvia hispanica* L.) is an annual plant whose edible seed has a high content of fatty acid of omega -3 which imparts health benefits especially in the prevention of cardiovascular diseases. To evaluate the adaptation of this specie in Chile, a study was conducted at the XV region of Arica and Parinacota, in the Valley of Azapa characterized by a desert climate with optimal conditions for growing Chia. The study considered the crop agronomic evaluation of two genotypes of Chia (white and gray) set in five sowing dates (04 / I, 18 / I, 04/II, 19/II and 06/III). The plants were evaluated in relation to phenological factors and seed yield. The onset of flowering is observed on average 60 days. The 50% anthesis was completed at 70 days. Harvest maturity was achieved at 136 days. The height of plants at harvest, showed a range from 50.9 to 148.6 cm for the dark seed genotype and 54.2 to 146.6 cm for genotype white seeds. The first crop biomass was 476 g fresh weight, with a tendency to decrease from the first to the fourth planting date and a slight increase in the fifth crop. Seed yield, by weight, values ranged between 1499.6 to 3098.0 kg ha⁻¹ for the dark seed genotype 1.307.5 to 2332.6 kg ha⁻¹ for white seed genotype. We conclude that the Azapa Valley presents optimal soil and climatic conditions for the establishment of chia in Chile, considering that the best planting date was the fifth seed, which has significant differences respect to the other planting dates. However, no significant differences were found between genotypes of the parameters evaluated.

Acknowledgments: FONDECYT N° 1120202

PS65

**EFFECT OF CONTROLLED WATER DEFICIT ON PHYSIOLOGICAL AND BIOCHEMICAL
PARAMETERS OF FOUR *PRUNUS* ROOTSTOCKS.**

Cristian Hernández¹, MaríaTeresa Pino², Marcia Bravo¹, Paula Pimentel¹, Manuel Pinto^{1,2}.

chernandez@ceaf.cl

¹Centro de Estudios Avanzados en Fruticultura (CEAF), Instituto de Investigaciones Agropecuarias (INIA) CRI Rayentué, Rengo, Chile. ²Instituto de Investigaciones Agropecuarias (INIA) CRI La Platina, Santiago, Chile.

Water stress is one of the most important abiotic stresses associated to the world climate change. The center of Chile -main fruit producing area- is one of the most affected areas by drought stress periods. However, the information about how the fruit-rootstocks respond to water deficit is still scarce. During season 2012-2013, CEAF studied the controlled drought stress treatment in four *Prunus* rootstock, Mariana2624, Garnem, Cab 6P y Mazzard F-12/1. Plants in 10 L pots were subjected to 100 % ET irrigation (control treatment) and water-deficit treatment (no irrigation until the stem water potential (Ψ_{stem}) reached -2 MPa). The experimental design was a randomized complete block with 4 replications. Under drought stress treatment, Garnem reached Ψ_{stem} of -2 MPa after 10 days, and Mariana2624 reached -2 MPa after 28 days. Cab 6P and Mazzard F-12/1 did not show changes in Ψ_{stem} throughout the whole assay. Under water deficit, Garnem and Mariana2624 showed a significant decrease of CO_2 assimilation and stomatal conductance by cross sectional area of trunk (TCSA) at day 10 and 28, respectively. However, Cab 6P and Mazzard F-12/1 did not show differences in leaf gas exchange. Moreover, Garnem and Mariana2624 showed a significant decrease in starch content and a significant increase in leaf total soluble sugars, proline and hydrogen peroxide after days 10 and 28 under drought stress, respectively. Cab 6P and Mazzard F-12/1 did not show significant differences. In addition, no significant differences in malondialdehyde content were observed for any of the genotypes tested. In conclusion, Garnem and Mariana2624 were more sensitive to drought stress treatment than Cab 6P and Mazzard F-12/1, showing a decrease in leaf gas exchange and an increase in osmolytes which are potent osmoprotectants that play a role in counteracting the effects of drought stress.

Acknowledgments: CEAF_R08I1001, CONICYT and Agromillora Sur S.A.

PS66

**PHENOTYPIC VARIABILITY IN THE ACCUMULATION OF WATER-SOLUBLE
CARBOHYDRATES IN SPRING WHEAT STEM**

M. Alejandra Yáñez Alegría¹, Alejandro del Pozo Lira¹, Gerardo Tapia²

maryanez@utalca.cl

¹Universidad de Talca, Talca, Chile; ²INIA-Quilamapu, Chillán, Chile

The Wheat (*Triticum aestivum* L.) is a crop produced mainly in mediterranean zone of Chile, characterized by low winter temperatures and a wide temperature variation and water shortages during the summer, which affect crop yield. The 80% of wheat is cultivated in upland soils and their production is concentrated in the VI to IX region under Mediterranean conditions. For these environments is necessary to identify wheat genotypes with increased tolerance to water stress, potential to be used as parents in the development of new varieties. Grain yield is the primary aim in drought tolerance of wheat in breeding programs. Characters used as selection criteria should help to improve the productivity and stability in water stress conditions. The main objective of this study is to evaluate the phenotypic variability in the accumulation of water-soluble carbohydrates (WSC) in spring wheat genotypes in response to terminal water stress. In this research we studied WSC at anthesis and physiological ripening in 384 genotypes of spring wheat from INIA Uruguay, CIMMYT México and INIA Chile. Genotypes were evaluated in three geographical ambient (VIII region in Santa Rosa with and without water stress and VII region in Cauquenes with water stress) Contrasting genotypes were selected and expression of genes related with fructans biosynthesis was analyzed. The results showed differences in sugar content of the stems at anthesis with values from 139,25 to 178,15 mgg⁻¹, found for restrictive water ambient and non-restrictive water ambient, respectively. No differences were found at Physiological maturity. Expression of fructan genes was differential in post-anthesis period. The results related to performance showed that crop yield depends on the environment; the number of grains per spike was genotype dependent. We found higher values of carbohydrates in water stress conditions.

Acknowledgements: This work was realized with financing of FONDECYT.

PS67

**CHARACTERIZATION OF VEGETATIVE PHASE CHANGE FROM JUVENILE
TO ADULT IN *PRUNUS SP.***

Adriana Bastías¹, Paula Pimentel¹, Boris Sagredo¹, Patricio Hinrichsen^{1,2}

adrianabastiasb@gmail.com

¹Centro de Estudios Avanzados en Fruticultura (CEAF), INIA CRI Rayentué, Rengo, Chile.

²INIA CRI La Platina, Santiago, Chile

Plants undergo several changes during their life cycle, including changes in the vegetative morphology, reproductive potential and flowering. The juvenile phase of a plant begins at its seedling stage. This phase is characterized by a variety of morphological traits including the shape, size, and insensitivity of the shoot to floral stimuli. The transition to the adult phase (vegetative phase change) is marked by changes in leaf morphology and an increase in reproductive potential. Flower induction begins during the adult phase, and depends on endogenous signals as well as environmental factors. In trees, besides flowering, there are other phenotypic changes associated with maturity characteristics: shoot growth, branch angle, branching pattern and physiological parameters. In *A. thaliana* the major phenotypic differences and molecular mechanism (miR156, miR172, *SPLs* y *TOEs*) associated with change of phase have been described. In this work the homologous genes involved in the phase change from juvenile to adult between *A. thaliana* and *Prunus persica* were determined *in silico*. Bioinformatic analysis of putative cis-regulatory elements of promoter regions of genes *PpSPLs* and *PpTOEs* suggest regulation by N metabolism, light, hormones and stress. In the *Prunus* rootstocks "Garnem" the change of phase is observed from plants of two to three years old. These plants are being used as example to study the change of phase in *Prunus sp.*, by comparison of morphological, physiological parameters and gene expression.

Acknowledgments: FONDECYT N°3120013. Plants were provided by Agromillora Sur S.A.

ECOPHYSIOLOGICAL RESPONSES OF ANTARCTIC PLANTS AGAINST THE IN SITU TEMPERATURE INCREASE

Patricia Sáez¹, León Bravo², Carla Alvear², Carolina Sanhueza³, Ángela Sierra-Almeida³,
Claudia Reyes³, Lohengrin Cavieres³

patrisaezd@gmail.com

¹Laboratorio Cultivo de Tejidos Vegetales, Centro de Biotecnología, Facultad de ciencias Forestales, Universidad de Concepción. ²Laboratorio de Fisiología y Biología Molecular Vegetal, Facultad de Ciencias Agropecuarias y Forestales. Center of Plant, Soil Interaction and Natural Resources Biotechnology, Scientific and Technological Bioresource Nucleus, Universidad de La Frontera, Temuco. ³ECOBIOSIS, Departamento de Botánica, Facultad de Ciencias Naturales y Oceanográficas, Universidad de Concepción.

Limiting factors for plant development in the Antarctic Peninsula are modified by the global warming (average air temperature increases 2.6°C over the last 50 years). Antarctic warming is correlated with a significant increase in plant size, plant cover and population size of antarctic plants. Here we studied the effect of *in situ* warming on ecophysiological characteristic of antarctic vascular plants. Warming was achieved in the field with open top chambers (OTC) installed in December 2012 in populations of *D. antarctica* and *C. quitensis* growing in King George Island (62° 09'S, 58° 28'W). Chlorophyll a fluorescence and growth were evaluated in February of 2013. Additionally, microclimate conditions were registered both inside and outside OTC. Air temperature was increased inside OTC. Air relative humidity was reduced inside the OTC during the day, but it was higher than outside overnight. Leaf temperature was increased inside OTC during the day, while during the night there were no differences between inside and outside OTC. While plant growth in *D. antarctica* was not affected by warming, in *C. quitensis* growth was positively affected by warming. This increase was consistent with increases in soil nitrogen. Photochemical measurements indicated that only *C. quitensis* responded positively to warming (i.e. significant increases in electron transport rate). These plants decreased their excitation pressure on PSII and increased their ability to drive light energy through the photochemical way. In contrast, *D. antarctica* showed no difference between plants growing inside and outside OTC. Our results show that both species have a different potential to respond to increases in temperature. Thus it is necessary to continue these studies to predict the likely effects of climate change on antarctic plants ecophysiology.

Acknowledgment: Project PIA ART 1102

PS69

**PHYSIOLOGICAL RESPONSES OF PLANTS OF CHIA (*SALVIA HISPANICA* L.)
TO WATER DEFICIT DURING THE VEGETATIVE GROWTH PHASE**

Sebastian Alister, María Paz Quezada, Cecilia Baginsky, Luis Morales, Herman Silva

sebastian.alister@ug.uchile.cl

Laboratorio Relación Suelo Agua Planta, Facultad de Ciencias Agronómicas,
Universidad de Chile.

We evaluated the effect of water availability on water relations and gas exchange in four accessions of chia: Antumapu (Chile), Santa Cruz (Bolivia), Atlixco and Acatic (México), established in the Región Metropolitana of Chile. In treatment T1 plants were maintained at field capacity during the vegetative growth phase (0-78 days after planting, DAP); in treatment T2 plants were submitted to total water deficit for 24 days (54-78 DAP). During the experimental period soil water content, vapor pressure deficit (VPD) and leaf turgor were recorded continuously using the non-invasive leaf patch clamp pressure probe (LPCP), as well as performing discrete measurements of gas exchange using an infrared gas analyzer system (IRGA) and water potential with the pressure bomb. During the first 54 DAP the mean values of photosynthetic rate, transpiration rate and stomatal conductance for the 4 provenances were $25 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$, $7.5 \text{ mmol H}_2\text{O m}^2 \text{ s}^{-1}$ and $310 \text{ mmol m}^2 \text{ s}^{-1}$, respectively, with a transpiration efficiency (TE) of $3.3 \mu\text{mol CO}_2 \text{ mmol H}_2\text{O}^{-1}$. From 54 to 78 DAP the plants of T2 had values of $9.5 \mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$, $4.5 \text{ mmol H}_2\text{O m}^2 \text{ s}^{-1}$ and $179.4 \text{ mmol m}^2 \text{ s}^{-1}$ in photosynthesis, transpiration and leaf conductance, respectively, with a TE of 2.1. The differences between provenances were not significant. Continuous measurement of cell turgor with the ZIM system showed high correlation between the values of leaf water potential, soil water content and VPD ($r = -0.8, 0.76$ and 0.95 , respectively). The results indicate that chia plants are highly resistant to the lack of water; they were able to support a level of water stress of -3.0 MPa and a 36% reduction of the TE compared to non-stressed plants and still recover. Both treatments flowered at 95 DAP. This high water deficit tolerance in chia may be explained by the presence of mucopolysaccharides in their leaf tissues observed through scanning microscopy.

Acknowledgments: FONDECYT N°1120202

SEED PROPAGATION OF *CARICA CHILENSIS* (PLANCH. EX A.DC.) SOLMS

Vania Leal^{1,2}, Carolina Uquillas¹, Carolina Zúñiga¹, Ariel Pinolef¹, Víctor Vásquez¹,
Carlos Magni², Iván Grez², Betsabé Abarca²

vania.leal.f@gmail.com

¹Instituto de Investigaciones Agropecuarias CRI La Platina.

²Facultad de Ciencias Forestales y Recursos naturales, Universidad de Chile.

Chile has a high number of species at conservation risk, due to the increasing exploitation of natural resources, being *Carica chilensis* one of the species listed as Vulnerable. Understanding the propagation limitations or complexity of the species under conservation risk is necessary for the design of conservation strategies and repopulation. The existing phenological and morphological knowledge on the species are both null, therefore the study of the spread is extremely important as generating a significant scientific contribution to the study of *Carica chilensis*. This research aims to establish a protocol for the seed propagation of *Carica chilensis* by optimizing its germination capacity. The assay was developed in the Laboratory of Grape Genetic Improvement of the *Instituto de Investigaciones Agropecuarias* of the Regional Center of Research *La Platina*. There are no previous backgrounds on germination in *C. chilensis*, so the four defined treatments were tested in *Carica pubescens* due to the difficulty to obtain *C. chilensis* seeds and by its physiological and morphological similarity. Extracted seeds were washed under running water to remove the surrounding mucilage. The evaluated treatments were: T₀, seeds were dried on a paper towel; T₁, seeds were immersed for 15 s. in hot water at 158°F (70°C) and submerged for 24 hours in distilled water; T₂, seeds were immersed in gibberellic acid (GA₃) for 15 s. at a 250 mg L⁻¹ concentration; T₃, seeds were air dried for one week ; T₄, seeds were submerged in 1.7% hydrogen peroxide for 12 hr., washed under running water for 10 s. and immersed in GA₃ for 24 hr. at 250 mg • L⁻¹ concentration. After applying the treatments, seeds were sown in a substratum, consisting of organic soil, vermiculite and peat, in the ratio of 2:1:1, respectively. T₄ was the most effective treatment for *C. pubescens* obtaining a 55 % of germination at 35 days. This treatment was applied in *C. chilensis*, obtaining a 15 % of germination at 35 days. Previous attempts of seed propagation have been unsuccessful. This research is important, as it allows us to use techniques that improve the germination capacity of *Carica chilensis*, specially, since there is no previous studies that can contribute to optimize its multiplication by seeds.

PS71

**PREHARVEST APPLICATIONS OF METHYL JASMONATE AND CHITOSAN IMPROVE
QUALITY AND TOLERANCE TO FUNGAL DECAY OF *FRAGARIA CHILOENSIS* FRUIT DURING
POSTHARVEST STORAGE**

Gabriela Saavedra, Nicolás Figueroa, Leticia Poblete, Carlos Figueroa

gabrisaavedra@udec.cl

Facultad de Ciencias Forestales y Centro de Biotecnología, Universidad de Concepción,
Concepción, Chile.

The Chilean strawberry is a highly perishable fruit, very susceptible to fungal invasion during postharvest. On the other hand, some natural elicitors, such as methyl jasmonate and chitosan, have received much interest for potential applications in agriculture due to potential effects on the accumulation of metabolites related to phenylpropanoid pathway and defense against fungal decay. The aim of this research was to determine the effect of preharvest applications of 250 μ M MeJA and 1.5% w/v chitosan on fruit quality attributes and tolerance to decay during 72 h of postharvest storage at 22°C. At 48 h, the decay incidence was 0% in MeJA treated fruit while in chitosan and control treatments a decay incidence was observed. Lignin content in MeJA-treated fruits was higher at 48 h and 72 h that could be related with the increasing tolerance to decay. On the other hand, strawberries treated with MeJA or chitosan have higher fruit firmness at 24 h and 48 h. Furthermore, anthocyanin content increased in both elicitor treatments at 0, 48 and 72 h compared to control. These results suggest that MeJA and chitosan applications could be used as favorable treatments to extend shelf-life and to improve the quality of *F. chiloensis* fruit.

Acknowledgements: FONDECYT N°11110171.

PS72

EFFECT OF DROUGHT AND HIGH RADIATION STRESSES IN ROS SCAVENGE ENZYME ACTIVITIES FOR GERMPLASM COLLECTION OF WILD TOMATOES.

Gerardo Tapia¹, Oscar Arrey^{1,2}

gtapia@inia.cl

¹Unidad de Recursos Genéticos Vegetales, Instituto de Investigaciones Agropecuarias, INIA-Quilamapu, Chillán. ²Departamento de Ciencias y Tecnología Vegetal, Universidad de Concepción, Los Ángeles.

Several environmental stresses induce production of reactive oxygen species (ROS), which cause damages to cellular components including proteins, lipids, carbohydrates and DNA, resulting in oxidized products. It produces senescence of photosynthetically active organs of the plant, reducing possibilities of growth until affect survival in the time. Under normal conditions ROS are produced, however, under unfavorable environmental conditions such as extreme temperatures, heavy metals, drought or salinity these enhance their production. Plants possess mechanisms for scavenger ROS mediating enzymatic and non-enzymatic way. Between the most important components of enzymatic way are catalase (CAT), Ascorbate peroxidase (AsPX) and Guaiacol peroxidase (POX). On the other hand, wild tomato species *Solanum chilense* and *Solanum peruvianum* have been characterized by their tolerance to several biotic and abiotic stresses, being a source of genes for cultivated breeding of tomato. This study covers the evaluation of CAT, AsPX and POX enzymatic activity in 14 different genotypes of *S. chilense* and *S. peruvianum*. These enzymes were analyzed during drought stress by two regimes of light intensities. The results show higher values of AsPX activity under high radiation (HR) than low radiation (LR). We did not observed differences between combinations of HR-control and HR-drought. During LR treatment drought induce increase of AsPX similar than HR. POX activity show an increase in HR compared with LR, but is independent of drought stress. CAT activity not showed significant differences between treatments. We also identified a genotype with higher level of AsPX and POX, which propose it as candidate for drought tolerance in tomato. These results suggest that drought and radiation affect differentially over activity of antioxidant enzymes in wild tomatoes.

Acknowledgements: FONTAGRO FTG-8071/08.

PS73

STUDY OF POLLEN DEVELOPMENT IN RICE UNDER CONTROLLED CONDITIONS

Gabriel Donoso, Mario Paredes, Viviana Becerra.

gabriel.donoso@inia.cl

Centro de Biotecnología de los Alimentos (CBA), Centro Regional de Investigación Quilamapu,
Instituto de Investigaciones Agropecuarias, Chillán, Chile.

Rice is a plant with high sensitivity to cold at reproductive stage, specifically when pollen is in development (tetrad stage). With the aim to find a better methodology to determine the tetrad stage, different varieties from rice breeding program from INIA Chile were studied. Seeds of Ambar-INIA, Oro, Brillante-INIA and Diamante-INIA were germinated in a growth chamber for three days at 28°C. After, they were transplanted into a plastic pot of 10 L with rice soil (Vertisol). Rice Plants were grown under controlled conditions at a PPFD of 300 mmol m⁻² s⁻¹, 28°C light / 22°C night and photoperiod of 14 h light / 10 h night. After 120 days, approximately, panicles of each plant were sampled with different auricular distances and were fixed in ethanol and acetic acid (3:1) at room temperature. Diameter of pollen grain, spikelet size and anther size were measured using ImageJ 1.46r software. The auricular distances showed a better correlation between pollen sizes with pollen phenology and allow us to find the tetrad stage to future cold tolerance treatment.

Acknowledgements: Grant FONDEF D10I1183

PS74

LOW TEMPERATURE FLUORESCENCE EMISSION FROM HYDRATED, DESICCATED AND REHYDRATED FRONDS OF TWO HYMENOPHYLLACEAE SPECIES (PTERIDOPHYTE)

Alejandra Flores-Bavestrello¹, Marianna Król², Alex Ivanov², Norman Hüner²,
Luis Corcuera¹, León Bravo^{3,4}

afloresb@udec.cl

¹Departamento de Botánica, Facultad de Ciencias Naturales y Oceanográficas, Universidad de Concepción, Chile. ²Department of Biology and The Biotron, Western University, London, Ontario, Canadá. ³Departamento de Ciencias Agronómicas y Recursos Naturales, Facultad de Ciencias Agronómicas y Forestales, Universidad de La Frontera, Chile. ⁴Center of Plant, Soil Interaction and Natural Resources Biotechnology, Scientific and Technological Bioresource Nucleus. Universidad de La Frontera.

Hymenophyllaceae is a desiccation tolerant family of Pteridophytes which inhabit shady and humid forests. Its species are poikilohydric epiphytes characterized by a mostly single-cell thick lamina and lack of cuticle, differentiated epidermis, and stomata. Since desiccation events usually occur during the day, it is likely that a combination of desiccation with a high light intensity occurs. Even in plants that inhabit zones with low light intensities, they can be subjected to high light intensities due to sunflecks inside the understory. The aim of this work was to study the energy distribution between PSII and PSI in two Hymenophyllaceae species (*Hymenophyllum dentatum* and *Hymenoglossum cruentum*) with contrasting vertical distribution during a desiccation and rehydration process. Low temperature fluorescence emission spectra from isolated thylakoids from hydrated, desiccated, and rehydrated fronds were taken at 77 K (-196°C). The spectra were recorded between 600 and 800 nm upon excitation at 436 nm. The emission peaks of LHCII, PSII and PSI became evident at 685, 695, and 730 nm, respectively. We observed that there were no significant variations in PSI/LHCII, PSI/PSII and LHCII/PSII parameters during the desiccation and rehydration process. It is concluded that there are minimal changes in energy distribution between PSII and PSI during desiccation in *H. dentatum* and *H. cruentum*.

Acknowledgements: FONDECYT N°1120964 (LB), CONICYT and MECESUP UCO 0708 fellowships to AF-B, Katalapi Park. NH is grateful for the financial support from NSERC, CFI and CRC research program.

PS75

**DEEP-SHADE ARCHITECTURE AND CROWN CARBON BALANCE OF SPROUTS AND
SAPLINGS OF *EUCRYPHIA CORDIFOLIA***

Antonio Escandón¹, Susana Paula², Rafael Coopman³, Roke Rojas³, Luis Corcuera¹

aescandon@udec.cl

¹Laboratorio de Fisiología Vegetal, Facultad de Ciencias Naturales y Oceanográficas, Universidad de Concepción, Concepción, Chile. ²Instituto de Ciencias Ambientales y Evolutivas, Facultad de Ciencias, Universidad Austral de Chile, Valdivia, Chile. ³LECOB, Instituto de Conservación, Biodiversidad y Territorio, Universidad Austral de Chile, Valdivia, Chile.

Light drives radical changes in plant architecture and leaf photosynthetic performance, especially in shady environments as those occurring in secondary temperate rainforests. In these ecosystems, many plants recruit from both seed and sprouts, being the latter a strategy to expand the regeneration niche. However, little is known about the physiological traits that allow sprouts to survive in shady microsites as well to colonize more open sites when compared with saplings. Here, we propose that the combination of gas exchange at leaf level and shoot architecture of sprouts determines a higher photosynthetic performance at crown level compared with saplings at low light availability. To test this hypothesis, we sampled *Eucryphia cordifolia* root suckers (sprouts) and saplings in a secondary temperate rainforest. Canopy openness (CO) and light contribution were calculated over each recruit. Gas exchange was measured in young fully expanded leaves. Plant architecture was captured by digitalization. Daily crown (DC) gas exchange was estimated according to the fitted light response curves, considering total leaf area and midday PPFD at 5% of CO. Sprouts architecture showed a higher crown density ($\text{m}^{-2} \text{m}^{-2}$; $\text{mean} \pm \text{S.E.} = 0.19 \pm 0.01$; $P < 0.001$) and self-shading ($\text{m}^{-2} \text{m}^{-2}$; $\text{mean} \pm \text{S.E.} = 0.15 \pm 0.01$; $P = 0.004$), therefore lower light interception efficiency (STAR, $\text{m}^{-2} \text{m}^{-2}$; $\text{mean} \pm \text{S.E.} = 0.42 \pm 0.003$; $P = 0.005$) than saplings ($\text{mean} \pm \text{S.E.} = 0.14 \pm 0.01$; 0.12 ± 0.01 ; and 0.44 ± 0.003 , respectively). However, sprouts showed higher carbon balance than saplings especially during cloudy days ($\text{mean} \pm \text{S.E.} = 0.92 \pm 0.11$ and 0.59 ± 0.09 , respectively; $P = 0.01$). Thus, the higher frequency of clonal growth in shady environments could be driven by higher photosynthetic performance than saplings given by their architecture and gas exchange capabilities.

Acknowledgments: FONDECYT 1110661 and Katalapi Park.

PS76

**DESICCATION TOLERANCE OF FILMY FERNS IS MEDIATED BY A CONSTITUTIVE
AND NOT INDUCIBLE CELLULAR MECHANISMS**

Marcelo Garcés Cea¹, Stephan Claverol², León Bravo¹

marcelogarcեսcea@gmail.com

¹Departamento de Recursos Naturales. Facultad de Ciencias Agronómicas y Forestales.
Universidad de la Frontera, Temuco, Chile. ²Pôle Protéomique, Plateforme Génomique
Fonctionnelle Bordeaux, Université V.Segalen Bordeaux 2, Bordeaux, France.

The Hymenophyllaceae Link is a primitive family within the Filicopsidae. One of the most outstanding features of this family of ferns is the presence of fronds with one or few cell layers (hence their name of filmy ferns) and therefore the absence of stomata. *Hymenophyllum caudiculatum* and *Hymenophyllum dentatum* are able to lose more than 82% of their water content, remain dry for some time and survive upon rehydration. The aim of this work was to understand if the adaptative strategy of Hymenophyllaceae for desiccation tolerance is constitutive or inducible. Chlorophyll fluorescence was used to monitor recovery in desiccation experiments, total sugar content to measure compatible solutes accumulation, and high resolution 2-DE to analyze proteome variation during a dehydration-rehydration cycle. For *H caudiculatum* 731 spots were identified, 112 spots showed significative differences between the hydration levels, only 17 from hydrated to dehydrated and 102 upon rehydration. For *H dentatum* 688 spots were identified, 70 spots showed significative differences between the hydration levels, only 26 from hydrated to dehydrated and 44 upon rehydration. Out of the 89 spots analyzed by MS/MS 83 were identified. Most of the identified proteins species were hypothetical proteins (47%), play a role either in photosynthesis (34.9%), Defense (6.0%) and carbohydrate metabolism (4.8%). A summary of the identified proteins, their putative functions and accumulation in hydrated or dehydrated states are present.

PS77

WATER STRESS INDUCED EFFECTS ON CARBON PARTITIONING IN GRAPEVINES

Nicolás Ríos, Luis Villalobos, Nicolás Franck, Claudio Pastenes

nicolas.rios@ug.uchile.cl

Facultad de Ciencias Agronómicas, Universidad de Chile.

Water stress, a common practice in vitiviniculture, promotes enological quality in red wine grapes, but reduces photosynthesis and the capacity for carbon partitioning; the latter, yet to be better elucidated on a whole plant system in field conditions. The present study aimed to simultaneously assess a) the effect of water stress on the non-structural carbon concentrations in leaves along the day and season, and b) on the berry growth kinetics and sugar concentration along the season in *Vitis vinifera* Carménère. The irrigation regimes were 0.8 mmha⁻¹h⁻¹ (T1), 1.6 mmha⁻¹h⁻¹ (T2), 3.2 mmha⁻¹h⁻¹ (T3) and 4.8 mmha⁻¹h⁻¹ (T4). The midday stem water potential were, on average -1,1MPa, -1MPa, -0,9MPa and -0,83MPa, respectively in the season. Significant differences occurred in sucrose and starch concentration in leaves, depending on the East or West side of the canopy, with higher contents for the more irrigated plants. Sugar import in berries was hastened, from veraison, in T2 and T3 at 5 days after *veraison* (DAV), but with no significant differences in sugar content later on. As for T1, the extremely depressed photosynthetic capacity limited the well-known impact of water stress on the hastening of sugar unloading at the berries site, resulting in similar sugar accumulation kinetic than T4. Sugar content, as per weight berry basis, resulted to be similar between treatments even though, due to the lower berry sizes in berries from the more stressed plants, lower sugar content was needed in such treatments. 20 (DAV), all the treatments reached the final sugar concentration in berries, inducing an increase in the starch content in leaves, suggesting a feedback regulation of sucrose synthesis depending on the sucrose unloading activity in clusters.

Acknowledgements: Fondecyt N° 1110193.

PS78

**INHERITANCE AND DISSECTION OF THE FRUIT TEXTURE COMPLEX TRAIT
IN PEACH USING DEEP PHENOTYPING APPROACH**

Alejandra Cifuentes-Esquivel^{1,2}, Miguel Rubilar², Claudio Meneses², Rodrigo Infante¹.

alej.cifuentes@gmail.com

¹Universidad de Chile, Facultad de Ciencias Agronómicas, La Pintana, Santiago, Chile;

²Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello, Santiago, Chile.

Peach fruit quality is one of the main objectives for breeding, and texture is one of the most important factors that define fruit quality and it influence in the consumer appreciation. Texture is a complex trait that depends on cellular structure and how this responds to forces, i.e. is dependent on the anatomical properties of the primary cell wall and on the cell turgor. Until now, peach texture has been related solely with firmness, but the evaluation of the different components of texture is a need for a more precise study of the phenotype. The aim of this work was to identify the components of fruit texture associate to melting and non-melting flesh types. Thus, a population of 100 individuals derived from the selfing an offspring of the cross of a melting and a non-melting variety were evaluated. The texture components measured through a TA-XTplus texture analyzer during 5 days since harvest were determined on all the individuals of the population. The curve force/ deformation for each individual were used to determine the maximum force, the work component, the Young module, or elasticity and the number of peaks. It was determined that the normal distribution doesn't exist for any of the parameters evaluated for texture. We also evaluated the time evolution using a linear regression analysis. Thus, we identified five groups related to the rate of softening, including Ross and Venus. With this approach we predicted the type of flesh pulp for individuals in the segregating population and we generated a new classification for melting and non-melting fruit. This information will support the study of physiological changes that occur during ripening, which are involved in the complex trait of texture.

Acknowledgements: Conicyt fellowship D-21120635 to ACE, Innova Corfo 09PMG-7240 and Fondecyt 1130198.

PS79

**COLD-REGULATED PROTEINS IN THE EXTREMOPHILE HAIR GRASS
DESCHAMPSIA ANTARCTICA DESV.**

Mario Díaz¹, Roxana Velásquez¹, Claudia Rabert^{1,2}, Ana Gutiérrez^{1,2}, Alejandra Sandoval^{1,2}

m.diaz18@ufromail.cl

¹Laboratorio de Fisiología y Biología Molecular Vegetal, Facultad de Ciencias Agropecuarias y Forestales. ²Center of Plant, Soil Interaction and Natural Resources Biotechnology, Scientific and Technological Bioresource Nucleus, Universidad de La Frontera, Temuco, Chile.

Temperature-induced stress has important implications for agriculture. Therefore, considerable efforts have been directed to understand the effects of temperature stress and adaptation to temperature in plants. Since temperature is a major climatic factor, more research is needed on inheritable characteristics for temperature stress resistance and on the mechanisms to increase extreme temperature tolerance. *Deschampsia antarctica* Desv. is the only monocot that thrives the harsh conditions of the Antarctic Peninsula and represents an invaluable resource for the identification of cold tolerance related genes. In order to identify proteins regulated by low temperature, we have initiated a detailed analysis of their expression. The objective of this work was to characterize cold regulated proteins in *Deschampsia antarctica* Desv. The soluble proteins were separated by two-dimensional polyacrylamide gel electrophoresis (2D-PAGE), an efficient method for the separation of complex protein mixtures. We found differences in the electrophoretic pattern of control and cold-treatment plants. The native Antarctic vegetation certainly must have one or various mechanisms that allow the maintenance of metabolism at low temperature during the Antarctic summer (growing season) and survival during the winter. The proteins could be relevant to understand the mechanisms by which this extremophile survives in its environment and contribute to the development of biotechnology in Antarctic species.

Acknowledgements: INACH 01- 03-Part II, DIUFRO DI13-0048.

**CHARACTERIZATION OF PHOTOSYNTHETIC RESPONSE OF TWO
PEACH VARIETIES TO DIFFERENT LIGHT INTENSITIES.**

Diego Andrade¹, Maria Paz Covarrubias¹, Gianfranco Benedetto¹, Eduardo Gusmão Pereira²,
Andrea Miyasaka Almeida¹.

di.andrade@uandresbello.edu

¹FONDAP Center for Genome Regulation, Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello, Santiago, Chile. ²Campus Florestal, Universidade Federal de Viçosa, Brazil.

The fruit of *Prunus persica* is highly demanded worldwide, being Chile the main exporter of peaches and nectarines in the southern hemisphere. To achieve a good quality and a maximized fruit yield, it is essential that the tree increases its photosynthetic rate to provide carbohydrates to the growing fruit. The objective of this study was to characterize the photosynthetic response of two peach varieties to different light intensities. The assays were performed in a nursery located in Malloa, VI region in Chile. The early harvest variety Magique and the late harvest variety Red Pearl were used. Fluorescence parameters were analyzed with the Fluorpen 100 equipment. It was obtained data of effective quantum yield of the photosystem II (ϕ_{II}) as well as the quantum yield of non-regulated (ϕ_{NO}) and regulated energy dissipation (ϕ_{NPQ}). The apparent electron transport rate (ETR), the non-photochemical (NPQ) and photochemical (qL) quenching were also evaluated. The two varieties showed high capacity to cope with excess light, once the values of ϕ_{NPQ} and NPQ increased with light intensities above $300 \mu\text{mol m}^{-2} \text{s}^{-1}$. The ϕ_{II} and qL were high in lower irradiances and diminishes when the light intensity was increased, in an inversely proportional way to ϕ_{NPQ} . At the same time ϕ_{NO} reaches a plateau in the same point ($300 \mu\text{mol.m}^{-2} \text{s}^{-1}$). The ETR was saturated also after light exposition above $300 \mu\text{mol.m}^{-2} \text{s}^{-1}$ and decays at $1000 \mu\text{mol m}^{-2} \text{s}^{-1}$, indicating a dynamic photoinhibition. In conclusion the photosynthetic response of both varieties were saturated by light intensities above $300 \mu\text{mol.m}^{-2} \text{s}^{-1}$ and the plants showed high capacity to quench excess energy above the saturation point.

Acknowledgements: FONDECYT 1130197, FONDAP CRG 15070009, Basal PFB-16, "El Tambo" Nursery.

PS81

**QUALITATIVE MEASSUREMENT OF LEAF AND FRUIT METABOLITES FROM TWO
PRUNUS PERSICA VARIETIES BY HPAE-PAD**

María Paz Covarrubias¹, Gianfranco Benedetto¹, María Luisa Valenzuela²,
Andréa Miyasaka Almeida¹

mariapaz.covarrubias@gmail.com

¹FONDAP Center for Genome Regulation, Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello, Santiago, Chile. ²Departamento de Ciencias Químicas, Facultad de Ciencias Exactas, Universidad Andres Bello, Santiago, Chile.

Nectarine [*Prunus persica* (L.) Batsch] is a stone fruit classified as a drupe, which means that possess the seed enclosed in a lignified endocarp, a fleshy mesocarp and a thin exocarp. This is a much appreciated fruit and the quality parameters defined by the consumers are size, juiciness, taste, aroma and color. The taste is determined by a balance between soluble sugars present in the mesocarp and the predominant organic acids. Biomass production and metabolites accumulation by fruits occur during the different developmental stages and depend on photosynthesis and carbon exportation by source leaves. Carbon supply to fruits can be potentiated through field practices of thinning (removal of flowers and fruits) that lead to a change in the source-sink balance favoring the development of fruits. It is well known that thinning leads to fruits with increased size, but it is not known how this practice could influence fruit quality in terms of metabolite composition. In this work we analyzed metabolite composition of fruit and leaf samples from two nectarine varieties at different development stages and sampled from trees submitted to different thinning treatments. Samples were processed with the objective of extracting water soluble sugars which were measured by high performance anion exchange liquid chromatography (HPAE-PAD). It could be seen that sucrose was the major sugar present in ripened fruit while in leaf accumulation of myo-inositol predominated. Other sugars, aminoacids and organic acids were also detected on both organs. We expected that analysis of 'metabolic fingerprints' of leaves and fruits would correlate to fruit quality traits and used as molecular predictor.

Acknowledgments: FONDECYT 1130197, FONDAP CRG 15070009, Basal PFB-16 , "El Tambo" Nursery.

PS82

EFFECT OF ROOTSTOCK ON GROWTH, PRODUCTIVITY AND FRUIT QUALITY OF CHERRY TOMATO

Juan Pablo Martínez^{1,3}, Luis Salinas¹, Alejandro Antúnez², Aníbal Ayala¹, Lida Fuentes^{3,1}, Stanley Lutts⁴, Francisco Pérez-Alfocea⁵

jpmartinez@inia.cl

¹Instituto de Investigaciones Agropecuarias, INIA-La Cruz, La Cruz. ²Instituto de Investigaciones Agropecuarias, INIA-La Platina, Santiago ³Centro Regional de Estudios en Alimentación y Salud (CREAS), Valparaíso, Chile; ⁴Université catholique de Louvain, Laboratoire d'Ecologie des Grandes Culture, Louvain-la-Neuve, Belgium. ⁵CEBAS-CSIC, Departamento Nutrición Vegetal, Murcia, España.

The study responds to the demand of tomato owners to mitigate the problem of a low productivity and fruit quality under greenhouse and soil conditions in Valparaíso Region. The study of rootstock has become a modern tool to improve the productivity and fruit quality of cultivated species. The trial was conducted to determine the effect of three rootstocks on growth, productivity and fruit quality in cherry tomato (*Solanum lycopersicum* var. *cerasiforme* L.). At 30 days after sowing, tomato plantings were grafted on three rootstocks (P1, P2 and P3) and fourteen after, grafted plantings were transferred into a greenhouse and transplanted individually. Grafted tomato plants were grown at a population density of 2.5 plants m⁻² under intermediate levels of nematodes (350 nematodes per 250 cm³) in the soil. The experimental design contained three treatments (rootstock), with six blocks localized by free random. Plant growth (fresh and dry aerial mass) and productivity (kg/m² and harvest index) were recorded while compatibility (stem diameter ratio superior to inferior) and fruit quality (size, colour, water content and pulp pressure). Results showed that the P1 rootstock presented a greater compatibility and harvest index than P1 and P2. On the other hand, P1 and P2 showed higher values of fresh and dry aerial weight than P3 while productivity levels were not affected significantly. P1 and P2 grafted plants showed changes in fruit quality (water content, pressure of fruit and diameter) while P3 grafted plants were less affected under stress conditions. Results suggested that the adequate variety-rootstock combination is a determinant factor to obtain a greater productivity and fruit quality in zones when tomato is cultivated, opening interesting opportunities for use traditional and new commercial varieties under biotic and abiotic stress conditions.

Acknowledgments: Proyecto INIA-CSIC N° 501736-70 and Proyecto R12C1001, CREAS, CONICYT-Regional GORE Valparaíso.

PS83

**EFFECT OF CALCIUM APPLICATIONS ON BERRY FIRMNESS DURING *IN VITRO*
GROWTH OF TABLE GRAPES CV. THOMPSON SEEDLESS**

Tamara Peredo¹, Iván Balic¹, Joaquín Delgado¹, Ariel Orellana¹, Humberto Prieto², Reinaldo Campos-Vargas¹

tamara.peredo@gmail.com

¹Universidad Andrés Bello, Facultad Ciencias Biológicas, Centro de Biotecnología Vegetal Santiago, Chile. ²Instituto de Investigaciones Agropecuarias, INIA La Platina, Santiago, Chile.

The loss of firmness in table grape berries is one of the main quality problems affecting the crunchiness perception by consumers. Thus, it is important to Chilean economy the development of strategies to reduce this problem. In different fruits and plant models, calcium has been strongly associated with tissue firmness due to its role in the formation of ionic bridges between homogalacturonan chains in cell wall pectins (the "egg-box" model). Foliar and soil calcium application is one of the most commonly used practices for growers in order to improve firmness of grape berries. However, until now there is not conclusive evidences regard their effectiveness about calcium mobility into the berry and its impact on texture. With the purpose of enhance calcium delivery we evaluated the effect of 0 to 5 g/L of calcium in *vitro* culture of young berries (3-4 mm of diameter) of Thompson Seedless variety. Results showed berry growth in all treatments with calcium but without significant differences. Berry firmness was determinate using a Texture Analyzer, and the data indicated a significant increase in this trait related with calcium concentration. Cell wall calcium determinations showed a correlation between firmness and calcium content. Our results support new insights in the relevance of calcium in texture parameters of table grape berries related with cell wall stability under berry development.

Acknowledgments: FONDECYT 1110406, UNAB DI-152-12-R, UNAB DI 415-13/I, IB Conicyt Doctoral Fellow.

IN VITRO MID-TERM STORAGE OF *FRAGARIA CHILOENSIS*

Gustavo Riveros, Gerardo Tapia, Marcela Alcorta

gf.riveros@gmail.com

INIA CRI Quilamapu, Laboratorio de Recursos Genéticos

Climate change, disease and pests, constitute a hazard to the preservation of genetic resources in the habitat where they grow. In vitro conservation of these genetic resources could help to keep these materials in time. The importance of *in vitro* conservation lies in the ability to propagate and maintain plants pathogen-free and also to maintain the genetic stability of an individual over time. The use of osmotic compounds in the medium and low temperatures have been successful methods to reduce explant growth, whereby in the present study we proceeded to develop in vitro mid-term storage studies in *Fragaria chiloensis*, because it is a native plant with commercial potential, that is reproduced vegetatively and therefore the *in vitro* storage is the more recommended conservation system. We proceeded to implement a methodology for in vitro explant introduction. We tested various types of explants such as apical meristem of rosette, and apical meristem of runners, also pretreatments with PVPP, activated charcoal or L-Cysteine to prevent oxidation. After performing the *in vitro* multiplication, a methodology for their conservation in the mid-term at 4°C was assessed. Rooted explants were used, their fresh weight was measured and the treatments were concentrations of mannitol at 0.1 M, 0.2 M, 0.3 M and 0.4 M. After 13 months at 4°C we proceeded to measure the increase in fresh weight and evaluate the development and survival of the explants. It was shown that higher concentrations of mannitol favored in retarding the plant growth, and it did not affect its viability or development once transferred into optimal conditions. Therefore the concentration of 0.4 M mannitol in the medium is appropriate for the preservation of *F. chiloensis* in the medium term.

PS85

**ALLELIC DIVERSITY OF CANDIDATE GENES IN RESPONSE TO HYPOXIC
STRESS IN *PRUNUS*.**

Claudia Gómez¹, María José Arismendi¹, Patricio Hinrichsen^{1,2}, Rubén Almada¹, Paula Pimentel¹,
Manuel Pinto^{1,2}, Rodrigo Contreras¹, Boris Sagredo¹

cgomez@ceaf.cl

¹Centro de Estudios Avanzados en Fruticultura (CEAF) INIA CRI Rayentué, Rengo, Chile;

²INIA CRI La Platina, Santiago, Chile.

In crops such as stone-fruit (*Prunus* sp.) deficiency of oxygen (O₂) in roots can adversely affect the performance and productivity of orchards. This phenomenon known as root hypoxia is exacerbated under situations of flooding and/or soils with drainage problems. The majority of *Prunus* species are sensitive to these conditions, while the subgenus *Prunophora*, mainly Myrobalan plums, exhibit good tolerance in soils with deficient O₂. It has been suggested that an important component of the phenotypic variation for this trait among *Prunus* populations, would be associated with allelic diversity of genes involved in the response to hypoxic conditions, responsible to maintain the energy homeostasis in roots plants. This study analyzed the allelic diversity of 11 candidate genes within a population of 22 varieties of *Prunus*, which are used as rootstocks. These genes encode for *LDH*, *ADH* and *PDC* from the fermentative pathway, 1 plant-hemoglobine, 1 antioxidant, 1 aquaporin and 1 ethylene transcription factor. Gene sequences of the candidate genes were obtained from the peach genome (*Prunus persica*) available at Phytozome platform, while SSR motif were evaluated from intronic sequences using the program SSRIT. The primers were designed using the PrimerPremiere software. These primers were used for SSR assays, using genomic DNA from young leaves as template amplified by TD-PCR. The SSR-amplified fragments were revised through a system based in electrophoresis capillary, Fragment AnalyzerTM, which provides the information necessary analysis of allelic diversity, by PROSize software. A dendrogram was generated using Dice and UPGMA distance, where clusters were able to differentiate into three subgenera studied by allelic diversity of candidate genes. In addition, a Venn diagram, unique alleles were identified in *Prunophora*, especially those present in Myrobalan plum as PDC1_184 and PIP2B_147, alleles which could explain the tolerance to hypoxia.

Acknowledgments: FONDECYT N°1121117 and CEAF_R08I1001.

PS86

**CHARACTERIZATION OF DIFFERENT SOURCES OF *PITAVIA PUNCTATA* MOL.,
A THREATENED SPECIES OF SOUTH-CENTRAL CHILE**

Helen Spielman, Carola Venegas

helspielman@udec.cl

Universidad de Concepción, Concepción, Chile.

In Chile, the loss and habitat fragmentation, are recognized by one of the most important threat that occurs to the different species of flora and fauna. The genetics analysis allow to study the effect of the fragmentation process in the reduction of the gene flow in populations and the genetic diversity within and between populations, moreover can also set priority species for conservation, defining evolutionarily significant units, among other actions. *Pitavia punctata* Mol., is one of the endemic threatened species without genetic information available, that has been subjected to high and consistent levels of fragmentation. A germination analysis says that would exist genetic difference between the populations. The aim of this study was perform an analysis of six backgrounds of *P. punctata* to provide information to conservation actions. The vegetal sample was collected from six populations of *P. punctata*. A habitat description was performed in function of variables which can influence in the survival, establishment and on the species current status. DNA was extracted from the phellogen, and the genetic diversity analysis used two molecular markers (AFLP and ISSR). The seeds of *P. punctata* were subjected to a cold stratification treatment and were sown. The germination capacity and the germination energy were measured. The increase of human population, the replacement of native forest by urban areas, forest plantations and agricultural land are the greatest threats for the species. *P. punctata* had a good germination capacity, so the seed reproduction is a good propagation method. Though, in situ had a low level of regeneration, so it is expected that genetic analysis show some problem. The study about a threatened species like *P. punctata* should consider an analysis of their attributes in all the levels to provide integral recommendations that allow the species persistence in the long term.

PS87

**ALLELIC IDENTIFICATION OF CANDIDATE GENES OF RESPONSE TO HYPOXIA
IN POLLEN OF *PRUNUS* SP. USING INTRON-SSR**

**Rodrigo Contreras¹, Simón Solís^{1,2}, Ruben Almada¹, Paula Pimentel¹,
Felipe Gainza¹, Boris Sagredo¹**

rcontreras@ceaf.cl

¹Centro de Estudios Avanzados en Fruticultura (CEAF), INIA CRI Rayentué Rengo-Chile.

²Universidad Andrés Bello, Santiago, Chile.

In breeding of *Prunus* sp., the pollen exchange is the simpler and faster alternative for germplasm transfer. Among their advantages, it is easy to store and ship to anywhere in the world, with less phytosanitary restrictions. The Centro de Estudios Avanzados en Fruticultura (CEAF) has imported pollen of different *Prunus* sp, from several regions of Europe, for the development of new rootstocks. Some of these pollens belong to genotypes that had been systematically evaluated for more than 15 years, for tolerance to hypoxia (oxygen deficiency) at the root level, which is one of the most important traits for the new *Prunus* rootstocks required for south-central regions of Chile. We used groups of genotypes with contrasting responses to root hypoxia for identifying allelic variants in candidate genes associated to hypoxia response, which were previously identified by RNA-seq. We analyzed the allelic diversity of 24 genes involved in tolerance to hypoxia within a panel of 32 genotypes, classified as highly (6), moderate (9), low (6) and very low (11) tolerant to root hypoxia. The complete DNA sequences of the genes under study were obtained from the peach genome available at Phytozome platform, while SSR were generated from their respective intron sequences using the program SSRIT. The SSR allele size was assessed in a Fragment AnalyzerTM. From an overall of 205 alleles that were found, 16 belong exclusively to the subgenus *Prunophora*, 18 to *Cerasus*, and 53 to *Amygdalus*. One allele (PIP2B_156) that belong to *Prunophora* subgenus was shared among all genotypes classified as highly and moderate tolerant to hypoxia. On the other hand, seven alleles were shared among susceptible genotypes that belong to the *Amygdalus* subgenus. The association of these alleles with tolerance to hypoxia in roots will be evaluated in segregant progenies of *Prunus* sp.

Acknowledgments: FONDECYT N°1121117 and CEAF_R08I1001.

PROTOPLAST FUSION IN TOMATO

Gustavo Riveros^{1,2}, Gerardo Tapia², Darcy Rios¹

gf.riveros@gmail.com

¹Facultad de Ciencias Forestales, Universidad de Concepción.

²INIA CRI Quilamapu, Laboratorio de Recursos Genéticos

The tomato (*Solanum lycopersicum* Mill) is the most widely grown vegetable in the world, and its demand is increasing. In Chile we can find wild relatives species such as *S. peruvianum* Mill. This species has characteristics of agronomic interest, such as pathogens resistance and abiotic stress tolerance. Studies of this kind lies in the ability of transferring these characters to commercial tomato. A methodology that provides this possibility is somatic hybridization that is the somatic cells fusion. For this reason, in this work we studied the technique stages that influence in the efficiency of the protoplast fusion. Therefore we proceeded to isolate intact protoplasts from leaves of both species. We achieved to adapt a purification methodology by using two sucrose gradients. Also we determine an appropriate electrofusion buffer which allowed the maintenance of protoplasts in a hypotonic medium. Besides, we standardized protoplasts alignment conditions as follows: 500 Vcm⁻¹ for 15 seconds for a concentration of 3.5×10^5 prot/ml and also we decrease the range of direct current which produced the most protoplast fusion, this range corresponded to two pulses of direct current between 3000 Vcm⁻¹-5000 Vcm⁻¹ for 40 seconds. As a conclusion, the electrofusion methodology is a technique that allows the fusion of protoplasts, but it is an extremely sensitive technique which requires an exhaustive standardization of each of the stages, from the donor plant status until further stages like ion concentration, prior to the isolation of protoplasts.

Acknowledgements: INNOVA BioBio

PS89

**BREEDING NEW CULTIVARS OF TABLE GRAPE RESISTANT TO POWDERY MILDEW BY
PYRAMIDING REN1 AND RUN1 RESISTANCE LOCI**

Mario Agurto^{1,2}, Rudolf Schlechter², Grace Armijo², Patricio Arce-Johnson².

meagurto@uc.cl

¹Programa de Doctorado en Ciencias de la Agricultura, Facultad de Agronomía e Ingeniería Forestal, Pontificia Universidad Católica de Chile. ²Departamento de Genética Molecular y Microbiología, Facultad de Ciencias Biológicas, Pontificia Universidad Católica de Chile.

The grapevine (*Vitis vinifera*) is the fruit species with the major national production, being Chile the world's largest exporter of table grapes. Powdery mildew is one of the most economically important diseases that attack the grapevine. The etiological agent is the biotrophic fungus *Erysiphe necator*, an obligate pathogen of the Vitaceae family that attacks all green tissues of the plant using its nutrients, reducing photosynthesis, affecting growth and causing important yield reduction. Its control implies both cultural and chemical techniques. However, nowadays there is a global trend toward decreased use of agrochemicals, becoming relevant the use of natural sources of resistance to grape powdery mildew in breeding programs. This work focuses on the development of potential new *V. vinifera* cultivars with natural resistance to powdery mildew. For this, Ren1 and Run1 dominant loci, responsible for this resistance, have been used. Ren1 and Run1 have been found in a North American species from Vitaceae family and a *Vitis vinifera* cultivar from Central Asia, respectively. We used these loci to develop new grapevine cultivars with resistance to powdery mildew by pyramiding resistance genes with the aim of providing a long lasting resistance. For this, we developed and selected four Ren1Run1 genotypes using six molecular markers that cosegregate with the resistance characteristic and two commercial table grape cultivars as parental lines used in controlled crosses. Progenies were obtained and assessed to direct *E. necator* infection by inoculating leaves. Several grape powdery mildew resistant segregants have been identified to date. These segregants constitute potential new powdery mildew resistant table grape cultivars and the next step is to evaluate these plants on different aspects of its defense response.

Acknowledgments: Programa de Mejoramiento Genético de la Vid - Consorcio Tecnológico de la Fruta S.A., Millennium Nucleus for Plant Functional Genomics and CONICYT Doctoral Fellowship.

PS90

MASSIVE PROPAGATION OF CHERRY GENOTYPES USING TEMPORARY IMMERSION SYSTEMS.

Luis Ortega⁴, Catalina Alvarez¹, Eduardo Tapia^{1,2}, Sebastián Godoy³, Humberto Prieto¹.

donzarpon@gmail.com

¹Instituto de Investigaciones Agropecuarias, CRI La Platina. ²Doctorado en Biotecnología PUCV/UTFSM. ³Departamento de Ing. en Biotecnología. UTCh INACAP.

⁴Doctorado en Biotecnología USACH

Chile is practically the only country in the Southern Hemisphere that has significant production of cherries fruit to supply the demands of Northern Hemisphere's in non-productive season. The cherry plantations represents almost 5% of total fruit crops production area of the country and 1.7 % of fruit? exports. *Prunus avium* plantations are located from Coquimbo to Aysén and with a harvest period between November to January, with low average yields, currently not exceeding 5 tons / ha. With these productive parameters, an increase in plant available biomass is a growing need for the sector. The clonal propagation of better rootstock and cultivars genotypes through temporary immersion systems (TIS) bioreactors is a very interesting technology, used worldwide and known to result in a higher plant multiplication yields. In a first stage, a 500 mL TIS was evaluated using explants with an average total weight of 2 g as starting material and weight increase in a 14 days period was measured. For Maxma 14? and Colt rootstocks 7.522 g and 7.416 g were obtained respectively, this was an important increase in productivity when compared to conventional *in vitro* culture values (3.322 g and 4.203 g respectively). In the second stage, a 2L TIS was used with the same 2 g as starting material. Two different culture media and immersion (length and number of runs) were evaluated. Van and Rainier genotypes were also included. The best conditions were obtained with culture medium II and 4 dips of 1 minute each. Using these parameters we obtained for Maxma 21 g, Colt 27.9 g, Van 35.8 g and Rainier 19.8 g. In conclusion, the incorporation of TIS platform for mass propagation of different cherry genotypes becomes efficient, productive and allows a continuous support to breeding programs, growers and producers of this fruit.

Acknowledgments: FONDEF G09I1008 and BIOFRUTALES.

PS91

FT-IR SPECTROSCOPY SUPPORTED BY PCA AND SIMCA ANALYSIS FOR THE STUDY OF EMBRYOGENIC POTENTIAL OF *PINUS RADIATA* D. DON CALLI.

Aileen Turner^{1,2}, Francisco Sepúlveda^{1,2}, Rodrigo Hasbún², Rosario Castillo³, Soraya Bravo¹.

aturner@udec.cl

¹Centro de "Biotecnología Gran Concepción", Facultad de Ciencias Biológicas, Universidad Andrés Bello. ²Laboratorio de Epigenética Vegetal, Facultad de Ciencias Forestales, Universidad de Concepción. ³Departamento de Análisis Instrumental, Facultad de Farmacia, Universidad de Concepción.

The loss of embryogenic potential and multiplication capacity of clonal lines propagated by somatic embryogenesis is one of the biggest disadvantages of this propagation method in forest species like *Pinus radiata*. These factors represent the most constraints to the application of intensive clonal forestry, reducing the competitiveness of Chilean forestry in the world. Previous results of our research group have shown remarkable differences between embryogenic and non-embryogenic calli, in terms of cellular composition and architecture, and in the quality and quantity of genomic DNA obtained after extraction. Aiming to develop a biotechnological tool that allows early detection of embryogenic potential of *Pinus radiata* calli, we analyzed by FT-IR (Fourier Transform Infra Red) spectroscopy, genomic DNA samples from embryogenic and non-embryogenic calli. Analysis showed marked differences in the profiles obtained for each category. We supplemented our research with the use of chemometric tools such as Principal Component Analysis (PCA) and a single independent modeling by class analogy (SIMCA). Thus, in this study we show that it is possible to establish a defined and robust separation between both categories, highlighting the potential of the resulting model to predict the category in a new sample.

Acknowledgements: CMPC Mininco, CMA BioBio.

PS92

**GENECTIC VARIABILITY IN SUSCEPTIBLE AND RESISTANT NICOSULFURON
POPULATIONS OF *SORGHUM HALEPENSE* (L) PERS**

Paloma Morales¹, Rodrigo Figueroa¹, Basilio Carrasco², Marlene Gebauer¹

pomorale@uc.cl

¹Departamento de Ciencias Vegetales, Facultad de Agronomía e Ingeniería Forestal, Pontificia Universidad Católica de Chile. ²Departamento de Fruticultura, Facultad de Agronomía e Ingeniería Forestal, Pontificia Universidad Católica de Chile.

Jonhson grass (*Sorghum halepense* (L) Pers.) is an allopolyploid weed that reproduces by self-pollination and also through vegetative propagation by rhizomes. The intensive use of acetohydroxyacid synthase (AHAS) inhibitors herbicides (Nicosulfuron) in maize fields, has accelerated the development of resistance in Jonhson grass (*Sorghum halepense* (L) Pers.). To establish relationships between AHAS inhibitor resistance and genetic variability of resistant and susceptible populations of Jonhson grass, a variability analysis was performed using SSR and ISSR molecular markers in 19 populations distributed in three regions of Chile (VI, VII, RM). In this investigation, 19 SSR isolated in *Sorghum bicolor* were amplified and validated. Finally, five SSR and two ISSR were used to characterize an average of 10 samples for each group. A total of 86 loci with 64.2 % polymorphism were observed in the 19 populations. Expected heterozygosity (H_e) within resistant populations (R) varied from 0.136 to 0.247, while the susceptible populations (S) ranged from 0.136 to 0.250. The mean Shannon index of susceptible populations was 0.22 and for resistance populations was 0.20. A higher degree of genetic differentiation was found among susceptible populations compared with Nicosulfuron-resistant populations, observing a 29% of genetic variation among S population with a $Ph_{ipt}=0.29$. There were found a 17 % of variation among R populations with a $Ph_{ipt}=0.22$. Those results could be explained by selection pressures due the herbicide application.

Acknowledgments: CONICYT, FONDECYT N°110535 for financial support.

PS93

**DEVELOPING A CUTTING PROTOCOL FOR PROPAGATION OF *PROSOPIS FLEXUOSA*,
AN ENDANGERED SPECIE FROM THE NORTH OF CHILE**

Marta Vargas¹, Maricela Rojas¹, Elda Jofré¹, Alejandra Araya¹, Ana María Vásquez¹,
Carlos Navarrete², Cristian Ibáñez¹

cibanez@userena.cl

¹Departamento de Biología, Facultad de Ciencias, Universidad de La Serena, Chile.

²Departamento de Matemáticas, Facultad de Ciencias, Universidad de La Serena, Chile

Since pre-Hispanic times, the Chilean species of the genus *Prosopis* spp have been useful for human settlement in the desert. *Prosopis flexuosa* is a tree legume (family Fabaceae) which is useful for forage, for medicinal purpose, for food (flour), for fuel and for dyeing material. Its ecological value has been recognized in such an environment. This tree is an endangered species and therefore it is important to generate successful methods of propagation. The aim of this study was to evaluate the effect of three concentrations of indole butyric acid (IBA; $T_0 = 0\text{ppm}$, $T_1 = 2.500\text{ppm}$, $T_2 = 7.500\text{ppm}$) on asexual propagation of *P. flexuosa*. For this, we set in closed glass jars three cuttings with their base embedded in IBA which we then put in a substrate of sand-vermiculite (2:1), with a total of 10 replications per treatment. After two months, we evaluated the rooting of the cuttings, the number of roots generated and the root dry mass. There were significant differences in the rooting of cuttings ($X^2 = 7.081$, $P = 0.029$), for T_0 , we obtained 57.1% rooting while T_1 reached 84.6% and T_2 manage 80.8%. Regarding generated roots and root dry weight, MANOVA analysis confirmed significant differences between our treatments (Pillai 2.27 = 0.24, $n = 10$, $p = 0.02$), this being the number of roots which developed ($p = 0.006$). We concluded that the addition of indole butyric acid stimulated both the rooting of cuttings and the number of roots generated in *P. flexuosa*. These results are a contribution to the knowledge of propagation strategies for this endangered tree with a high ecological value for the arid zone of Chile.

Acknowledgments: Fondo de Investigación de Bosque Nativo, proyecto CONAF 037/2011.

PS94

**MICROMORPHOLOGICAL ANALYSIS OF PRO-EMBRYOGENIC STRUCTURES FROM
COTYLEDONS OF MATURE GERMINATED *EUCALYPTUS GLOBULUS* LABILL. SEEDS**

Natalia Avilés¹, Fabiola Avilés², Manuel Sánchez¹, Darcy Ríos¹.

¹ Laboratorio de Cultivo de Tejidos Vegetales, Facultad de Ciencias Forestales y Centro de Biotecnología, Universidad de Concepción. Chile. ²Laboratorio de Biotecnología, BIOFOREST, Concepción, Chile.

drios@udec.cl

Eucalyptus globulus plays an important role in an economic as a commercial area, however, the *Eucalyptus* genus, has a limitation in their asexual propagation, specially, via cuttings, making impossible to establish successful clonal plantations. The main problem, is their low percentage of rooting, therefore, leaving a large number of genotypes selected out of production due to low rooting capacity. By this reason, the somatic embryogenesis technique applied to *E. globulus*, would bring benefits in terms of time to clones generation, reduced physical space, and most importantly, to overcome the low rooting capacity. In this study we evaluate the induction of embryogenic callus, which it used to germinate cotyledons. The culture medium used was MS, where 2 treatments were evaluated: MS without hormones (T0) and MS + 3 mgL⁻¹ Picloram (T1). In this last treatment the presence of pro-embryogenic structures was observed. Treatment T0 showed no reaction. After 25 days of culture, micromorphological analysis was performed by disintegrations-smear, which allowed us to determine the existence of meristematic nests characteristic of embryogenic callus, corresponding to small cells with prominent nuclei, surrounded by large parenchyma cells which have a prominent central vacuole. These cells present extremely thin walls, which could indicate that when they are exposed to a hormonal inducing agent they could be potentially differentiated into secondary type meristem cells.

Acknowledgments: Proyecto 12.300-EM.TES INNOVA BIOBIO

PS95

CLONING AND MICROPROPAGATION OF CURUBA (*PASSIFLORA MOLLISSIMA* BAILEY) FROM LEAF SOMATIC EMBRYOS

Maria del Pilar Acosta Zambrano¹, Carolina Andrea Vega²

pilaracosta@unach.cl

¹Departamento de Ciencias Básicas. Universidad Adventista de Chile.

²Pedagogía en Biología y Ciencias Naturales. Universidad Adventista de Chile.

The curuba (*Passiflora mollissima* Bailey) is a native fruit of the Andean area. It is characterized for being a perennial, herbaceous plant, which grows in a wild way. In the food industry it is used like raw material for the preparation of sherbets (iced drinks), jams, ice creams, juices and natural flavoring. It presents a high nutritional value, being a valuable source ascorbic acid, niacin and vitamins, in addition, it possess medicinal properties, such as: anti-inflammatory and antihistamine activities. Due to the difficulties that its spread presents in natural conditions, *in vitro* propagation tests were carried out for its multiplication from somatic embryos originated from leaves. It was designed a disinfection protocol for the seeds and then they were introduced in Murashige and Skoog (MS) medium supplemented with 1 mg/l of Gibberellin to induce the germination. For the multiplication *in vitro* explants were placed in MS medium supplemented with 2 mg/l of Benzylaminopurine (BAP) and 1 mg/l of Acid Naftalenoacetic (NAA). The most vigorous leaves were selected for inoculation in MS and Woody Plant Medium (WPM). From the third week embryos formation was observed. At week six the seedlings, formed from these embryos, were inoculated in WPM supplemented with 1 mg/l of BAP. For the rooting period a medium of WPM mg/l of 2-ip was used. Finally they were acclimatized in a peat soil-vermiculite mixture for one month previous to their transference to containers with soil until subsequent phytochemical studies.

PS96

**GENETIC DIVERSITY AND PATERNITY TEST IN JAPANESE PLUM
(*PRUNUS SALICINA* LINDL.)**

Marco Meneses¹, Paloma Morales¹, Máximo González¹, Herman Silva², Marlene Gebauer¹, Basilio Carrasco¹

mmmenese@uc.cl

¹Facultad de Agronomía e Ingeniería Forestal. Pontificia Universidad Católica de Chile, Santiago, Chile. ²Universidad de Chile, Facultad de Ciencias Agronómicas, Departamento de Producción Agrícola, Laboratorio de Genómica Funcional and Bioinformática, Santiago, Chile.

The Japanese plum is a diploid, alogamous and self-incompatible species. The self-incompatibility ensures outcrossing, reduces the inbreeding rate and maintaining high level of genetic variability within population. The genetic characterization of parental lines and its progenies is a useful tool for assess the progress of breeding programs. That allows verifying the remanents level of genetic diversity and the affectivity of breeding cycles. In this work a base population of 29 cultivars of Japanese plums was evaluated through 17 microsatellites. The results showed a high level of genetic diversity among cultivars and a high discriminate power with a low number of microsatellites. Then, the microsatellites allele frequencies allowed to carry out a paternity test to a population of 112 offspring developed in our breeding program. Microsatellites displayed a high discriminant power to verify the paternity assignment to each generated progeny. Moreover, this study allows analyzing the reproductive biology of Japanese plum.

Acknowledgements: CONICYT, FONDECYT/Regular N°1120261 and Programa de Mejoramiento Genético de Carozos, Consorcio Tecnológico de la Fruta SA, FIA/INNOVA.

PS97

**ASSOCIATION MAPPING OF SEED QUALITY TRAITS IN *BRASSICA NAPUS* L.
USING GWAS AND QTL CANDIDATE APPROACHES**

Humberto A. Gajardo^{1,2}, Benjamin Wittkop³, Rod J. Snowdon³, Maria L.
Federico¹ Iniguez-Luy¹

federico.iniguez@cgna.cl

¹Agriaquaculture Nutritional Genomic Center (CGNA), Genomics and Bioinformatics Unit, Chile. ²Programa de Magíster en Ciencias Vegetales, Universidad Austral de Chile. ³Department of Plant Breeding, IFZ Research Centre for Biosystems, Land Use and Nutrition, Justus Liebig University, Germany.

Single nucleotide polymorphisms (SNPs) are considered as the molecular marker of choice in plant and animals genetic studies. Recent low cost for their discovery and genotyping had favored their use for association mapping (AM). In this work, we evaluated both genome wide association study (GWAS) and QTL candidate approaches (cQTL) in order to search SNPs-seed quality traits associations in a diversity panel of *Brassica napus* composed of 89 winter lines. Six seed quality traits were evaluated in different localities. For GWAS 4025 SNPs distributed along the *B.napus* A and C genomes were genotyped using a 6K Illumina array platform. Moreover, for cQTL, 100 SNPs previously discovery in seeds quality QTL by sequence capture were genotyped using the KASPar assay. The effects of population structure and relative kinship of the lines tested were taken into account for both approaches. The population structure analysis was conducted by three different methodologies which suggested the presence of two subpopulations. Indeed, 82% of the lines present low relative kinship ranging from 0 to 0.1. For AM, we conducted a general lineal model (GLM+Q) using TASSEL, finding significant associations for four traits in GWAS: seed oil content (SOC), seed linolenic acid content (SLAC), seed glucosinolates content (SGC), seed hemicelluloses content (SHC), and for two traits in cQTL: SGC and SHC. When the relative kinship effect was taken into account, only SGC showed SNP-trait associations. The associated SNPs showed stability through the different localities and seasons evaluated. Additionally, the associated SNPs were mapped in, or close to previously reported QTL for SGC. These results suggest a good potential use for these markers to assist a breeding *B. napus* program in order to select lines with specific seed quality traits.

Acknowledgements: FONDECYT 1100732 and CONICYT REGIONAL GORE ARAUCA-NÍA/CGNA/R10C1001.

PS98

AMPLIFICATION OF THE LYCE 3A AND 3B GENES FOR GENETIC IMPROVEMENT OF DURUM WHEAT (*TRITICUM TURGIDUM* L.SSP. *DURUM*) USING THE TILLING METHOD

Daniela Richaud and Andrés Schwember

aschwember@uc.cl

Departamento de Ciencias Vegetales, Facultad de Agronomía e Ingeniería Forestal, Pontificia Universidad Católica de Chile

To increase β -carotene (i.e., a vitamin A precursor) of durum wheat grains is important to potentially improve pasta quality. *LYCE* (Lycopene Epsilon cyclase) is a key enzyme that diverts the carotenoid biosynthetic pathway into two metabolic branches. The main objective of this work was to amplify the *LYCE* gene for genetic improvement by using the TILLING method (Target Induced Lesions In Genomes). Durum wheat is a tetraploid species with high similarity between the genomes A and B (98%), large genome size (16,000 Mb) and genes (the *LYCE* gene is about 4,000 bp) This made necessary to design specific primers for the A and the B genomes for a specific region of 1500-1700 bp within the *LYCE* gene. Polymorphic indels and SNPs (single nucleotide polymorphisms) between the homeologous genes were used to design specific primers. Conserved regions, truncation mutation percentage, GC percentage and exonic regions were studied for the region selected. We amplified the *LYCE* 3A and 3B genes and we are currently validating them with nullisomic lines. We expect to find 5,1% and 5,5% truncation mutations in the A and the B genomes, respectively, and 40,8% missence mutations on both genomes in those regions selected. The identification of *LYCE* mutants has great value because they will allow the creation of new alleles, which enriches the breeding programs by generating genotypes with higher grain quality (i.e., genotypes with increased levels of grain β -carotene). In addition, the TILLING technique also creates genetic variability that can be used in functional genomics research, and it may elucidate how specifically the carotenoid biosynthetic pathway works in cereals.

Acknowledgements: Fondecyt N°11110066.

PS99

VALIDATION OF MOLECULAR MARKERS FOR USE IN SWEET CHERRY BREEDING PROGRAMS IN CHILE

Roberto Bascuñán de la Fuente^{1,2}, Gerardo Nuñez¹, Hugo Portillo^{1,2}, Sebastián Tapia³,
Claudio Meneses¹, Andrea Miyasaka Almeida^{1,2}

ro.bascunan@uandresbello.edu

¹ Centro de Biotecnología Vegetal, Facultad de Ciencias Biológicas, Universidad Andrés Bello, Santiago, Chile. ² FONDAP Center for Genome Regulation, Santiago, Chile.

³ Instituto de Investigaciones Agropecuarias, INIA La Platina, Santiago, Chile

Molecular markers have been introduced over last decades, which have revolutionized the entire scenario of biological sciences. Traditionally, breeders have relied on visible traits to select improved individuals. Molecular markers rely on identifying DNA sequence markers that are inherited alongside with a desired trait. Over the last years, there has been a significant increase in the application of molecular markers in breeding programs of various crop plants. However, in Chile the use of molecular markers have been implemented only in some cereal species. Cultivation and export of fresh sweet cherry (*Prunus avium*) has generated huge profits in recent years in the country. However the varieties currently used are from other countries which has the disadvantage that are not fully adapted to our conditions. Breeding programs that aim to obtain new varieties through the traditional method requires a large time investment, physical, human and economic. The use of molecular markers for selection of genetic advanced lines could minimize the cost of breeding programs. The aim of this work was to validate molecular markers available in the literature on thirty-one sweet cherry varieties present in Chile. Eight microsatellite (SSR) and one inter single sequence repeats (ISSR) markers were selected. Among the markers selected there was one for the first intron of the S-RNase, which is related with self-incompatibility, markers to assess time of harvest and skin color. Promising results were observed for time of harvest markers which showed strong association with this trait in the varieties analyzed. Molecular marker for self-incompatibility showed to be an important reference in determining the compatibility groups for *Prunus avium*. However, data skin color data were inconclusive. The results show that these molecular markers can be used in *Prunus avium* breeding programs in Chile.

Acknowledgements: FONDEF G09I1008; FONDAP CRG 15070009 and Basal PFB-16.

PS100

**PROPAGATION OF *RHODOPHIALA PRATENSIS* (AMARYLLIDACEAE)
AND CHARACTERIZATION OF ALKALOIDS. OPTIMIZATION TECHNIQUES
THROUGH BIOTECHNOLOGY *IN VITRO***

Evelyn Bustos¹, **Camilo Gatica**², Matilde Uribe², Claudia Pérez¹, José Becerra¹

camilogatica@udec.cl

¹Laboratorio de Química de Productos Naturales, Universidad de Concepción. ²Laboratorio de Cultivo Tejidos Vegetales, Centro de Biotecnología, Universidad de Concepción, Chile

Approximately 1,600 new chemical structures obtained from higher plants are described each year, where a large number of them show biological activity. Therefore, the need arises to use more and better technologies for their production, characterization and optimization. Plant biotechnology is considered one of the cutting edge technologies since it is an alternative way to obtain molecules with diverse biological properties, complex chemical synthesis or from plants growing in limited geographic areas. The Amaryllidaceae alkaloids represents a large (over 300 alkaloids have been isolated) and still expanding group of biogenetically related isoquinoline alkaloids that are found exclusively in plants belonging to this family. Excelling among the compounds are galanthamine, which is being used in the treatment of Alzheimer's disease. The goal of this work was to propagation of the endemic specie *Rhodophiala pratensis*; *in vitro* seed were incubated on solid and supplemented medium MS, determining the best treatment for hormonal induction and subsequent multiplication of bulbs. Also, we used increasing combinations of benzylaminopurine (BAP) and naphthaleneacetic acid (NAA). The treatments were incubated in a growth chamber with controlled conditions of temperature, humidity and a photoperiod of 16 hours light. The best treatment for multiplying bulbs was obtained from bulbs sections and yielded greater shoot production in a shorter time. According to these results, subsequent subcultures were done to increase the biomass concentration varying bulbs addition of sucrose to get the optimal concentration. Our research has allowed us to establish the best protocol for multiplication of bulbs using *in vitro* micropropagation. Actually we are establishing the variation in alkaloid chemical structures collected at time of flowering with those obtained by *in vitro* cultures, characterizing by gas chromatography coupled with mass spectrometry (GC-MS) and spectroscopic methods the chemical structures for derived Galanthamine and Lycorine derived alkaloids.

Acknowledgments: INNOVA BIOBIO 12.462 and University of Concepción

PS101

**STANDARDIZATION OF IN VITRO CULTURE OF *ACTINIDIA DELICIOSA* (KIWI)
AND ITS TRANSFORMATION OVEREXPRESSING A CAROTENOGENIC GENE
OF *DAUCUS CAROTA* (CARROT)**

Luis Felipe Quiroz, Carolina Rosas, Claudia Stange.

Luisfe.quiroz.bt@gmail.com

Plant Molecular Biology Laboratory, Facultad de Ciencias, Universidad de Chile.

Carotenoids are isoprenoid molecules of industrial and nutritional relevance. These molecules are synthesized by all photosynthetic organisms and some non-photosynthetic microorganisms. In plants, carotenoids are synthesized in plastid and they are involved in photosynthesis, photo-protection and synthesis of abscisic acid (ABA). The β -carotene, the main carotenoid in carrots, is a precursor of vitamin A and has antioxidant properties when is ingested by mammals. The lycopene β -cyclase enzyme (LCYB), which catalyzes the conversion of lycopene into β -carotene, is a key enzyme in the biosynthesis of carotenoids. In *Daucus carota* two LCYB have been identified (DcLCYB1 and DcLCYB2). Previously in our laboratory we showed that *DcLcyb2* gene is preferentially expressed in modified carrot root and its over-expression in *N.tabacum* allows to increase β -carotene levels. On the other hand, *Actinidia deliciosa* (kiwi), the third main exported fruit in Chile, has high content of vitamin C but not carotenoids. Among the different varieties of kiwi, golden kiwi (*Actinidia chinensis*) has an orange colored fruit; this phenotype is due to lower levels of chlorophyll, in comparison to the green variety, but not because of carotenoid content. Therefore, in order to increase the carotenoid content in *Actinidia deliciosa* (Kiwi), we used metabolic engineering to express *DcLcyb2* gene in this plant. For this, we standardized successfully a transformation system for *Actinidia deliciosa*, which include *in vitro* culture from seeds, somatic organogenesis, *Agrobacterium tumefaciens* transformation and transformed plant selection. Thus we propose organogenesis and transgenesis as efficient methods for the acquisition and improvement of commercial interest plants such as kiwi, one of the most important fruit plants for the economy of our country.

Acknowledgments: FONDEF-VIU [110046].

PS102

**POLYSACCHARIDES ISOLATED FROM *ALOE BARBADENSIS* MILLER (*ALOE VERA*)
ARE EXCELLENT PREBIOTIC MOLECULES.**

María Paz Quezada¹, Carlos Salinas¹, Martin Gotteland², Liliana Cardemil¹

lcardemi@gmail.com

¹Laboratorio de Biología Molecular Vegetal, Departamento de Biología, Facultad de Ciencias, Universidad de Chile.²Departamento de nutrición, Facultad de Medicina, Universidad de Chile.

Aloe vera plants have medicinal properties. We isolated two polysaccharides (PS): PS1 from green leaf tissue and PS2 from leaf gel. Both molecules have glycosidic linkages resistant to enzymatic hydrolysis by the intestinal tract and by enterobacteria present in the intestine. By in vitro bacterial growth analysis, the prebiotic potential of these isolated and purified polysaccharides was tested. For this, the growth of 8 *Lactobacillus* and *Bifidobacterium* strains was evaluated in a culture medium containing these polysaccharides as the sole carbon source. Bacterial growth was compared with that induced by glucose (Glc) or a commercial prebiotic polysaccharide, Nutra Food. The results show that PS1 induces growth similar to that induced by Glc and higher than that induced by Nutra Food. PS2 increases growth to levels similar to Nutra Food while a mix of PS1 and PS2 induces growth lower than that induced by Glc but higher than that of Nutra Food. By quantitative RT-PCR, the benefit of each polysaccharide in the specific intestinal *Lactobacillus* and *Bifidobacterium* populations present in human stools was evaluated. Stools were cultured in a bioreactor and population growth was compared to that induced by Glc. After 48 h in the bioreactor, PS2 was a better carbon source than PS1 for *Lactobacillus* and comparable to Glc for *Bifidobacterium* populations. These results indicate that both polysaccharides have prebiotic properties.

Acknowledgments: Project VIU 110015, FONDECYT 1130025.

PS103

**CHARACTERIZATION OF A NEW CARLAVIRUS INFECTING NATIVE POTATOES
IN THE SOUTH OF CHILE**

Elizabeth Peña¹, Thierry Candresse², Armelle Marais², Marlene Rosales¹

erpena@uc.cl

¹Facultad de Agronomía e Ingeniería Forestal, Pontificia Universidad Católica de Chile.

²Equipe de Virologie, INRA Centre Bordeaux-Aquitaine, Bordeaux, France.

Potato is one of the most important foods crops in the world. In Chile, the potato crop is present in several regions along the country, including native potato varieties originating in the Chiloé archipelago, which are very diverse in terms of color, shape, texture, size and flavor. However, viral diseases are an important factor limiting potato production. Many viruses, such as Potato virus Y (PVY), Potato virus X (PVX), Potato leafroll virus (PLRV), Potato virus S (PVS), Potato virus M (PVM) cause severe symptoms, and yield losses. Preliminary studies of potato viruses led to the discovery of a new virus. The determination and analysis of its partial nucleotide sequence provided evidence that this agent may represent a new species of the genus *Carlavirus*. Complete RNA genomic sequences of one isolate of new virus was determined using reverse transcription-long distance PCR and the 5' rapid amplification of cDNA ends (5' RACE) method. Sequence analysis revealed that the new virus had the typical genomic organization of members of the genus *Carlavirus*, with a positive-sense single-stranded genome of 8422 nt. The amino acid sequences of the coat protein and RNA dependent RNA polymerase showed a sequence identity between 28.7 to 71.8% and 39.1 to 56.7%, respectively, compared with those reported *Carlavirus*. Although the discovery of this new virus is important, it remains to elucidate their origin, their distribution in natives and traditional potato crops, its host range, as well as its vector transmission, and the effects of co-infection with other important viruses of the potato as PVY or PVX for complete characterization.

Acknowledgments: ECOS-CONICYT Project 2010-016.

PS104

**RHIZOBACTERIA STRAIN ISOLATED FROM COQUIMBO FOR BIOCONTROL
OF SCLEROTINIA AND BOTRYTIS SP.**

Williams Arancibia^{1,2}, Stefanie Maldonado², Andrés Rodríguez-Aguilera², Luis Castillo³,
Alexandra Stoll², Jaime Bravo^{4,2}

williams.arancibia@gmail.com

¹Facultad de Ciencias, Escuela de Agronomía Universidad de La Serena. ²Laboratorio de Microbiología, Centro de Estudios Avanzados en Zonas Áridas. ³Biología, Universidad de La Serena. ⁴Microbiología, Universidad de Pénjamo, México.

In Chile, horticultural sector provides a wide range of products highly ranked by their organoleptic characteristics. However, the international phytosanitary standards require better products from nutritional and ecological point. In this context, we need reduce agrochemical use as an important theme to generate higher quality products. For all that, this work aims to characterize native bacterial strains antagonistic over fungi *Sclerotinia* and *Botrytis* sp. Its cause problems in lettuce crop (*Lactuca sativa*), in arid zones of our country. We obtain approximately 1200 bacteria, from agricultural soil lettuce crop of Coquimbo region. Then, we were isolated *Bacillus* sp. Then, we had selected 110 strains with some control degree over fungus, using and adversarial trial. Five strains were selected to quantify its antagonistic power over *Sclerotinia* and *Botrytis* sp. We showed a reduction of fungal area between 25-50% and decreased the sclerotia formation in 80%. Also, we examined the bacterial strains for its ability to produce siderophores, indol acetic acid, phosphate solubilization and nitrogen fixation, like other promoting growth plant rhizobacteria characteristics. Finally we tested bacterial strains antagonist ability over lettuce plants previously infected. The results suggest a partial biocontrol with the five bacterial strains for *Sclerotinia* sp and *Botrytis* sp pathogens. The perspective work is establishment an assay on commercial crop conditions.

Acknowledgements: project GORE Coquimbo FIC 2012 BIP 30127532-0.

PS105

**BIOTECHNOLOGICAL APPROACHES TO IDENTIFY VARIETAL CONTAMINATION
AND VIRAL DISEASE IN RED RASPBERRY CV HERITAGE**

Pamela Rojas¹, Marina Gambardella², José San Martín¹, Boris Sagredo¹

projas@inia.cl

¹Instituto de Investigaciones Agropecuarias (INIA) CRI Rayentue-Raihuen, Chile.

²Departamento de Fruticultura y Enología, Pontificia Universidad Católica de Chile.

Red Raspberry (*Rubus idaeus*) is one of the main berry crops in central regions of Chile, and 80% of its cultivated area is occupied by cv Heritage. Chile is the principal producer of red raspberry of the south hemisphere but problems affecting fruit quality have reduced its market price. Most raspberry orchards are established with plants propagated by growers and the use of certificated plant material is infrequent. Therefore, virus accumulation in orchards could be at the basis of problems affecting fruit quality and yield. Additionally, volunteer plants germinated from open pollinated raspberry seeds can contribute to the fruit quality loss. Nowadays, molecular techniques such as SSR, ELISA and RT-PCR are valuable tools for cultivars and viruses identification, respectively. Aiming to establish if the varietal contamination and/or viral diseases are related with loss of quality in raspberry orchards, we collected and analyzed samples of eight orchards from Romeral (Maule region), using 8 SSR and ELISA test for detection of *Tomato ringspot virus* (ToRSV), *Raspberry bushy dwarf virus* (RBDV), *Arabidopsis mosaic virus* (ArMV) and *Strawberry latent ringspot virus* (SLRSV). ELISA positive results were confirmed by RT-PCR. Neither cultivar contamination nor detection of RBDV, ArMV and SLRSV were found in the analyzed samples. Three of the eight orchards evaluated were positive to ToRSV but in a low infection level. However, it is strongly necessary assess a wider panel of viruses and study novel possible viruses affecting raspberry orchards in Chile to develop control strategies for viral diseases in the field.

Acknowledgements: Mejoramiento Genético de Frambuesas en Chile (08CT11PUD-14) funded by INNOVA-CORFO, FDF, Consorcio Tecnológico de la Industria Hortofrutícola S.A, PUC and INIA.

PS106

**IDENTIFICATION AND MYCORRHIZA ABUNDANCE IN EUCALYPTUS NITENS
(DEANE & MAIDEN) NURSERY AND PLANTATIONS OF THE VII AND VIII REGION OF CHILE.**

Daniela Torres¹, Angélica Casanova-Katny², Götz Palfner¹

danielatorres@udec.cl

¹Laboratorio de Micología y Micorrizas, Departamento de Botánica; Universidad de Concepción, Concepción, Chile. ²Departamento de Microbiología, Facultad de Ciencias Biológicas, Universidad de Concepción, Concepción, Chile

Many studies have reported the occurrence of two major mycorrhizal associations: ectomycorrhiza (EM) and arbuscular mycorrhiza (AM) in the genus *Eucalyptus*. There are different factors that affect the establishment of mycorrhiza, such as plant age and fertilization. Based on these factors, a study of the mycorrhiza community in *Eucalyptus nitens* (Deane & Maiden) in nurseries and plantations of south central Chile was performed. To this, we analyzed seven *Eucalyptus nitens* plants of seven different ages classes. Three of them were obtained from nursery and 4, from plantations. In this analysis we observed five morphotypes of ectomycorrhiza (*Telephora terrestris*, *Descomyces sp*, *Scleroderma sp*, *Hydnangium sp* and *Hysterangium sp*). There was no significant difference in the abundance of the morphotypes with respect to age classes. According to the source of the specimens, the colonization was lower in nursery compared to plantation. Regarding arbuscular mycorrhiza, there was no significant presence in the different age classes. These results suggest that in the studied area *E. nitens* is largely colonized by ectomycorrhiza, this in nursery as well as in plantation. However there are several factors that affect the establishment of one or another type of mycorrhizal symbiosis or dual. *T. terrestris* as root morphotype was dominant in all age classes, independent of the crop factors, suggesting that this species appropriated as inoculums for seedlings.

Acknowledgements: Innova Bío-Bío

PS107

**FOLIAR DAMAGE IN *DESCHAMPSIA ANTARCTICA* AT CONTRASTING NATURAL
AND EXPERIMENTAL SITES.**

Sebastián Morales-Munita¹, Angélica Casanova-Katny^{1,2}, Eugenio Sanfuentes¹, Götz Palfner³

¹Laboratorio de Patología Forestal, Centro de Biotecnología, Universidad de Concepción, Chile. ²Departamento de Microbiología, Facultad de Ciencias Biológicas, Universidad de Concepción, Chile. ³Laboratorio de Micología y Micorrizas, Departamento de Botánica; Universidad de Concepción, Chile.

sebasmorales@udec.cl

Populations of *Deschampsia antarctica* in the maritime Antarctic commonly show foliar damage with typical symptoms being decoloration, blackish spots or bands and partial necrosis. At least partially, this damage can be associated with fungi: Light microscopy and in vitro culture of affected leaf segments yielded evidence for presence of important phytopathogenic genera of Ascomycetes and Oomycetes like *Leptosphaeria*, *Mycosphaerella*, *Pleospora* and *Pythium*, among others. Interestingly, frequency of damaged/infected leaves shows differences between population sites and *in situ* experimental treatment, probably depending on microclimatic factors like temperature and moisture. We compared frequency of leaves with the mentioned symptoms in adult plants of *Deschampsia antarctica* between populations on King George Island, Fildes Peninsula, on one hand between two sites with contrasting exposition, viz.: Fildes Bay (protected) vs. open site to Drake Sea (exposed) and on the other hand between colonies having grown during five years in open top chambers (OTC) and the respective control (without OTC). Frequency of foliar damage was lower in plants growing at the exposed sites (Drake Sea 54%) than in protected populations (Fildes Bay 62%). Similarly, lower values were found in plants growing outside than inside OTC (46% vs. 73%, respectively). One possible reason for the higher frequency of damage in the protected habitats could be increased virulence of phytopathogenic fungi, stimulated by increased temperature which is about 3°C higher inside OTCs. Further experiments are necessary to confirm the effect of these fungi on Antarctic vascular plants under a climate change scenario.

Acknowledgements: FONDECYT 1120895, INACH PR_05-12.

PS108

**ASSESSMENT OF THE *EUCALYPTUS NITENS* CUTICLE BY FOURIER TRANSFORM
INFRARED SPECTROSCOPY (FTIR) IN RESPONSE TO COLD ACCLIMATION**

María Navarrete¹, Daniela Alvarado¹, Catalina Lagos¹, Rosario Castillo², Verónica Emhart³,
Sofía Valenzuela^{1,4}, Marta Fernández^{1,4}

marianavarrete@udec.cl

¹Facultad de Ciencias Forestales, Universidad de Concepción, ²Facultad de Farmacia,
Universidad de Concepción, ³Forestal Mininco S.A., ⁴Centro de Biotecnología,
Universidad de Concepción.

The primary environmental factor responsible for triggering the increase in freezing tolerance in some plants is low nonfreezing temperatures, a phenomenon known as cold acclimation. *Eucalyptus nitens* shows tolerance to cold temperatures, allowing its establishment in areas where other species have growth limitations, as is the case of *Eucalyptus globulus*. However, the nature of the mechanisms involved in freezing tolerance that activate the cold-acclimation response are not well known. *Eucalyptus* species have a hydrophobic layer, which gives a glaucous appearance to leaves and is known to play an important role in protecting aerial organs from damage caused by environmental stress. In this work, the main functional groups deposited on the leaf cuticle of *E. nitens* in response to acclimation were identified by FTIR spectroscopy with Attenuated Total Reflectance (ATR). FTIR analysis was applied in different areas of the leaf (lamina and vein), without sample preparation. A cold chamber assay was established by assessing young plants of four open-pollinated families of *E. nitens*, under the following conditions: non-acclimated (NA, 12/20 °C day/night), cold acclimated to non-freezing low temperature (CAC, 4/8 °C day/night), acclimated to freezing temperature (CAF, 6/12 °C day/night), and de-acclimated (DA, 6/12 °C day/night). Leaves from the third verticil were collected from three plants per family at NA and CAF. A night frost of -6 °C during DA was applied to determine survival and the percentage of leaf damage for each family to assess their tolerance to a simulated late-spring frost. The results showed variability in freezing tolerance among the four families. FTIR spectra were analyzed by vibration frequencies of macromolecules. The principal component analysis (PCA), showed the presence of amino, hydroxyl groups, fatty acids, amide I, amide II compounds which showed the highest peaks at 3728, 3620, 2916, 1500, 1644, respectively in the lamina of leaves from CAF plants.

Acknowledgments: FONDECYT 11121559.

AUTHOR INDEX

A

Abarca, B.	122
Aceituno, U.	86
Acevedo, F.	82
Acosta, M.	147
Acuña, M	59, 97, 99
Aguirre, C.	111
Agurto, M.	141
Alberdi, M.	29
Alcorta, M.	136
Alister, S.	121
Almada, R.	40, 59, 87, 89, 97, 99, 101, 137, 139
Almonacid, D.	107
Alvarado, D.	160
Álvarez, C.	28, 33, 142
Álvarez-Gerding, X.	110
Alvear, C.	29, 120
Alzate-Morales, J.	53
Andrade, D.	132
Antúñez, A.	134
Arancibia, W.	156
Araya, A.	145
Araya, C.	43
Araya, M.	43
Arbona, V.	18
Arce-Johnson, P.	110, 141
Arellano, M.	78
Arismendi, M.	137
Armijo, G.	141
Aros, D.	63
Arraño, P.	73
Arrey, O.	30, 124
Avilés, F.	146
Avilés, N.	146
Ayala, A.	61, 134
Ayala, H.	105, 106
Azócar, C.	74

B

Baginsky, C.	116, 121
Balbontín, C.	92, 105, 106
Balic, I.	27, 36, 135
Barba, P.	111
Bascuñán, R.	151
Bastías, A.	119
Beaudry, R.	44
Becerra, J.	152
Becerra, V.	81, 85, 86, 125
Benedetto, G.	54, 132, 133
Berrios, G.	60
Blanco, F.	64, 65, 73 94
Bravo, J.	46, 156
Bravo, L.	29, 60, 90, 120, 126, 128
Bravo, M.	117
Bravo, S.	83, 143
Bremer, A.	53
Brown, M.	21
Bucki, L.	53
Bustos, E.	66, 152

C

Caballero, J.	53
Cabrera, S.	77
Campos, K.	42, 71
Campos-Vargas, R.	25, 27, 36, 135
Candresse, T.	155
Cardemil, L.	154
Carrasco, B.	43, 62, 77, 144, 148
Carrasco, C.	55
Carvajal, D.	111

VIII Reunión de Biología Vegetal

Casanova-Katny, A.	158, 159
Castillo, L.	156
Castillo, R.	143, 160
Castro, A.	28, 111, 112
Cavieres, L.	120
Chávez, S.	65
Chilian, J.	81
Cifuentes, A.	25
Cifuentes-Esquivel, A.	130
Claverol, S.	128
Concha, C.	108
Contreras, C.	44
Contreras, R.	101, 137, 139
Coopman, R.	127
Corcuera, L.	126, 127
Correa, F.	87
Correa, J.	43, 72
Couture, C.	112
Covarrubias, M.	132, 133
Cruz, N.	104
Cuba-Díaz, M.	31

D

Davis, T.	17
de Ollas, C.	18
Decroocq, V.	111, 112
Defilippi, B.	36, 61, 75
del Pozo, A.	118
Delgado, J.	135
Dell'Orto, P.	111
Di Génova, A.	26, 61
Díaz, A.	20
Díaz, M.	29, 131
Dimeglio, L.	17
Dittrich, J.	20
Donoso, G.	85, 86, 125
Doñas, D.	98

E

Elizondo, D.	63, 94
Emhart, V.	160
Escandón, A.	127
Escobar, H.	116

Espejo, R.	19
Espinoza, C.	110

F

Federico, M.	93, 149
Fernández, M.	160
Figueroa, A.	104
Figueroa, C.	34, 108, 123
Figueroa, L.	79
Figueroa, N.	108, 123
Figueroa, P.	68
Figueroa, R.	88, 144
Flores-Bavestrello, A.	126
Franck, N.	80, 129
Friedt, W.	96
Fuentes, L.	61, 134

G

Gaete-Eastman, C.	58
Gainza, F.	40, 105, 106, 139
Gajardo, H.	149
Galilea, B.	64, 73
Gambardella, M.	157
Garcés, M.	128
Gatica, C.	152
Gatica, M.	107, 115
Gebauer, M.	88, 144, 148
Godoy, S.	142
Gómez, C.	137
Gómez-Cadenas, A.	18
Gonzalez, A.	32
González, C.	100
González, E.	69, 70
González, J.	86, 115
González, M.	62, 77, 148
González, S.	69
González, W.	53
González-Agüero, M.	26, 75, 103
González-Ramírez, C.	49
Gotteland, M.	154
Gratacós, E.	41

Greze, I.	122	Klagges, C.	41
Guajardo, V.	40	Krause, P.	96
Gusmão, E.	132	Król, M.	126
Gutiérrez, A.	60, 90, 131		
Gutiérrez, R.	48	L	
Guzmán, A.	41	Laborie, D.	72
		Lagos, C.	160
H		Laiz, P.	72
Handford, M.	39, 47	Larama, G.	60
Hasbún, R.	78, 143	Latapiat, V.	35
Henríquez, C.	114, 117	Leal, V.	122
Hernández, M.	88	Lemus, G.	25, 40
Hernández, O.	37	León, R.	88
Herrera, R.	34, 35, 38, 55, 56, 57, 58, 59, 76, 104, 109	Leyton, M.	85
Hicks, G.	21, 48	Lisboa, K.	81
Hincha, D.	53	Liu, B.	17
Hinrichsen, P.	26, 45, 72, 91, 97, 99, 103, 119, 137	Lizana, R.	56, 109
Hooley, R.	21	López-Emparán, A.	93
Hummer, K.	17	Lundberg, M.	17
Hüner, N.	126	Lutts, S.	134
		M	
I		Maass, A.	26, 61
Ibacache, A.	100	Magni, C.	122
Ibáñez, C.	32, 145	Mahoney, L.	17
Infante, R.	25, 102, 130	Maldonado, J.	62, 105
Iniguez-Luy	149	Maldonado, S.	46, 156
Inostroza-Blancheteau, C.	110	Mamani, M.	45, 72
Ivanov, A.	126	Manque, P.	60
		Mansur, L.	41
J		Marai, A.	155
Jiménez, N.	42, 71	Martin, R.	20
Jofré, E.	145	Martínez, J.	61, 134
Jorrín, J.	86	Mathias, M.	82
		Matus, I.	42, 71
K		Meisel, L.	41, 102
Keller, K.	20	Mejia, N.	75
		Mella, C.	29
		Méndez, M.	43
		Meneses, C.	25, 27, 63, 130, 151
		Meneses, F.	94
		Meneses, M.	148
		Miccono, M.	107
		Miranda, G.	68

VIII Reunión de Biología Vegetal

Mitina, I.	114	Olivos, A.	109
Miyasaka-Almeida, A.	54, 79, 83, 84, 107, 115, 132, 133, 151	Olmedo, P.	36
Molina, R.	75	Oñate, F.	108
Molinett, S.	76	Opazo, I.	40
Monsalve, L.	61	Orellana, A.	25, 26, 27, 54, 64, 65, 94, 98, 103, 113, 114, 135,
Montes, C.	111, 112		
Montoya, M.	100	Ortega, L.	142
Moraga, C.	54	Ortiz, M.	40
Morales, C.	92	Osorio, C.	48, 67
Morales, E.	82		
Morales, L.	116, 121	P	
Morales, P.	62, 144, 148	Palfner, G.	158, 159
Morales-Munita, S.	159	Paredes, M.	81, 85, 86, 125
Morales-Quintana, L.	34, 35, 57, 58, 59	Parra, J.	113
Moreno, A.	36, 65, 73, 113, 114	Pastenes, C.	80, 129
Mosier, N.	20	Paula, S.	127
Moya-León, M.	34, 35, 37, 38, 55, 56, 67, 58, 74, 76, 109	Pavicic, M.	68
Mujica, K.	102	Peña, E.	155
Muñoz, C.	26, 40, 103	Peredo, T.	135
Mura, I.	62	Pérez, C.	66, 152
N		Pérez, F.	95
Navarrete, C.	145	Pérez-Alfocea, F.	134
Navarrete, M.	160	Perez-Donoso, A.	39
Navarro-Retamal, C.	53	Pimentel, P.	40, 59, 87, 89, 91, 97, 99, 101, 117, 119, 137, 139
Nilo, R.	54		
Norambuena, L.	48, 49, 67, 104	Pino, M.	117
Núñez, G.	25, 63, 151	Pinochet, J.	40
O		Pinolef, A.	122
Olea, P.	77	Pinto, M.	40, 72, 87, 89, 91, 97, 99, 101, 117, 137
		Pizarro, L.	48, 49, 67, 116
		Poblete, C.	99
		Poblete, L.	108, 123
		Portillo, H.	115, 151

Prieto, H.	28, 83, 84, 107, 111, 112, 115, 135, 142	Ruiz-Lara, S.	69, 70
Q		S	
Quezada, M.	121, 154	Saavedra, G.	70, 123
Quiroz, D.	28	Saens, M.	57
Quiroz, L.	153	Sáez, P.	33, 120
Quito-Avila, D.	20	Saez-Aguayo, S.	98
		Sagredo, B.	40, 43, 59, 87, 89, 91, 97, 99, 101, 119, 137, 139, 157
R		Salazar, C.	66
Rabert, C.	29, 131	Salazar, E.	43, 62, 77
Raikhel, N.	21	Salinas, C.	154
Ramos, P.	38, 55, 74, 104	Salinas, L.	134
Ravest, G.	45	Salvatierra, A.	87, 89, 91, 101
Razmilic, I.	37	Salvo-Garrido, H.	82
Recabarren, C.	64	San Martin, J.	40, 157
Reyes, C.	120	Sánchez, A.	26, 103
Reyes, F.	98	Sánchez, E.	111, 115
Reyes, M.	29	Sánchez, M.	146
Richaud, D.	150	Sánchez-Olate, M.	33
Rios, D.	33, 140, 146	Sandoval, A.	131
Ríos, N.	80, 129	Sandoval, C.	79
Rivas, P.	78	Sandoval, O.	114
Riveros, G.	136, 140	Sanfuentes, E.	159
Roa, R.	70	Sanhueza, C.	120
Rodríguez, F.	102	Sanhueza, D.	27, 36
Rodríguez-Aguilera, A.	46, 156	Sarver, K.	20
Rodríguez-Furlán, C.	64	Schlechter, R.	141
Rojas, M.	145	Schulthess, A.	42, 71
Rojas, P.	97, 99, 157	Schwember, A.	42, 71, 150
Rojas, R.	127	Sepúlveda, F.	143
Rojas-Pierce, M.	48	Shields, M.	17
Rosales, M.	155	Sierra-Almeida, A.	120
Rosas, C.	153	Silva, C.	27
Rothkegel, K.	83	Silva, H.	41, 62, 105, 116, 121, 148
Rubilar, J.	105, 106		
Rubilar, M.	82, 130		
Rubio, J.	111	Silva, R.	72
Rubio, S.	95		
Rubio-Astudillo, J.	112		

VIII Reunión de Biología Vegetal

Silva, S.	45
Silva-Sanzana, C.	36
Snowdon, R.	96, 149
Solís, S.	101, 139
Spielman, H.	138
Stange, C.	153
Stappung, Y.	30, 56, 76, 105, 109
Stoll, A.	46, 156

T

Tapia, D.	90
Tapia, E.	111, 115, 142
Tapia, G.	30, 92, 118, 124, 136, 140
Tapia, S.	84, 151
Tejerina, L.	75
Thalhammer, A.	53
Torres, C.	37, 74
Torres, D.	158
Torres, P.	82
Travisany, D.	61
Troncoso, M	59
Turner, A.	143
Tzanetakis, I.	20

U

Undurraga, P.	75
Uquillas, C.	122
Urbina, D.	34
Uribe, M.	66, 152
Utz, D.	47

V

Valenzuela, C.	55, 104
Valenzuela, F.	58
Valenzuela, M.	133
Valenzuela, S.	160
Valledor, L.	33
Van de Ven, W.	21
Vargas, M.	145
Vásquez, A.	145
Vásquez, V.	122
Vega, C.	147

Vega, J.	107
Velásquez, R.	131
Venegas, C.	138
Verdonk, J.	79
Vergara, A.	48
Viera, E.	109
Villalobos, L.	80, 129
Villegas, D.	39
Vizoso, P.	27

W

Westphal, C.	32
Wittkop, B.	149
Wood, D.	17
Y	
Yang Y.	17
Yáñez, M.	70, 118
Young, M.	21

Z

Zamudio, M.	75
Zhang, C.	21
Zhang, Q.	17
Zúñiga, C.	122
Zúñiga, F.	19, 107
Zúñiga, M.	93
Zurita-Silva, A.	100

