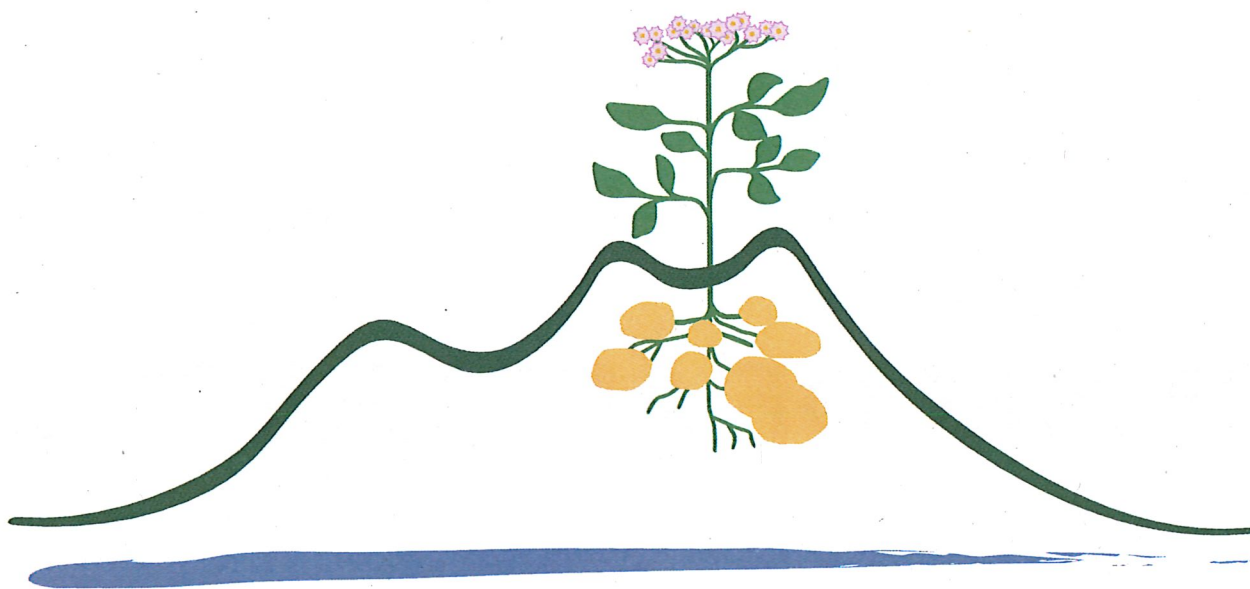




EUCARPIA 



BOOK OF ABSTRACTS

2015

EAPR Breeding and Varietal Assessment
Section and EUCARPIA Section Potatoes

18TH JOINT MEETING

Vico Equense, Italy - November 15th – 18th, 2015



BOOK OF ABSTRACTS

2015 EAPR Breeding and Varietal Assessment
Section and EUCARPIA Section Potatoes

18TH JOINT MEETING

Vico Equense, Italy - November 15th – 18th, 2015

Local organising committee: Riccardo Aversano, Amalia Barone, Maria Raffaella Ercolano, Edgardo Filippone, Luigi Frusciante (University of Naples Federico II, Dept. Agricultural Sciences)

Scientific committee: Glenn Bryan, Domenico Carputo, Finlay Dale, Luigi Frusciante, Dan Milbourne, Vanessa Prigge

Dear Participants,

It is a great pleasure to welcome you to the 18th Triennial Meeting of the EAPR Section 'Breeding and Varietal Assessment' and the EUCARPIA Section 'Potatoes' in Vico Equense, Italy. This meeting has been designed to bring experts and students together from disciplines covering all relevant aspects of modern potato genetics and breeding. We hope the relaxing environment stimulates new proficuous collaborations.

We are very glad that so many delegates are coming from all over the world to share their knowledge and expertise. There will be 6 keynote lectures, 33 oral presentations and several posters. The meeting will also have a special session on the occasion of Dr. Christiane Gebhardt's retirement, a great opportunity for the entire potato community to celebrate her exemplary career.

We would like to thank EAPR, EUCARPIA, the Department of Agricultural Sciences of the University of Naples Federico II and all the sponsors for their financial contribution, the Scientific Committee for continuous support and useful suggestions, PlantGeM and PQS for assistance in the organization of the meeting. Last but not least, many thanks to all of you for participating.

We hope you enjoy the scientific program as well as the unique environment of the Sorrento Peninsula!

The Local Organizing Committee

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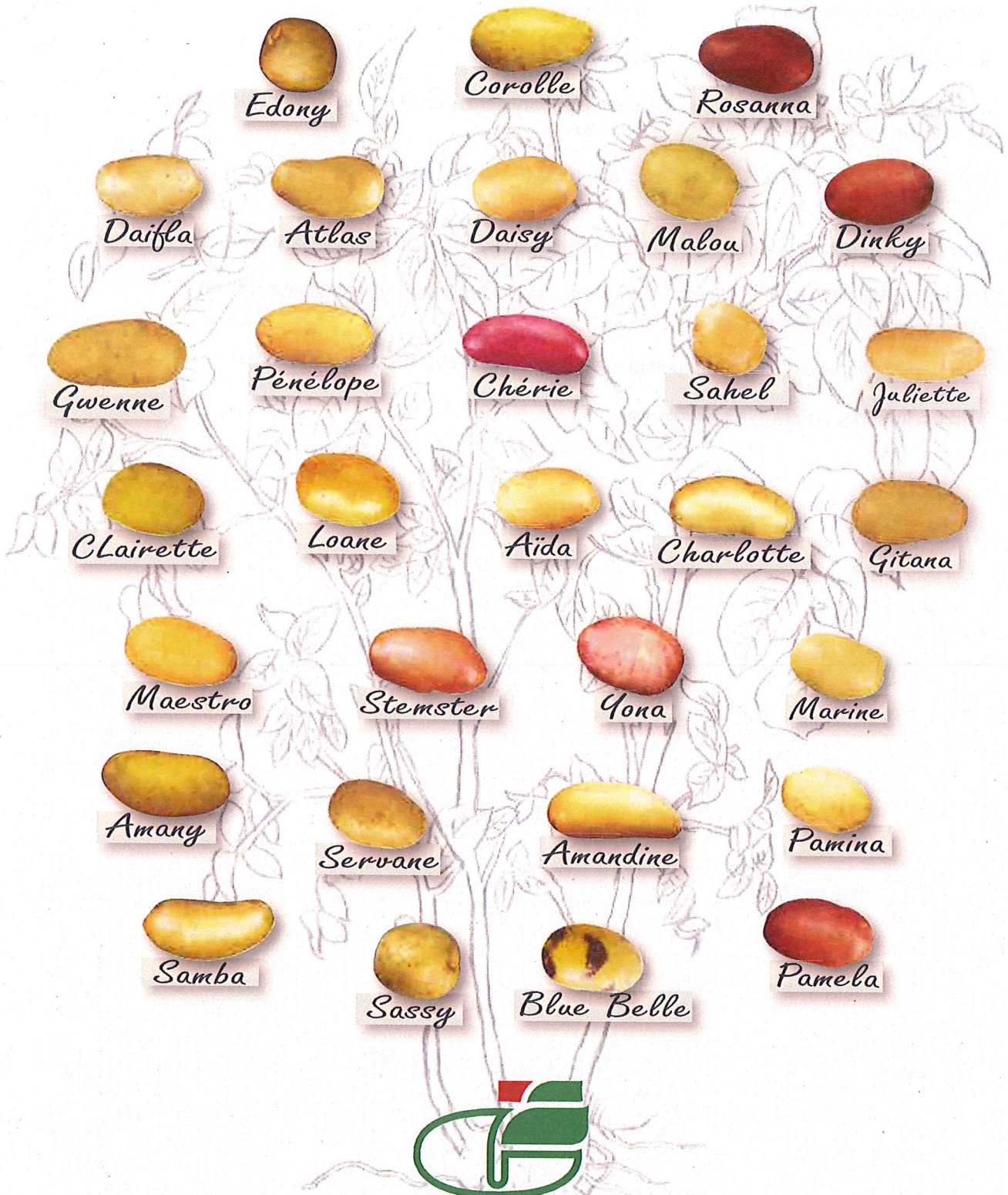
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GERMICOPA

CRÉATEUR VARIÉTAL DE POMME DE TERRE
POTATO VARIETIES CREATOR



SCIENTIFIC PROGRAMME

MONDAY, NOVEMBER 16

08.30 - 09.00

Opening ceremony

09:00 - 12:40 Session 1 – BIOTIC INTERACTIONS

Chairpersons: Prigge Vanessa, Visser Richard

09.00 - 09.30

Keynote Lecture

Bradeen James

Borrowing from neighbors: comparative genomics approaches for resistance gene discovery and deployment

09.30 - 09.50

Chen Xinwei

Application of novel enrichment and sequencing approaches for rapid marker development and fine-mapping of a broad-spectrum blight resistance locus from *S. verrucosum*

09.50 - 10.10

Fadina Oksana

Late blight resistance genes in complex interspecific potato hybrids

10.10 - 10.30

Hutten Ronald

Rpi-gene mediated late blight resistance in potato tubers

10.30 - 10.50

Vossen Jack

Novel R gene differential set plants for monitoring virulence in *P. infestans*

10.50 - 11.20

COFFEE BREAK AND POSTER DISPLAY

Session 1 – BIOTIC INTERACTIONS (cont.)

11.20 - 11.40

Milbourne Dan

Stability of resistance conferred by pyramiding two QRLs for *G. pallida* Pa2/3

11.40 - 12.00

Valls Marc

Molecular and genomic insights on potato resistance to bacterial wilt

12.00 - 12.20

Jacobs Jeanne

Soft rot resistance in potato lines derived from a somatic hybrid with *Solanum palustre*

12.20 - 12.40

Discussion

12.40 - 14.30

LUNCH

14.30 - 16.00 Session 2 - QUALITY TRAITS

Chairpersons: Milbourne Dan, Mosquera Teresa

14.30 - 15.00

Keynote Lecture

Sliwka Jadwiga

Most QTL for leaf sucrose content map to positions similar to positions of QTL for tuber starch content in diploid potato

15.00 - 15.20

Mandolino Giuseppe

Identification of new alleles for carotenoid biosynthesis and degradation in a collection of potato varieties

15.20 - 15.40

Diretto Gianfranco

Metabolic engineering of carotenoids in potato affects ABA metabolism and tuber shelf-life

15.40 - 16.00

Ngobese Nomali

Evaluating the yield and tuber quality performance of eight European cultivars in Pietermaritzburg to improve potato production in South Africa

A selection of our
Promising varieties

PRIMARY USE:



TRADITIONAL



RODEO



FABULA



FARIDA



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SIFRA



MEMPHIS

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SALAD



RETAIL FRESH



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PANTHER



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16.00 - 16.30 **COFFEE BREAK AND POSTER DISPLAY**

16.30 - 18.20 Session 3 - ABIOTIC STRESSES

Chairpersons: Zimnoch-Guzowska Ewa, Jacobs Jeanne

16.30 - 17.00 **Keynote Lecture**
Visser Richard
Unraveling drought stress in potato

17.00 - 17.20 *Aversano Riccardo*
Stress-tolerant *Solanum commersonii*: genome sequence and manipulation

17.20 - 17.40 *Hosang Laura*
Selection method for salt tolerance in potato

17.40 - 18.00 *Prashar Ankush*
Sensing the plants: can we detect stress

18.00 - 18.20 *Discussion*

20:30 **DINNER**

TUESDAY, NOVEMBER 17

08.30 - 12.40 Session 4 - DIVERSITY, BREEDING AND SELECTION - 1

Chairpersons: Douches David, Van Eck Herman

08.30 - 09.00 **Keynote Lecture**
Lindhout Pim
Be aware: diploid hybrid potatoes are coming!

09.00 - 09.30 **Keynote Lecture**
Jansky Shelley
Progress toward the development of recombinant inbred lines

09.30 - 09.50 *Douches David*
Progress in diploid potato breeding with self-compatibility

09.50 - 10.10 *Greplová Marie*
Somatic hybrids in sexual hybridization of potato

10.10 - 10.30 *Smyda-Dajmund Paulina*
Genetic diversity and composition of *S. x michoacanum* (+) *S. tuberosum* somatic hybrids and 4x *S. x michoacanum* evaluated by Diversity Array Technology and PCR markers

10.30 - 10.50 *Iwama Kazuto*
Genotypic differences in solar radiation used efficiency intercepted by plant canopy and its relation to early tuber bulking in a CxE diploid potato population

10.50 - 11.20 **COFFEE BREAK AND POSTER DISPLAY**

Session 4 – DIVERSITY, BREEDING AND SELECTION - 1 (cont.)

11.20 - 11.40 *Visser Richard*
Towards durable resistance against *P. infestans*

11.40 - 12.00 *Rogozina Elena*
New superior parental material developed by pre-breeding from potato collection at the Vavilov Research Institute of Plant Industry (VIR)

12.00 - 12.20 *D'Amelia Vincenzo*
New insights into the role of anthocyanin duplicated genes in response to cold stress in the potato plant

12.20 - 12.40 *Discussion*

12.40 - 14.30 **LUNCH**



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DALLA PREPARAZIONE DEL CAMPIONE

ALL'ANALISI BIOINFORMATICA DEI DATI

1 Sequencing

Sequenziamento Sanger: Sequenziamento di prodotti di PCR - Sequenziamento di vettori plasmidici - Walking primer

Next Generation Sequencing: Sequenziamento de novo di genomi batterici - Caratterizzazione metagenomica di campioni complessi - Rilevazione di patogeni - Identificazione di SNPs - Analisi Epigenetiche e di Metilazione - Sequenziamento Whole Genome - Sequenziamento Trascrittoma

3 Oligo Synthesis

Sintesi di DNA/RNA primers, DNA probes, siRNA

5 Sample Processing

Estrazione di DNA/RNA da campioni biologici, RT-PCR, QPCR

2 Microarray

Profili di espressione genica - profili di espressione di miRNA - Interazione proteina /DNA Chip-on-chip - Metilazione del DNA mediante immunoprecipitazione meDIP-chip - Genomica Comparativa CGH

4 Protein Expression

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**14.30 - 16.45 Session 5 – SPECIAL SESSION ON THE OCCASION OF THE
RETIREMENT OF DR. C. GEBHARDT**

Chairpersons: Bryan Glenn, Carputo Domenico

- 14.30 - 15.00 *Salamini Francesco*
Christiane Gebhardt: what I do remember
- 15.00 - 15.30 *Mosquera Teresa*
Potato genetic studies in *Solanum tuberosum* group Phureja
- 15.30 - 16.00 *Debener Thomas*
Transcriptome sequencing as a tool to study the potato wart interaction
- 16.00 - 16.45 *Gebhardt Christiane*
Molecular diagnostics of complex agronomic traits in tetraploid potato
- 16.45 - 17.15 **COFFEE BREAK AND POSTER DISPLAY**
- 17.15 - 18.00 **Section Meeting**
- 20.30 **Social Dinner**

WEDNESDAY, NOVEMBER 18

09.00 - 13.00 Session 6 – DIVERSITY, BREEDING AND SELECTION - 2

Chairpersons: Bradeen James, Sliwka Jadwiga

- 09.00 - 09.30 **Keynote Lecture**
Bryan Glenn
Analysis of potato traits in crosses and association panels
- 09.30 - 09.50 *Van Eck Herman*
Haplotype reconstruction in tetraploids using sequence read based alignments
- 09.50 - 10.10 *Lemm Jana*
A simple approach to score SNP markers in a dosage-dependent fashion in tetraploid potato
- 10.10 - 10.30 *Rodríguez Luis Ernesto*
Yellow diploid potato breeding through participatory selection for food security in Colombia
- 10.30 - 10.50 *del Rio Alfonso*
Use of potato genetic diversity to challenge abiotic stresses in the high Andes of Peru
- 10.50 - 11.20 **COFFEE BREAK AND POSTER DISPLAY**

Session 6 – DIVERSITY, BREEDING AND SELECTION - 2 (cont.)

- 11.20 - 11.40 *Ruiz de Arcaute Roberto*
Accelerated breeding choosing best parentals for chip quality
- 11.40 - 12.00 *Slater Tony*
Improving the selection efficiency in potato breeding
- 12.00 - 12.20 *Getahun Baye*
Assessment of genetic diversity and relationships for agronomic nitrogen use efficiency and related traits in potato (*Solanum tuberosum* L.) under contrasting N regimes
- 12.20 - 12.40 *Young Vanessa*
James Hutton Limited: The link between molecular marker development and commercialisation
- 12.40 - 13.00 *Discussion and closing ceremony*
- 13.00 **LUNCH**

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Top latest publications

Aversano, R., Contaldi, F., Ercolano, M. R., Grosso, V., Iorizzo, M., Tatino, F., et al. (2015). *The Solanum commersonii Genome Sequence Provides Insights into Adaptation to Stress Conditions and Genome Evolution of Wild Potato Relatives*. *The Plant Cell*, tpc.114.135954-60.

Cabañes, F. J., Sanseverino, W., Castellá, G., Bragulat, M. R., Cigliano, R. A., & Sánchez, A. (2015). *Rapid genome resequencing of an atoxigenic strain of Aspergillus carbonarius*. *Scientific Reports*, 5.

Sanseverino, W., Hénaff, E., Vives, C., Pinosio, S., Burgos-Paz, W., Morgante, M., et al. (2015). *Transposon Insertions, Structural Variations, and SNPs Contribute to the Evolution of the Melon Genome*. *Molecular Biology and Evolution*, msv152.

Pérez-García, P., Ma, Y., Yanovsky, M. J., & Mas, P. (2015). *Time-dependent sequestration of RVE8 by LNK proteins shapes the diurnal oscillation of anthocyanin biosynthesis*. *Proceedings of the National Academy of Sciences*, 112(16), 5249-5253.

Takahashi, N., Hirata, Y., Aihara, K., & Mas, P. (2015). *A Hierarchical Multi-oscillator Network Orchestrates the Arabidopsis Circadian System*. *Cell*, 163(1), 148-159.

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ORAL PRESENTATIONS

2015 EAPR Breeding and Varietal Assessment
Section and EUCARPIA Section Potatoes

18TH JOINT MEETING

Vico Equense, Italy - November 15th – 18th, 2015

Borrowing from neighbors: comparative genomics approaches for resistance gene discovery and deployment

Van Eck L, Bradeen JM

Department of Plant Pathology and the Stakman-Borlaug Center (SBC) for Sustainable Plant Health, University of Minnesota, St. Paul, MN, United States, jbradeen@umn.edu

Comparative genomics has revolutionized modern plant improvement, allowing researchers to use genetic knowledge gained from one species to identify, isolate, and deploy genes of related function in a second species. Genes affording disease resistance traits are of particular interest to plant improvement efforts. Disease resistance proteins of the NB-LRR type act as cellular sentinels and switches, identifying pathogens and activating defense responses. Each plant genome encodes dozens to hundreds of NB-LRR genes. Because these genes change to keep pace with pathogens, they are among the fastest evolving plant genes. These attributes combine to limit the utility of traditional comparative genomics approaches for discovery and deployment of disease resistance. Working in the Rosaceae (apple, peach, and strawberry), we have developed a new and novel approach to comparative genomics for NB-LRR genes. A standardized analytical pipeline for assigning NB-LRR genes from across species to diversity bins, application of phylogenetic approaches, and creation of a standardized, cross-species terminology has yielded a community resource positioned for rapid discovery of novel resistance phenotypes in genebank collections, development of markers for marker-assisted breeding, and efficient NB-LRR gene mapping and cloning. In the Rosaceae, our approach has revealed candidates for non-host resistance, identified allelic diversification patterns associated with economically important traits, and yielded hypotheses about the impact of plant domestication and large-scale cultivation on NB-LRR evolution. Next steps in this project include automating our informatics and data visualization pipelines and application to the Solanaceae. We have particular interest in discovering novel alleles at known late blight resistance loci in genebank collections of potato species. The resulting community resources from these efforts are both adaptable and expandable. Future efforts will include deep allele mining of Rosaceae and Solanaceae genebank collections and expansion/refinement of our analytical frameworks.

Application of novel enrichment and sequencing approaches for rapid marker development and fine-mapping of a broad-spectrum blight resistance locus from *S. verrucosum*

Chen X¹, Bayer M², Bradshaw JE¹, Baker K¹, Armstrong M¹, McLeanK¹, Harrower B¹, Lewandowska D¹, Witek K³, Jones JD³, Lees AK¹, Bryan JI¹, Hein I¹

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Late blight disease, caused by the oomycete pathogen *Phytophthora infestans*, remains a major threat to potato production worldwide. We have characterised a novel, broad-spectrum resistance gene to *P. infestans* from the diploid, Mexican, tuber bearing inbreeding species *Solanum verrucosum*. Utilising the sequenced potato genome, we have devised enrichment and sequencing strategies for the rapid, SNP-based, mapping of the resistance in a bulked-segregant analysis. To reduce the genome complexity and to achieve sequencing depth sufficient for SNP calling, two specific bait libraries have been generated. The first library, referred to as GenSeq, enables mapping of any dominant trait without prior knowledge of the underpinning gene or gene family and is based on enrichment, sequencing and SNP discovery within COS genes alongside additional, single or low-copy genes that are distributed equally across the potato chromosomes. The second library, referred to as RenSeq, specifically targets NB-LRR genes for sequencing and SNP discovery. Bulk segregant analysis of the datasets revealed the resistance gene resides on Chr. 9. SNPs from COS markers and NB-LRRs at the target region were pursued and converted into allele-specific PCR markers for genotyping of the segregating population. Analysis of genotypic data in combination with phenotypic data localised the resistance to a 10 MB interval between genes DMG400010287 and DMG400017146 on Chr. 9 according to DM genome. Additional sequence data for genes within this interval are currently being generated to facilitate the fine-mapping of the resistance.

Late blight resistance genes in complex interspecific potato hybrids

Fadina OA¹, Beketova MP¹, Sokolova EA¹, Kuznetsova MA², Rogozina EV³, Khavkin EE¹

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² Institute of Phytopathology, Bol'shiye Vyazemy, Moscow region, Russia

³ N.I. Vavilov Institute of Plant Industry, St. Petersburg, Russia

Pyramiding late blight resistance genes of diverse race specificity by backcrossing (Kim et al., 2012; Rietman et al., 2012) and genetic engineering (Haverkort et al., 2009; Jo et al., 2014; Jones et al., 2014; Zhu et al., 2012) has paved the road to new potato cultivars with high, broad-spectrum and durable resistance. Complex interspecific potato hybrids that comprise genetic material from up to eight wild *Solanum* species and manifest high foliar and tuber resistance in field and laboratory tests (Rogozina et al., 2014) are promising sources for breeding new potato cultivars. Forty clones of standard potato varieties and interspecific hybrids were screened with SCAR markers for late blight resistance genes *R1*, *R2/Rpi-blb3*, *R3a*, *R3b*, *RB/Rpi-blb 1*, *Rpi-vnt1* and *Rpi-blb2*. Pedigrees of some hybrids were partly verified with DNA markers specific for *S. bulbocastanum* and *S. demissum* germplasm, *Solanum* genome B and *Rysto* resistance genes. SCAR markers for the *R* gene were verified by cloning the corresponding amplicons and comparing their sequences with the prototype *R* genes and their orthologues already characterized in wild *Solanum* section *Petota*. Multiple alignments of these sequences support the initial suggestion that when stacked in the interspecies hybrids, the *R* genes from different *Solanum* species considerably enhance late blight resistance of the hybrids under study. Although resistance in the laboratory tests was significantly related to the presence of SCAR markers, we did not observe a clear-cut correlation between the level of resistance and the number of markers per hybrid. Evidently a considerable portion of the registered resistance depended on as yet uncharacterized genes and the genes that we failed to recognize because of the limited set of markers. Such discrepancy was especially pronounced in the case of insufficiently researched wild progenitors of interspecific hybrids, such as South American *Solanum* species.

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Rpi-gene mediated late blight resistance in potato tubers

Hutten RCB, Wouters TCAE, Van Der Zweep M, Vossen JH, Visser RGF

Plant Breeding, Wageningen University & Research centre, The Netherlands.
Email: ronald.hutten@wur.nl

Late blight (*Phytophthora infestans*) is a devastating disease that infects both foliage and tubers of potato. Within the DuRPh (Durable Resistance against *Phytophthora*) project the late blight resistance genes *Rpi-blb1*, *Rpi-blb3*, *Rpi-*chc1**, *Rpi-vnt1* and *Rpi-edn2*, driven by their native promoter, were transferred to potato variety Désirée. For each *Rpi*-gene a set of 3 true-to-type transformants was selected with a level of foliage late blight resistance comparable to conventional breeding material containing the same *Rpi*-gene. The identical genetic background of all transformants allows accurate characterization of the effect on tuber blight resistance of the individual *Rpi*-genes relative to each other and to the wildtype variety Désirée. Tuber slices from field grown tubers were inoculated with *P. infestans* isolates 'Katshaar' (avirulent to all 5 *Rpi*-genes) and 'IPO-C' (virulent to *Rpi-blb3*). The amount of newly formed sporangia after droplet inoculation was estimated and compared to the amount of sporangia formed on variety Désirée. Compared to wildtype Désirée, isolate 'Katshaar' produced significant less sporangia on transformants containing *Rpi-blb3*, *Rpi-*chc1** or *Rpi-vnt1*. Isolate 'IPO-C' produced significant less sporangia on transformants containing *Rpi-*chc1** or *Rpi-vnt1*. From the experiments can be concluded that late blight resistance genes *Rpi-blb3*, *Rpi-*chc1** and *Rpi-edn2* are expressed in the tuber.

Novel *R* gene differential set plants for monitoring virulence in *P. infestans*

Vossen JH¹, Kessel GJK², Hutten RBC¹, Boonenkamp P², Jacobsen E¹, Visser RGF¹

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Potato cultivation is highly dependent on biocide applications to protect the crop against a wide range of diseases and pests. Breeding for resistance is therefore economically highly relevant and mandatory to improve sustainability of potato production. In the *Solanum* germplasm natural resistance against most diseases is amply available but introgression of the resistance (*R*) genes into potato varieties is very complicated and time consuming due to the tetraploid heterozygous genome of potato. Introduction of these cisgenic *R* genes through *Agrobacterium* mediated transformation into established varieties is a very attractive approach that bypasses several limitations from classical introgression breeding. In recent years more than 20 *R* genes providing resistance against late blight caused by *Phytophthora infestans*, have been cloned, among which the broad spectrum genes *Rpi-chc1*, *Rpi-vnt1*, *Rpi-edn2*, *Rpi-blb3* and *Rpi-sto1*. All these cloned late blight *R* genes encode NB-LRR proteins that recognize different pathogen derived effectors. The introduction of (combinations) of these *R* genes into varieties like Désirée, Première, Aveka, Atlantic and Bintje by marker-assisted and marker-free transformation has resulted in sets of isogenic potato plants that only differ in the introduced *R* gene(s). These so called "differential sets" are excellent tools to monitor virulence in *P. infestans* populations. The compilation of monogenic and digenic differential sets will be explained. Also several examples of the application of the differential sets in *P. infestans* virulence monitoring will be presented.

Stability of resistance conferred by pyramiding two QRLs for *G. pallida* Pa2/3

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The potato cyst nematodes (PCN) *Globodera rostochiensis* and *Globodera pallida* are significant pests of potatoes worldwide. The most effective control methods are crop rotation and the deployment of resistant varieties. Complete resistance to *G. rostochiensis* based a single resistant gene, has successfully been integrated into many varieties. However resistance to *G. pallida* has not been as successful to date with current varieties only exhibiting partial resistance. Combining partially effective quantitative resistance loci (QRLs) can increase the strength and breadth of the resistance. We have previously demonstrated an additive effect on resistance on combining two QRLs from *Solanum tuberosum* spp *andigena* (*GpaIVsadg*) and *Solanum vernei* (*GpaV*). However populations of *G. pallida* can be quite divergent and it was unclear whether the relative effects of the individual QRLs and the combined additive effect would be consistent across different *G. pallida* Pa2/3 populations. Using a mapping population segregating for both QTLs, we examined the effect of the QRLs individually and combined on four UK-derived field populations of *G. pallida* pathotype Pa2/3, and found that the relative effects of the individual QRLs and the additive effect of the combination were consistent across all populations.

Molecular and genomic insights on potato resistance to bacterial wilt

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Ralstonia solanacearum causes bacterial wilt on more than 200 plant species. This disease is devastating in tropical and subtropical countries due to the high persistence of the bacterium in the fields, its wide host range and the lack of resistances in commercial varieties. *Solanum commersonii* is a wild potato species that exhibits significant resistance to bacterial wilt (also called brown rot in potato).

We will present two complementary approaches to gain insight into the defense mechanisms used by *S. commersonii* accessions with contrasting resistance to the pathogen.

First, we have developed a simple, high-throughput method that allows quantitative detection of bacterial colonisation, using a light-emitting *R. solanacearum* reporter strain. This non-disruptive methodology also allows for the identification of latent infections on symptomless plants and can be applied to evaluate potato germplasm for resistance to bacterial wilt in breeding programmes.

Second, we are using deep sequencing of RNAs (RNA-seq) to determine the potato and *R. solanacearum* genes that are key for the establishment of infection. We made a de novo potato transcriptome assembly and identified some 3,000 unigenes with no homologues in the *Solanum tuberosum* Group Phureja DM sequenced genome. These *S. commersonii*-specific genes constitute the majority of differentially-expressed transcripts upon inoculation. Interestingly, genes controlling the metabolism of certain plant defence hormones were differentially regulated in the susceptible and resistant plant accessions. We will describe these results and our current experiments to determine bacterial gene expression *in planta*.

Soft rot resistance in potato lines derived from a somatic hybrid with *Solanum palustre*

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Pectobacterium atrosepticum can cause two distinct diseases in potato; black leg of foliage and soft rot in tubers. Both can result in severe economic losses to the potato crop. Resistance to soft rot was identified in a hexaploid somatic hybrid between the non-tuber-bearing diploid species *Solanum palustre* and cultivated tetraploid potato (*S. tuberosum*). Thirteen pentaploid hybrids were obtained from hybridisation of the hexaploid somatic hybrid with cultivated potato. All pentaploid lines consistently exhibited high resistance to tuber soft rot. Further hybridisation of three pentaploid lines with cultivated potato resulted in progeny lines with DNA content ranging from tetraploid to pentaploid as determined by flow cytometry. Field-grown tubers of these second-generation progeny lines showed a wide spectrum of soft rot resistance when inoculated with *P. atrosepticum* ICMP8975. Selected tetraploid/near-tetraploid progeny have retained soft rot resistance in field-grown tubers in bioassays over four years. We are currently in the process of identifying genome regions of *S. palustre*, present in these lines, with the aim to develop markers for introgression of the resistance into potato cultivars. We are making use of a vast amount of potato genome and transcriptome data generated over recent years. In the near future we will exploit this growing resource further to improve our understanding of the mechanisms of *Pectobacterium* resistance in the somatic hybrid derived lines.

Most QTL for leaf sucrose content map to positions similar to positions of QTL for tuber starch content in diploid potato

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In growing potato tubers, starch biosynthesis is the dominant pathway of carbohydrate metabolism. It is under control of many genetic and environmental factors. In the present study, using a diploid potato mapping population and Diversity Array Technology (DARt) markers, we identified twelve quantitative trait loci (QTL) for tuber starch content on seven potato chromosomes: I, II, III, VIII, X, XI, and XII. The most important QTL spanned a wide region of chromosome I (42.0–104.6 cM) with peaks at 63 cM and 84 cM which explained 17.6% and 19.2% of the phenotypic variation, respectively. ADP-glucose pyrophosphorylase (AGPase) is the key enzyme for starch biosynthesis. The gene encoding the large subunit of this enzyme, AGPaseS-a, was localized to chromosome I at 102.3 cM and accounted for 15.2% of the variance in tuber starch content. A more than 100-fold higher expression of this gene was observed in RT-qPCR assay in plants with the marker allele AGPaseS-a1334. This study is the first to report QTL for sucrose content in potato leaves. QTL for sucrose content in leaves were located on eight potato chromosomes: I, II, III, V, VIII, IX, X and XII. In 5-week-old plants, only one QTL for leaf sucrose content was detected after 8 h of darkness; four QTL were detected after 8 h of illumination. In 11-week-old plants, six and three QTLs were identified after dark and light phases, respectively. Of fourteen QTL for leaf sucrose content, eleven mapped to positions that were similar to QTL for tuber starch content. Our results provide knowledge on genetics of tuber starch accumulation and indicate its relationship with genetic factors controlling leaf sucrose content.

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Identification of new alleles for carotenoid biosynthesis and degradation in a collection of potato varieties

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Genetics of carotenoid accumulation by potato tubers has intensely been investigated in recent years, due to the potential benefits of these compounds on human health. It has been demonstrated the variability, within wild and cultivated potato germplasm, of the amount and type of carotenoids accumulated; the work carried out aimed to disclose new allelic variants of the main carotenoid biosynthetic genes influencing carotenoid accumulation in tubers, and to determine their phenotypic effects, taking the newly-released, yellow-fleshed cv. Melrose as a starting point.

Melrose cv. was characterized, as well as its agronomical and productive value, also for potential nutritional value based on its carotenoid content and type; these data were compared with a collection of ten 2x and 4x potato genotypes representing the contrasting tuber flesh color, grown in the same years and conditions and including the wild species *S. chacoense*.

Then, cv. Melrose was crossed to either white-fleshed potato cv. Daifla, or self-pollinated to obtain an S1 progeny with the aim to analyze the flesh color segregation; this S1 progeny at harvest was analyzed for flesh color, carotenoid content and allele constitution at the *CHY2* and *ZEP* loci.

The allele analysis at the main loci determining carotenoid amount and type (*CHY2*, *ZEP*, *CCD4*) was extended to all the ten potato clones used as comparison.

In particular, the allelic variability for *CCD4* locus has been analyzed in the frame of this work for the first time. Besides, new alleles were identified at the *ZEP* locus. Compared to other variants at the same loci, some of the new alleles identified displayed significant variations in transcript levels, correlated with differences in the amount and profile of tuber carotenoids, suggesting their relevance in carotenoid accumulation.

Metabolic engineering of carotenoids in potato affects ABA metabolism and tuber shelf-life

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Carotenoids are secondary metabolites involved, in animals, in the prevention of several animal diseases including cancers and cardio-vascular pathologies. In plants, they play essential functions as photosynthetic pigments in leaves, secondary metabolites in fruits and flowers, and hormone precursors (ABA and strigolactones); three out of them (*a*-/*b*-carotene, *b*-cryptoxanthin) cover a fundamental role in human nutrition as precursors of vitamin A.

We have previously generated potato transgenics enriched in β -carotene and total carotenoids, so defined "Golden", by expressing simultaneously three genes of bacterial origin (*CrtB*, phytoene synthase; *CrtI*, phytoene desaturase; *CrtY*, lycopene *b*-cyclase). In a different approach, potato tubers have been engineered with the "Or" gene from cauliflower, resulting in β -carotene and total carotenoid levels. In both cases, transgenic tubers also displayed higher levels in ABA. We have performed a global profiling at transcriptional (RNA-Seq), metabolomic (LC-MS) and phenomic level on all these materials. Several nutritional (aa, vitamins, flavonoids, tocopherols, quinones) and antinutritional (amides, alkaloids) resulted altered in "Or" and "Golden" tubers, suggesting a dramatic remodeling of primary and secondary metabolisms. Integration of Gene expression profiles, integrated and high-throughput metabolomics, revealed unscheduled transcript-metabolite correlations and shed light on novel co-regulatory dynamics which emerged in tuber metabolism and, limited to "Golden" samples, in post-harvest storage resulting in an elongated shelf-life.

A strong influence of carotenoid/ABA accumulation on tuber maturation kinetics was observed, while GO enrichment analysis allowed identification of gene classes specifically regulated in "Golden"/"Or" tubers. Overall, these analyses revealed the central role of carotenoids in regulating tuber metabolism and development and large-scale network analysis proved to be a valuable approach for rational design of new biofortified crops by through identification of higher correlative power nodes (hubs of the network), as potential targets in future breeding programs.

Evaluating the yield and tuber quality performance of eight European cultivars in Pietermaritzburg to improve potato production in South Africa

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Potato has become one of the most economical produce for addressing food security challenges in South Africa due to its reputation as the highest-yielding crop. Developments in the local potato industry have resulted in a number of foreign cultivars introduced into the country in an effort to improve productivity. However, the performance of many cultivars has not been well-established as most are from differing agro-ecological conditions. The aim of this study was to evaluate the yield and potato quality performance of eight cultivars introduced in South Africa within the last decade. These were Mondial, Panamera, Electra, Savanna, Navigator, Melanto, Sifra and Innovator, which are of white to yellow flesh colour. Experiment was carried-out in Pietermaritzburg, under temperate agro-climatic conditions, using a Randomised Complete Block Design. Results showed significant ($p \leq 0.05$) differences in most of the parameters measured, with Innovator having the highest dry matter content (20%) and specific gravity (1.063). The highest and lowest yields achieved were of Electra and Navigator, yielding 21 and 54 t ha⁻¹, respectively. All cultivars had a low occurrence of internal defects ($\leq 10\%$) and produced high marketable yield ($\geq 80\%$). Only Navigator had a processing yield lower than 60%, and Melanto and Electra had the lowest dry matter content (16%). These results assert Innovator as a good cultivar for industrial processing of French fries, and suggest five out of the eight cultivars to be economical for production, which may be targeted to improve crop yields in South Africa. Only Navigator, Panamera and Savanna did not exceed the 35 t ha⁻¹ yields currently achieved in the country.

Unraveling drought stress in potato

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Potato (*Solanum tuberosum*) is the most important non-grain food commodity consumed all over the world, but it is highly sensitive to drought. Because of climate change the need for improved drought tolerance in potato has gained global research and agricultural interest. Tolerance to drought stress is a complex trait determined by many minor effect loci. In different experiments conducted in the greenhouse from 2012 – 2015 using a biparental population segregating for drought response and potato cultivars with contrasting drought responses we mapped many QTL of which the transcriptional regulation contributes significantly to observed phenotypes. Under drought, a major eQTL hotspot was identified on chromosome V. Nuclear factor γ subunit C4 (*NF-Y-C4*) was identified as the key candidate for adaptation to drought in the biparental population studied. It is well known that one of the major physiological processes affected by drought stress is carbon partitioning. Under severe drought stress tuber setting and growth and thus yield is strongly affected. Carbon partitioning and its relation to yield involve many processes including photosynthesis, sucrose metabolism, transport of metabolites, and starch biosynthesis. These were studied and our results indicate that one of the most severe effects of drought stress is the arrest of stolon differentiation and formation of tubers. Our phenotypic studies also point to some physiological traits that affect photosynthesis and eventual tuber yield. Some of the key enzymes and genes studied may be inclusive breeding targets for drought tolerance in potato.

Stress-tolerant *Solanum commersonii*: genome sequence and manipulation

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S. commersonii possesses several resistance traits not found in cultivated potato and its freezing tolerance and capacity to cold acclimate is its main attractive features. We sequenced and de novo assembled the genome of *S. commersonii* to 105x coverage using Illumina technology producing ~830 Mb draft genome with N50 of 44,303bp. Gene annotation by ab initio prediction supported by RNA-seq data allowed the production of a catalogue of 1,703 predicted microRNAs, 18,882 long noncoding RNAs, 39,290 protein-coding genes with a significant repertoire of non-redundant nucleotide binding site-encoding genes and 126 cold-related genes that are lacking in *S. tuberosum*. In addition, phylogenetic analyses offered new insights into recent duplications and divergence between *S. commersonii* and the domesticated potato. Whole-genome expression data of frost stress acclimated (AC) and non-acclimated (NAC) plants highlighted enhanced expression of genes affecting reactive oxygen species (ROS) scavenging enzymes, those involved in cell repair, and those encoding proteins that may function as osmoprotectants. In addition, remarkable features of genes known to be critical in cold signalling pathways, such as C-repeat binding factors (CBFs) and cold responsive (COR) genes, were found. We also used the sequenced *S. commersonii* as model to understand the consequences of autopolyploidization. A high fraction of parental genes changed expression in synthetic tetraploids of *S. commersonii* and an unusually high percentage of differentially expressed genes were found in the pericentromeric regions of several chromosomes (6, 9, 10 and, to a lesser extent, 3, 5 and 11). The information we generated provides insights in elucidating the molecular and evolutionary mechanisms of crop domestication, and in facilitating translation of acquired knowledge into advances in crop stability in light of global climate and environmental changes.

Selection method for salt tolerance in potato

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Due to the growing world population and the associated growing demand for food, there is an increasing need for farmland and irrigation water. A lack of sufficient water of good quality leads to the irrigation with salt water in several countries [1]. Furthermore, there is an increasing siltation of farmland. Worldwide 7% of the farmland is salt-affected, which corresponds to 1 billion ha.

Agrico sells seed potatoes to more than 75 countries worldwide, among which are countries that make use of salty irrigation water, for example Egypt and Israel. Therefore, there is a growing demand for salt tolerant potato varieties.

In order to determine the salt tolerance of the available Agrico varieties and to start breeding for salt tolerance a good test or selection method is required. In 2013 we started with a small scale pot trial with 10 varieties and two salt concentrations. Although this trial was not optimal, there were indications of differences in tolerance between varieties, which also has been described in literature [2]. In 2014, this trial was extended to a pot trial with more varieties and to a field trial. Here we present the results of this project, where the main conclusion is that the pot trials show significant differences between varieties (yield), but no correlation between two consecutive years. Furthermore, there was no correlation with data of the field trial. Repeatability of the field trial will be determined in 2015.

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Sensing the plants: can we detect stress?

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Recent development in imaging sensors has offered the chance of exploiting high throughput phenotyping (HTP) and crop monitoring non-destructively by gaining information on canopy state and variation.

We have used Infra-red thermography as a sensitive and HTP method to understand variation in canopy temperature in potato under field conditions and have demonstrated its reproducibility and consistency at different plant growth stages and times of the day. Further, the phenotypic results were used to identify QTL regions for canopy temperature and our study suggests complex interactions between crop productivity traits and stomatal conductance.

Data from canopy temperature is also being related to morpho-physiological characteristics by using 3D imaging approach in field and exploring the possibility for understanding growth and development in potato.

Be aware: diploid hybrid potatoes are coming!

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In the last hundred years the breeding of many crop species has been accelerated after the implementation of hybrid breeding. For potato, attempts have been made by using prickle pollination to induce haploid plants from egg-cells (gynogenesis) or to generate haploid plants from anther culture (androgenesis). Though many haploids and double-haploid plants were generated, these homozygous diploid plants were showing severe reduced vigour and fertility, which has blocked the development of inbreeding systems in potato. Alternatively, homozygous plants could be generated by selfings. However, diploid potato is self-incompatible and the progress in increasing homozygosity of tetraploid potato is too slow. As a consequence, it has long been perceived impossible to develop a hybrid breeding system for potato. Recently, major breakthroughs have been achieved:

- 1) The self-incompatibility of diploid potatoes has been overcome by introgressing the *Sli-gene* from *S. chacoense*
- 2) Inbreeding depression has been overcome by many rounds of crossings, selections and selfings.

The first essentially homozygous self-compatible potato genotypes have already been generated in 2012 and the vigour of these plants was still quite low. The level of homozygosity was assessed by using SNP-markers to investigate the effect of inbreeding on phenotypic traits. A strong correlation between overall level of homozygosity and lack of self-compatibility was observed. By new series of crosses, selections and selfings the agronomic performance of the inbred lines continuously improved.

In the summer season of 2014 and in the winter of 2014/15 the most vigorous inbred plants were used as parents to generate experimental hybrids. The agronomic performance of these experimental hybrids was tested in a replicate field trial in 2015. The yield and tuber quality of these hybrids will be presented.

Progress toward the development of recombinant inbred lines

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Recombinant inbred lines (RILs) offer new opportunities for mapping traits of interest to potato breeders. We are developing a set of six RILs, which will comprise a nested association mapping population. The common parent is M6, an inbred line of the diploid wild relative *Solanum chacoense*. It is homozygous for a dominant self-incompatibility inhibitor. Other germplasm in the RILs includes *S. tuberosum* Tuberosum Group, *S. tuberosum* Phureja Group, and *S. berthaultii*. The parents of the RILs have been selected based on tuber morphology and yield, disease resistance, and processing quality. We plan to create 100 F6 inbred lines of each of the six RIL populations. Currently, we have populations ranging from the F2 to F6 generations. During inbreeding, a reduction in vigor has been observed, as expected. However, the major challenge is maintaining adequate flower production and fertility for seed production. The RILs will be shared with the research community for phenotyping and mapping.

Progress in diploid potato breeding with self-compatibility

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Breeding at the tetraploid level in potato is an inefficient process. Diploid breeding offers advantages that are compounded by the introgression of self-fertility. Using the dominant self-incompatibility inhibitor *Sli* gene from *Solanum chacoense* (M6) as well as other sources, we are introgressing the self-incompatibility trait into diploid species hybrids and dihaploids of *S. tuberosum* for the purpose of creating diploid inbred lines. We have developed a recurrent selection population for photoperiod adaptability, self-compatibility and cultivated tuber traits. The germplasm is composed of a mixture of six species (*S. berthaultii*, *S. chacoense*, *S. microdontum*, *S. tuberosum* Grp. Phureja, *S. tarijense* and *S. tuberosum*). The foundation lines and selections from two cycles of recurrent selection have been SNP genotyped to monitor population diversity and structure. We also constructed two gynogenic dihaploid ($2n = 2x = 24$) populations from cvs. Atlantic and Superior. The population sizes were 134 and 67 individuals, respectively. Field trials were conducted in Michigan in 2014-15. Multiple traits were evaluated to study vigor and phenotypic traits associated with fitness (tuber emergence, total tuber yield, average tuber weight, number of tubers per plant, plant height, vine vigor, specific gravity, and tuber shape). Although extreme variation for emergence created bias in the phenotypic analysis, these dihaploid populations also showed extreme phenotypic variation for vigor, tuber number and total yield. The best performing dihaploids have been used as donors to introgress self compatibility from various self-compatible species hybrids. Among the selections from the recurrent selection and species-diploids hybrids ($2x-2x$ crosses) made in the field in 2014, genome-wide SNPs were used to separate tetraploid from diploid progeny. The frequency of tetraploid progeny suggests a high frequency of $2n$ gametes in the germplasm pool.

Somatic hybrids in sexual hybridization of potato

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The sexual cross ability of two somatic hybrids was evaluated. Two somatic hybrids (marked as SH292 and SH1003) with high level of resistance to *Phytophthora infestans* (PI) were employed. Both originated from protoplast fusion of 1EBN diploid *Solanum pinnatisectum* 8166 + 2EBN *Solanum tuberosum* dihaploid 299. Four pollinators (tbr cvs: Barbora, Valfi, Flavie, Karin) were successful. Overall 19 berries, 343 seeds, 90 seedlings in crossing generation 1 (CG) were gained from more than four hundred pollinated flowers. Seeds germination was 21 % in average. Almost all genotypes bloomed and had good pollen fertility. More than one third of genotypes had long stolons. Chosen genotypes (after laboratory test on Petri dish) showed repeatedly high level of resistance to PI. In the field, most of genotypes were moderately resistant and three genotypes showed even high level. One hybrid with moderate level of resistance to PI had very good table quality.

In CG2, more than one thousand flowers were pollinated. As result of that pollination, 27 berries, 1032 seeds and 418 seedlings were obtained. Seeds germination reached 41 %. One third of them flowered; half of them had good pollen productivity as well as female fertility. No genotype had stolons. Chosen genotypes were again highly resistant to PI in laboratory test. Reciprocal crosses between somatic hybrids failed and the same situation was in reciprocal crosses of them with two hybrids of CG1.

During the years 2009 – 2014, the laboratory test on Petri dish showed good predictive value for the first selection of breeding materials.

This work (supported by RO1011) demonstrated the possibility of somatic hybrids to be sexually crossed with *S. tuberosum* cultivars and also proved that the resistance to PI was passed from wild *S. pinnatisectum* 8166 to somatic hybrids SH292 and SH1003 and also to CG1 and CG2 progenies.

Genetic diversity and composition of *S. × michoacanum* (+) *S. tuberosum* somatic hybrids and 4x *S. × michoacanum* evaluated by Diversity Array Technology and PCR markers

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The somatic fusion process leads to new nuclear and cytoplasmic combinations and generates structural changes in somatic hybrids' genomes. Information about diversity and composition of a somatic hybrid genome is essential for its efficient exploitation. DArT markers in combinations with available genetic and physical maps of potato permits detailed analysis of composition of nuclear DNA of somatic hybrid. Markers specific to chloroplast DNA (cpDNA) and mitochondrial DNA (mtDNA) were used for analysis of changes within the organellar genomes.

The goal of this study was to determine the composition of nuclear and cytoplasmic genomes of 97 *S. × michoacanum* (+) *S. tuberosum* [mch (+) tbr] somatic hybrids and 11 4x *S. × michoacanum* [4x mch] lines obtained from autofusion of 2x mch. We tried to explain the low number of somatic hybrids resistant to *Phytophthora infestans*.

Out of 5,358 DArT markers generated in a single assay, greater than 2,000 markers were polymorphic between parents, of which more than 1,500 have a known chromosomal localization on potato genetic or physical maps. DArT markers were distributed along the entire length of all the 12 chromosomes of hybrids. We observed the elimination of DNA of wild and tbr fusion components. The nuclear genomes of individual somatic hybrids were diversified with a predominance of tbr-specific markers. From 97 mch (+) tbr somatic hybrids, two hybrids and all 11 4x mch were resistant to late blight. The analysis of the structure of chromosome VII, where the resistance gene *Rpi-mch1* is located, indicated that the lack of resistance of somatic hybrids might be potentially caused by the loss of one homologous mch chromosome containing the *Rpi-mch1* gene, gene silencing or loss of other genetic factors important for resistance. Random and non-random segregations of cp- and mtDNA with no recombination in their structure were noted. Statistical analysis indicated a significant positive correlation between cytoplasmic DNA type D and the percentage of nuclear DArT markers specific to the mch parent.

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Genotypic differences in solar radiation used efficiency intercepted by plant canopy and its relation to early tuber bulking in a CxE diploid potato population

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Drought is a complex trait and both escape from drought and drought avoidance at all stages of development are thought to be genetically controlled. Early tuber bulking is important for drought escape, but is generally corresponding to a lower partition ratio of dry matter to root (Iwama 2008, Deguchi *et al.* 2010, 2015), and reduces root growth, resulting in weakened drought avoidance. We need to break down the tradeoff relation between early tuber bulking and root growth to breed new genotypes with both drought escape and drought avoidance. In this study we focused on a genetic difference in plant dry matter production in relation to aboveground traits. We measured an accumulated solar radiation intercepted by plant canopy (SRi), and increase of shoot and tuber dry weight (DW) till 35 days after sprouting (DAS) within a CxE diploid potato population (89 genotypes) planted in an experimental field of Hokkaido University. SRi was estimated from the measured vegetation cover ratio at three intervals till 35 DAS by using a digital camera equipped with a near-infrared light filter. At 35 DAS tuber DW positively correlated not only with partition ratio of dry matter to tuber (PRT, $R^2=0.849^{***}$), but also with total DW ($R^2=0.782^{***}$) which related positively to the ratio of total DW per unit SRi (solar radiation used efficiency intercepted by plant canopy, RUE, $R^2=0.606^{***}$). In multiple regression analysis of tuber DW at 35 DAS (dependent variable, $R^2=0.948^{***}$), the partial regression coefficient of independent variables was largest in PRT (0.638^{***}), followed by RUE (0.406^{***}) and smallest in SRi (0.270^{***}). The present results suggest that we may be able to find QTL(s) regulating RUE independently with PRT and thus plant maturity class.

Towards durable resistance against *P. infestans*

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Potato suffers from the devastating late blight disease, which is caused by *Phytophthora infestans*. This oomycete pathogen secretes host-translocated RXLR effectors that include avirulence (AVR) proteins, which are targeted by resistance (R) proteins from wild *Solanum* species. We and others have localized and mapped many *Rpi*-genes on the genome and cloned a range of these genes. They have in common that they belong to the NB-LRR class of intracellular immune receptors. However, virulent races of the pathogen quickly evolve to evade recognition by these cytoplasmic immune receptors. Another, yet unexploited, layer of apoplastic immunity occurs at the surface of plant cells and generally has a broader spectrum and is based on recognition of conserved proteins of pathogens. We have used this effectoromics strategy to show that the receptor-like protein ELR from the wild potato *Solanum microdontum* mediates extracellular recognition of the elicitor domain, a molecular pattern that is conserved in *Phytophthora* species. ELR mediates broad-spectrum recognition of elicitor proteins from several *Phytophthora* species, including from *P. infestans*. Transfer of ELR into cultivated potato resulted in enhanced resistance to *P. infestans*. When having R-genes available stacking of those genes is a defense strategy that is highly successful in wild relatives of potato and thus also seems a good approach for creating new varieties by introgressing multiple new late blight R genes from related *Solanum* species. Introgression can be done by classical means or by more advanced means such as genetic modification preferably by using cisgenesis. In order to achieve truly durable resistance we believe it is necessary to combine different resistance mechanisms. One of these approaches is focused on looking at genes involved in the signaling pathway once the late blight disease is evolving in susceptible plants. Another approach focusses on so called *Susceptibility (S-)* genes. Disabling plant susceptibility (*S*)-genes results in resistance to pathogens. We have tested in potato the susceptibility function of several *S*-genes identified in Arabidopsis and demonstrated that silencing orthologs of specific Arabidopsis genes in potato gives rise to resistance against late blight. We do believe that combining the different mentioned approaches for obtaining resistant varieties in combination with a thorough knowledge of the pathogen population at the growing sites will lead to a broader and potentially more durable resistance to this devastating plant pathogen.

New superior parental material developed by pre-breeding from potato collection at the Vavilov Research Institute of Plant Industry (VIR)

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To broaden the genetic basis for conventional potato breeding, a pre-breeding program for introgressing economic traits from South American wild relatives was initiated in the Vavilov Institute of Plant Industry (VIR) in 2000. Accessions of 16 tuber-bearing *Solanum* species: *S. abancayense*, *S. alandiae*, *S. avilesii*, *S. ambosinum*, *S. okadae*, *S. doddssii*, *S. famatinae*, *S. hondelmannii*, *S. gandarillasii*, *S. incamayoense*, *S. marinasense*, *S. multiinterruptum*, *S. multidissectum*, *S. oplocense*, *S. vidaurreiet*, *S. vermei* from Peru, Bolivia and Argentina, were deployed in crosses either with dihaploid of cultivars or other wild species. Preliminary screening revealed that some of these *Solanum* species could be sources of resistance to different pests such as nematodes, late blight, Colorado potato beetle, etc. (Bamberg et al. 1986). The most promising hybrid progenies came from the cross combination dihaploid Atzimba x *S. alandiae* (n=24). Next, best F1 genotypes were crossed to potato varieties. Each of superior BCF1 clones combined the desirable traits from both parents. Field and laboratory tests demonstrated that the resistance of these hybrid clones to late blight and golden potato nematode Ro1 ran together with good appearance and acceptable yield of tubers, starch content and taste. DNA analyses showed that the patterns of race-specific late blight resistance genes in some hybrid clones differed from those in commercial potato cultivars (Khavkin et al. 2014, Rogozina et al. 2012). Currently these hybrid clones are further used as donor parents to develop new potato varieties.

New insights into the role of anthocyanin duplicated genes in response to cold stress in the potato plant

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An interesting phenotype in cold tolerant potato species is the presence of anthocyanins in leaves, while sensitive species are generally not pigmented. Despite this, little is known about anthocyanins associated with cold stress resistance in potato. In our previous work we established the *MYB/bHLH* complex involved in anthocyanin regulation in potato and, in particular, the role of *StAN1* and *StJAF13* in leaf anthocyanin production. In this present study we compared the behavior of the anthocyanin *MYB/bHLH* complex after cold treatment using cold-tolerant species *Solanum commersonii* and three different genotypes of the cultivated potato. Cold treatment caused a slight increase in total anthocyanin content specifically in the wild species, with anthocyanin structural genes and *bHLHs* consistently up regulated. We also detected a 10-fold increase of *AN2* in *S. commersonii*, whereas the expression of its homolog *AN1* did not change. Yeast-Two Hybrid analysis showed that in *S. tuberosum* "Double Fun" *AN1* and *AN2* shared the same *bHLH* partners for interaction. Interestingly, in *S. commersonii* *ScAN1* and *ScAN2* were able to interact with *ScbHLH1*, but not with the *ScJAF13*, indicating that a potential mutation in *ScJAF13* affected the interaction. In silico analysis of the *S. commersonii* genome revealed that the two genes are closely linked on the same chromosome, at a distance of less than 40 Kb. Furthermore, putative gypsy retro-transposons in the inter-genic region were found. These results suggest a possible origin of the two species from an ancestral duplication with accompanying neo-functionalization of *AN2* related to the cold stress response.

Potato genetic studies in *Solanum tuberosum* group Phureja

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The south of Colombia is a center of diversity for diploid potatoes. This germplasm is important for genetic studies and it is used as a genetic resource in potato breeding programs. Since a decade, Colombia started studies in potato genetics, in close scientific collaboration with Christiane Gebhardt from Max Planck Institute. This collaboration has allowed knowing the potato genetic resources and to build scientific capabilities for Colombia. The potato genetics studies started with the construction of genetic map of *S. phureja* and the identification of QTL linkage with resistance to late blight, these studies evolved to studies of association genetics. The potato population employed is suitable for association studies and several complex traits such as resistance to late blight, quality of tuber and nutritional quality have been studied using association mapping analysis. Through candidate gene approach, we found five SNPs at two loci associated with quantitative resistance to late blight. One locus *StTin1* encodes a potato homologue of TMV-induced protein I and presents three SNPs in near complete linkage disequilibrium (LD) and explained 10.5% of phenotypic variation. The locus *StTL15A* encodes a potato homologue of Thylacoid lumen 15 kDa and presents two SNPs in LD and explain 13% of the phenotypic variation. For quality of tuber related with sugar content, association analysis was carried out with 111 SNP markers identified in ten candidate genes, with key function in carbohydrate metabolism. This analysis revealed four SNP markers in the locus *InvGE* from an apoplasmic invertase and one SNP marker in the locus *SssI* from a soluble starch synthase with significant effect in sugar content and frying color. Most of the associated SNPs were low-frequency variants. Currently, we are employing GBS data in order to use them in GWAS analysis in order to develop markers for assisted selection and to look for genetic interactions.

Transcriptome sequencing as a tool to study the potato wart interaction

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Synchytrium endobioticum is an obligate biotrophic fungus without the development of visible mycelium that causes wart diseases in potato. As a quarantine pest, incidence of the fungus causes severe economic losses. Over 40 *S. endobioticum* pathotypes were reported, out of which, pathotypes 1, 2, 6, 8 and 18 are the most important and widespread pathotypes in Europe. So far molecular diagnostic tools to characterize the pathotypes are not available. A small scale next generation sequencing of *S. endobioticum* pathotype 18 resulted in 5,440 ESTs and 427 corresponding genomic contigs. We used this partial genomic and transcriptomic information to generate molecular markers that are able to discriminate some of the pathotypes which so far could only be distinguished based on laborious and time consuming bioassays.

The identified markers shall aid a fast, accurate and unified cataloguing of *S. endobioticum* pathotypes. The marker-based grouping into three classes followed the virulence level of the pathogen, which will also help to forecast severity of an infection. The ESTs and the genomic sequences reported here can also be used in future experiments to understand the interaction of the pathogen and its host plants

Molecular diagnostics of complex agronomic traits in tetraploid potato

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Most characters important for variety development are controlled by multiple genetic and environmental factors. Their phenotypic evaluation requires multiple year and location trials. The efficiency and precision of the selection process can be enhanced by DNA-based markers, which are diagnostic for superior or inferior alleles of quantitative trait loci and are applicable early in the breeding cycle. Linkage and association genetics in experimental and natural populations of varieties and breeding clones were used to develop a first toolbox of diagnostic DNA markers for the complex traits tuber starch content, yield, starch yield, chip quality (reducing sugar content), susceptibility to bruising, resistance to late blight (*Phytophthora infestans*), potato wart (*Synchytrium endobioticum*) and the root cyst nematode *Globodera pallida*. The first diagnostic DNA markers were obtained based on DNA variation in candidate genes. Candidate genes are known or suspected to play a functional role in the targeted trait. For example, genes functional in starch-sugar interconversion were candidates for the traits tuber starch, starch yield and reducing sugar content (chip quality). Novel candidate genes were identified by comparative proteome and transcriptome profiling between groups of tetraploid genotypes with contrasting phenotypes and yielded novel diagnostic markers. More recently, genome wide approaches such as SNP genotyping with the 8.3 k SoICAP SNP array and genotyping by sequencing resulted in further marker-trait associations that can be useful for potato breeding. The results of 30 years of potato genetic and molecular research at the MPI for Plant Breeding Research are ready for translation in practical breeding applications.

Analysis of potato traits in crosses and association panels

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The Potato Genome Sequencing Consortium (PGSC) published the genome of the homozygous DM genotype in 2011, and a much improved version (v4.03) of the genome pseudomolecules was published in 2013. The availability of a full genome sequence for potato has facilitated the development of extensive tools for performing genetic analysis in this important crop. One of the most well-known of these is the SolCAP 8303 SNP array which has been used extensively for trait analysis in segregating populations and association panels. We have recently published a dense linkage map of a diploid cross as well as an analysis of tuber shape and eye depth. Ongoing analysis of this crop focuses on other agronomic traits as well as resistances to late blight and Potato Virus Y. Genotyping by sequencing (GBS) approaches are also a cost-effective method for genotyping in potato and we have used this approach for both linkage and QTL analysis in biparental populations and association mapping in variety panels. Results from these and other analyses suggest that association mapping is a very effective method for the detection of marker trait associations in tetraploid potato. In performing this type of study a careful assessment of population structure is required to avoid spurious associations arising from systematic differences in allele frequencies due to differences in sample ancestries. Various models have been examined, insights from which will be presented. A further development has seen the development and use of capture array technology for genetic and genomic analysis in potato. Such approaches are now being extended using a recently constructed exome wide capture platform that is being used for various mapping studies, as well as the study of diversity and evolution in potato. Results from some recent analyses will be presented.

Haplotype reconstruction in tetraploids using sequence read based alignments

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The dissection of quantitative characters is always challenging. In tetraploid outbreeders the number of segregating alleles, and the number of allele combinations in descendants, severely complicates the identification of markers associated with trait variation. In case of Genome Wide Association Studies (GWAS) the distantly related individuals comprise even higher numbers of allele combinations. Our research does not only aim to identify QTL, but more importantly, to estimate the contribution of the various alleles and allele interactions to the trait value. SNP markers are not necessarily haplotype-specific, and each SNP-allele may be indicative of a diversity of alleles. To establish a direct relation between a trait allele and a DNA marker, the identification of haplotype specific SNPs (hs-SNPs) is desirable. However, there is no guarantee that each allele has hs-SNPs. Therefore it is our aim to develop a method to analyse the composition of the four haplotypes per individual and the many haplotypes in the potato gene pool.

The *StCDF1* and *Ro* locus are involved in the monogenically inherited characters of plant maturity and tuber shape, respectively. It is our aim to identify a complete collection of haplotypes at these loci and to estimate the quantitative contribution of each haplotype to the trait value. In this way we may understand how the continuous variation for plant maturity and tuber shape can be explained. This presentation will outline our progress in haplotype identification using alignment of DNA sequence reads to deduce the linkage phase of sequence variants. Eventually this effort may expand into a better understanding of long range decay of linkage disequilibrium and a genome-wide HapMap in potato. This research is supported by NOW grant 831.14.002 to JHW

A simple approach to score SNP markers in a dosage-dependent fashion in tetraploid potato

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The accurate scoring of SNP markers on genotyping arrays in tetraploid potato is challenging since five allelic states have to be discriminated (AAAA, AAAB, AABB, AB BB, BBBB). So far, this is not easily possible in a reproducible way using the available software solutions. In order to develop such a method, we have genotyped a set of tetraploid potato samples with the new 12K potato Infinium array. Subsequently, we have developed two cluster files for the scoring of the markers in a diploid mode (AA, AB, BB) whereby we have defined the clusters for the three alternative calls in two different ways. The first cluster file was developed as for a diploid organism with calling all heterozygous states (AAAB, AABB, AB BB) as heterozygous. This cluster file is also useful for the scoring of diploid potato. The second cluster file was adjusted in a way that only the AABB calls are called heterozygous while the other clusters were assigned to the respective homozygous classes. After entering the allele calls with the two different cluster files into our marker data base, we have developed bioinformatics tools to extract all five allelic states into a final allele table. We find this approach simple, reliable and accurate to reproducibly call all five allelic states in tetraploid potato.

Yellow diploid potato breeding through participatory selection for food security in Colombia

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Solanum tuberosum group Phureja, is a valuable genetic resource of potato with a center of diversity in Nariño, southern of Colombia, an important cropping area, with high consumer acceptance. Native diploid potato species and varieties of Phureja have been used in potato breeding with good results, but they have used just a small base of Phureja genetic diversity. Native cultivars present high variability in traits like color in flesh and skin, shape, nutrient content, and rusticity that are not frequent in modern cultivars. Breeding of diploid potato for higher yield, nutritional quality and resistance to late blight disease is considered to be one important option to improve the economic stability and daily diet of communities. Breeding potato programs involving the communities in cropping and selecting new varieties has proven to be a powerful change motivator. In a process of participative selection research with male and, especially female, through a participatory selection process, researchers collaborated with native families of Nariño province and participants in Farmer Field Schools. 100 advanced clones of diploid potato of the breeding program were multiplied, from them, 30 advanced clones were selected in a first trial. In a second trial, eight advanced clones based in their yield, yellow skin and flesh color, round shape, shallow eyes, palatability, resistance to late blight, good performance in abiotic stresses, and high nutritional value and acceptance for potato producers were selected. Each trial evaluated eight advanced clones and two commercial controls in 8 environments. The results allowed identifying the genotypes with biggest phenotypic value for each trait studied. The estimated average specific gravity ranged between 1.081 and 1.100 and yield between 32 and 40 t.ha⁻¹. Three genotypes were selected taking into account quality for fresh consumption and for industrial processing. These results are a consequence of F1 greater hybrid vigor. New potato cultivars contain up to 19% more iron, 17% more zinc, and higher level of protein and dietary fibre than the Colombian commercial cultivars. These new potato varieties have become a sustainable option to offset deficiencies in micronutrients. In addition to improved nutritional content, these new varieties are twice as resistant to late blight disease and have shown higher yield.

Use of potato genetic diversity to challenge abiotic stresses in the high Andes of Peru

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Unexpected frost is one of the most serious constraints in the Andean region and, recent climate change models are predicting even more severe and untimely frost episodes. Since agriculture is dependent on climate and sensitive to climate change, work is needed to keep it sustainable. From a long term effort at the US Potato Genebank and the University of Wisconsin-Madison, frost tolerant potatoes with cultivar potential have been developed. These clones originated from initial crosses between two frost hardy species *S. acaule* and *S. commersonii* which were subsequently crossed with cultivated and Andean native potatoes. True seeds from some of the crosses were sent to CIP in 2006 and they were placed and evaluated in fields located in Central and Southern Peru. After several screenings in the highlands we have identified promising selections that are frost hardy and unlike traditional hardy native potatoes in the Andes, these selections are non-bitter potatoes. In addition, they have equal or better yield as compared to the native potato cultivars. We are currently evaluating these clones with and without supplemental calcium application in large scale field trials in Puno, in collaboration with the Peruvian program for agriculture (INIA) and with local farmers. In 2014-2015, cold tolerant selections are being tested at three locations in Puno to verify their levels of hardiness and productivity.

Accelerated breeding choosing best parentals for chip quality

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The potato breeding program developed in APPACALE S.A. since 1992 utilized for crosses reputed parentals that included several characters related with virus resistance, agronomic performance, tubers appearance and quality for fresh or processed food (Isla S et al. 2000). Selection of progenies for PVY resistance involved a MAS scheme developed for routine use in the company (Ortega F and Lopez-Vizcon C, 2012), but for agronomical characters and quality the selection work was an empiric process. To accelerate and improve the selection process, a half-diallel mating design (Griffing B, 1956) was developed involving 7 parentals commonly used in the program were selected for progeny testing (4 of them were commercial varieties and 3 were clones from the program). From crosses, 17 progenies of 21 were obtained, and during two years about 900 individuals from those 17 progenies and 7 parentals were cultivated and evaluated about agronomic performance and quality for processed chips. Field evaluation was made for plants and tubers, and quality was evaluated in a small-scale laboratory for potato processing, including the process of frying chips. Results of these experiences will be presented and discussed.

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Improving the selection efficiency in potato breeding

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Potato breeders consider a large number of traits in the development of new cultivars. A conventional potato breeding strategy uses outcrossing, then screening of derived lines to identify improved cultivars. Recently, we investigated methods, including molecular approaches, to make the program more efficient. Implementation of marker-assisted selection (MAS) was highly desirable and our initial targets for MAS were qualitative disease resistances. We undertook a cost-analysis of MAS and conventional screening to identify if MAS would be cost-effective within a breeding program. As the majority of target traits are quantitative and controlled by several to a very large number of genes, we also looked at methods to address these traits. We investigated a quantitative genetic analysis technique used in livestock breeding to develop estimated breeding values (EBVs) for these complex traits. We then designed a breeding scheme using a combination of MAS, EBVs and conventional screening methods for rapid selection of cultivars with multiple desirable traits, which saw the breeding cycle reduce from over 10 to 4 years. We have now investigated strategies to incorporate genomic selection (GS) in potato breeding. We have found that genetic gain can be accelerated through GS as the breeding cycle can be reduced to 1 year. The application of these new techniques, alongside conventional screening will see significant genetic gain in a range of traits.

Assessment of genetic diversity and relationships for agronomic nitrogen use efficiency and related traits in potato (*Solanum tuberosum* L.) under contrasting N regimes

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One of the strategy of the Ethiopian potato breeding program is to develop new potato cultivars with better resistance, nutritional and market value that provide reliable yields under lower-input conditions. To study the genetic diversity for agronomic nitrogen use efficiency (NUE) and related traits in potato, ninety seven cultivars (eighty-eight Dutch) and nine Ethiopian cultivars were evaluated at two nitrogen levels (40kg/ha, and 120kg/ha) for 18 quantitative traits in Debre-Tabor and Injibara, (Ethiopia) in the 2013 main growing season. Plant height, maximum canopy cover (Vmax), area under the canopy curve (Asum), lower leaf chlorophyll content and NUE were significantly affected by N levels across locations. NUE, tuber dry matter content %, and days to maturity had higher mean values at LN than at HN in both locations. The tuber yield reduction due to N limitation was significant, and tuber number per plant reduced total tuber weight more than average tuber weight especially in Injibara. High phenotypic coefficient of variation, genotypic coefficient of variation, heritability and genetic advance as a percent of mean were observed for tuber number per plant, average tuber weight and NUE in Debre-Tabor and plant height, tuber number per plant and average tuber weight in Injibara under both N regimes. Strong phenotypic correlation coefficients were observed between NUE and tuber number per plant, days to maturity, tuber dry matter %, Vmax and Asum under both low and high N input conditions. Dutch cultivars showed rapid initial canopy development and matured early, compared to the Ethiopian cultivars at both N levels and locations. Higher NUE values were observed for late maturing potato cultivars at both N rates. Our study indicate that potato cultivars can be exploited for NUE improvement through improving and pyramiding of component traits at both low and high N levels.

James Hutton Limited: The link between molecular marker development and commercialisation

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James Hutton Limited (JHLtd) is one of the largest plant breeders in the UK and currently manages eleven commercial potato breeding programmes for both the fresh and processing markets, for both UK and internationally based customers. Our competitive advantage is gained through access to some of the largest and well-characterised germplasm collections in the world backed up by world leading scientific expertise in potatoes from the James Hutton Institute. JHLtd has recently established a new molecular diagnostics unit offering a range of diagnostic services for many crops to both in-house and external customers. In potatoes, disease resistance markers are routinely used to screen germplasm and advanced breeding lines (e.g. 57 R marker for *Globodera rostochiensis* resistance) and molecular fingerprinting services are offered to determine trueness to type or to resolve varietal identity issues. JHLtd works very closely with the scientists at the James Hutton Institute with an aim to provide the link from research to commercialisation of products. With potato cyst nematode (PCN) resistance high on the agenda in the UK, the James Hutton Institute and JHLtd are currently developing new markers for *Globodera pallida* resistance with an aim to improve parental /breeding lines through marker assisted selection (MAS) and to offer new and improved diagnostic markers.

POSTER PRESENTATIONS

2015 EAPR Breeding and Varietal Assessment
Section and EUCARPIA Section Potatoes

18TH JOINT MEETING

Vico Equense, Italy - November 15th – 18th, 2015

P1 Investigation of the potato virus Y status in seed potatoes in Romania (preliminary studies)

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The cultivar's selection is one of the most important critical component in the PVY control. Potato varieties with demonstrate some resistance to PVY can be managed effectively in most regions and maintain their certification status. However, knowledges regarding each cultivar's reaction to PVY and the various strains of PVY (especially the necrotic one) are criticals. This preliminary studies investigating the status of the Potato virus Y (PVY) during two years (2014 and 2015), in five main seed potato growing areas of Romania, revealed large differences in PVY incidence. The samples tested were from the following cultivars: Christian, Roclas (romanian cv.), Riviera, Carrera, Bellarosa, Jelly, Hermes and Red Lady. Serological investigations by DAS ELISA showed that 49.5% of the PVY positive samples in 2014 and 68.4% in 2015, viruses belonging to the PVYN group were found. A selection of 126 samples collected in 2014 and all 72 serological positive PVY^N samples collected in 2015 were further typified by molecular tests and by biological assays on tobacco and potato plants. The tests largely confirmed the predominance of the PVY^N group and within this group, the prevalence of recombinant PVY^{NTN}, with 79.4% and 88.6% in 2014 and 2015, respectively.

Keywords: potato virus Y, seed potato, necrotic strains.

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P2 Repression of catalase activity by Antisens and co-suppression strategies accelerated dormancy breakage and sprouting of potato tubers (*Solanum tuberosum* L.)

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Potato (*Solanum tuberosum* L.) is the fourth most important food crop in the world mainly due to its starch content and high quality protein, substantial amounts of essential vitamins, minerals and very low fat content. At harvest and for a finite period thereafter, potato tubers will not sprout and are both pre- and post-harvest conditions. Excessively long dormancy poses a problem in sprouting of seed tubers for early crop installation. Controlling the length of dormancy period could therefore be of considerable importance. In several vegetable species a relationship between the metabolism of active oxygen species and breakage of dormancy has been established. However, scarce information is available on the role of oxidative stress in potato tubers. The aim of the present investigation is to study the effect of catalase activity on the dormancy and the sprouting of potato, by generation of transgenic plants deficient in their CAT activity. Two constructs *pCat2AS* (*Cat2*, antisense orientation) and *pCatGH* (*GH2*, sense orientation) were mobilised into *Agrobacterium tumefaciens* (strain C58) and used to transform internodal explants of potato, cv. Désirée. The genes *Cat2* from *Nicotiana plumbaginifolia* and *GH2* from *Gossypium hirsutum*, both coding for the CAT2 isoform were used for suppression of catalase activity by antisense approach (2AS lines) or gene silencing by co-suppression approach (lines GH). The repression of CAT activity was associated with Ascorbate peroxidase (APX) accumulation and a decrease in the Hydrogen peroxide (H₂O₂) in potato tubers. The sprouting of transgenic tubers was accelerated as compared to the non-transformed control and was associated with an increase of APX activity.

P3 Efficacy of bio-fungicides for potato late blight control

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Late blight of potato, caused by *Phytophthora infestans* (Mont.) de Bary, is one of the most important diseases in all potato (*Solanum tuberosum* L.) growing areas of Tunisia.

Field experiments were conducted to evaluate the efficacy of two bio fungicides namely Fungastop and Cuivox, using seven varieties of potato (Loane, Bellini, Eden, Derby, Spunta, Royal, Oceania and Santana), against this disease using the foliage resistance in order to enlarge the list of fungicides in organic potato production. The foliage resistance was analyzed using an estimation of 1–9 scale values. Those observations are subsequently used to calculate the Area under the Disease Progress Curve (AUDPC). Results showed that repeated preventative application of fungicides is the primary means of control on susceptible crops, especially in the organic production, where the numbers of fungicides are limited. AUDPC permits to classify variety into 3 groups: Tolerant varieties (Derby, Eden and Bellini), with leaf destruction level between 1 and 8 %; Moderately susceptible varieties (Oceania and Santana Loane) with a percentage of leaf destruction ranged from 15 to 25%; Susceptible varieties (Spunta, Royal) with a percentage of leaf destruction varying from 30 to 70%.

Fungastop showed the same results with Cuivox for the Tolerant varieties, but in for the other cultivars, Cuivox seemed to be more efficient against potato light blight than the Fungastop.

Keywords: Potato light blight, Variety, Fungastop, Cuivox

P4 Screening of organic potato cultivars for resistance against late blight (*Phytophthora infestans*) in Tunisia

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During 2010, an assessment was conducted to determine the host resistance of organic potato cultivars against *Phytophthora infestans*, at the trial site of Chott-Mariem. Eight potato cultivars (Loane, Bellini, Eden, Spunta, Derby, Royal, Oceania, and Santana) were used for foliage and tuber evaluation symptoms. On foliage, the symptoms were analyzed using an estimation of 1–9 scale values. Those observations are subsequently used to calculate the Area under the Disease Progress Curve (AUDPC). Results showed that late blight could affect all the tested cultivars, but with a consistent differences between the AUDPC of cultivars. Spunta (the most used in Tunisia) and Royal seemed to be the most susceptible in leaves test. Those cultivars showed an early and fast starting of the disease.

The cultivars Eden, Derby and Bellini were the least susceptible and they revealed that they could be able to delay the beginning of the disease for twenty days. However, Oceania, Santana and Loane cultivars were moderately susceptible and they can delay the disease for one week. Our results demonstrated the for all tested cultivars, there is no correlation between the appearance of Late Blight symptoms on leases and those on tubers.

Keywords: *Phytophthora infestans*, resistance, cultivar, Potato, organic, Tunisia.

P5 The evaluation of homeostasis capacity on some potatoes landraces from West part of Romania

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On 31 potato landraces collected in hilly and mountain areas: Arad County (AR) and Alba (AB), Hunedoara (HD), Maramureș (MM) Counties respectively was assessed the ability to grow in *ex situ* conditions. The owners preserved them for their resistance to unfavorable condition and for their good resistance to viruses (41.66%). In comparison with their *in situ* evolution the Timișoara *ex situ* conditions are different and rather unfavorable. The high temperatures and drought are the main restrictions. The drought was kept under control using irrigation. During 15 years of conservation *ex situ/in vivo* the yield and other traits were visible diminished. To point out the homeostasis of potatoes landraces the genotypes were cultivated in different environments: very good Red Water / Târgu Secuiesc - Covasna County, good Măru - Caraș Severin County and unfavorable Cenad – Timiș County. The obtained data were compared with the performances emphasize in Timișoara. The obtained data were compared with the performances highlighted in Timișoara. The biometrical determination emphasized the most sensitive traits: tuber number, weight and yield g/nest and kg/ha (CV%=96.97). Only 18.2% of landraces performed an yield average over 10 t/ha. The Variance Analysis pointed out significant differences in genotype' homeostasis. In Timisoara if it was used irrigation the landraces behavior was good. The traits as yield/nest and kg/ha was higher than in Cenad and Măru (d= -290.78g000 and d= -108.52g000 respectively). In the most unfavorable conditions (2012) more than 75% of landraces collected in Hunedoara area emphasized high homeostasis; the yield/nest over-passed 300g. In the same time in Covasna County the best place for potatoes cultivation (year with extreme drought conditions) the landraces performances was significant low (d= -75.36g00) the g/nest average was less than 120g. The good homeostasis of some potatoes landraces gives the opportunity to be preserved in Timișoara *ex situ* conditions.

P6 Molecular and microstructural analysis of potato varieties

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Potato is one of the most common starchy food, with a popularity that is being increasing due to human needs expected by next few decades. Tuber crops contain 70–80% water, 16–24% starch and trace quantities (<4%) of proteins, lipids, minerals, etc. Starch is the major component of the dry matter, accounting for 70% of the total solids and exhibits differences in various properties in accordance with source and genotype. In this study we investigated the molecular diversity and the microstructural characteristics of several potato varieties, to understand their genetic variation and possibly detect its relationship with microstructure. Molecular analysis with SSR markers generated 186 polymorphic alleles from 23 loci, with an average of 8 alleles per locus. A core set of highly informative primers was identified (PIC values higher than 0.8). Scanning Electron Microscopy (SEM) analysis of selected varieties allowed the characterization of their potato tissue and raw potato tuber cells. Tuber parenchyma from all of them showed a reasonable preservation of the polygonal cell wall outline. The shape of parenchyma cellular compartments was genotype-dependent: elongated and hexagonal, or roughly spherical, more and less regular and defined. Also tuber parenchyma cells appeared to be genotype-specific, showing inclusions of starch granules with a range of different shapes and sizes. The genetic fingerprinting through SSR markers and the microstructural classification through SEM analysis provided useful information to discriminate a large set of potato genotypes.

P7 Functional characterization of Parallel Spindle Like (PSL) genes in potato

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In plants, whole-genome doubling largely contributed to plant evolution and diversification. Gametes with somatic chromosome number ($2n$) are widely recognized as the major route of plant polyploidization. The parallel arrangement of the spindles (ps) in the 2nd division of male meiosis is the most common mechanism generating $2n$ pollen through the formation of dyads and triads. In *Arabidopsis*, Parallel Spindle1 (*AtPS1*) was reported as implicated in the control of the ps meiotic defect. We identified three Parallel Spindle Like loci (*PSL1-3*) in the potato genome. To establish their putative role in the formation of $2n$ male gametes, two functional strategies were undertaken. Firstly, the *Arabidopsis* mutant *Atps1*, kindly given by Dr. R. Mercier (INRA, Versailles), was transformed with a construct containing the *PSL3* gene. The *PSL3* gene expression was detected by qRT-PCR in the *Atps1* transgenic lines. However, these lines still showed the mutant phenotype, i.e. dyads and triads, thereby indicating that the transgene, *PSL3*, failed to functionally complement the *Atps1* mutation.

In the other strategy, RNAi silencing of *PSLs* was performed in a wild diploid species (*Solanum commersonii*) and in a tetraploid potato (*S. tuberosum* cv Desirée). Hairpin-RNAi expression vector containing a 400 bp sequence common to all *PSLs* was utilized for transformation experiments by *Agrobacterium tumefaciens*. Several transgenic plants from both species have been obtained. The silencing of *PSL1* was confirmed in leaf by qRT-PCR. *PSLs* silencing in floral buds and pollen ploidy are under evaluation.

P8 Potato gene pool in the Czech Republic

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In the Czech Republic bank of potato (*Solanum* spp.) genetic resources works in the Potato Research Institute Havlíčkův Brod. The institute is in charge of this activity in the framework of the National Programme of Conservation and Utilization of Plant Genetic Resources and Agro-biodiversity and it is the only organization in the Czech Republic with the long-term engagement in this objective. The bank's mission is an assembly, evaluation, documentation, maintenance and delivery of potato genetic resource accessions. The collection involves 2.497 accessions maintained in an *in vitro* culture and divided into six sub-collections (*Solanum tuberosum* varieties, *Solanum tuberosum* tetraploid hybrids, dihaploids and diploids, cultivated *Solanum* species, wild *Solanum* species, interspecific *Solanum* hybrids). Assembled accessions are evaluated for 44 morphological and 12 agronomic traits and characteristics using a 9-point scale; and levels of resistances to 12 pests and diseases are determined. Derived information is a part of the genetic resource database maintained in the Czech Republic, which could be found on the website <http://www.genbank.cz/genetic/resources> and from June 2015 also on <https://grinczech.vuvr.cz/gringlobal/search.aspx>. The data are also accessible on the website <http://europotato.org>. The long-term maintenance is based on slow-growth culture and *in vitro* tuberization. Microtubers are induced using appropriate media and modified culture conditions. Chitting microtubers after dormancy period or surviving stem segments are subjected to the regeneration passage onto new media in 14-18 months. A revitalization and valorization program is running in the gene bank with the focus on virus infection eradication and screening of maintained material for presence of quarantine viruses, viroids and bacteria, complying with the EU directives.

Keywords: *Solanum* species, gene pool, gene bank, *in vitro*

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P9 Characteristics deciding on suitability for washing in potato varieties

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Based on results from five-year (2009-2013) field trials involving 47 potato varieties, an effect of variety and year was evaluated on characteristics of potato tubers intended for washing. Simultaneously, dependence between studied characteristics was calculated. Washed tubers were evaluated for skin smoothness, presence of black scurf and silver scurf, skin firmness, peeled tuber discoloration, greening, sprouting and presence of common scab.

The results show that silver scurf was explicitly the decisive parameter considering suitability for washing in individual varieties. A statistically significant variety effect besides the group of medium-late varieties was also recorded for skin smoothness (most varieties passed for this), greening (most varieties did not pass for this), sprouting and ambiguously for black scurf (only medium-early varieties) and common scab (very early and medium-early varieties).

The year effect was more pronounced compared to variety effect and reached significance in many cases also for indicators, for which the variety effect was not significant. Direct dependence between skin smoothness and silver scurf was detected ($r = 0,7152^{**}$ for the whole set). Since potato greening could be technologically eliminated (e.g. limiting the time between washing and sale, minimizing the exposition of washed tubers to light etc.) silver scurf infection is decisive for suitability for washing.

The results indicate that suitability for washing could be expected in Liliana, Valetta, Annabelle, Baccara and Marabel from the set of evaluated varieties. The suitability for washing decreases with increasing growing period duration, on the other hand, appropriate technological measures limiting occurrence of silver scurf, but also skin greening could have a positive impact on potato quality after washing.

Keywords: *Solanum tuberosum* varieties, suitability for washing

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P10 Software for genome-wide association studies in autopolyploids and its application to potato

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Genome-wide association studies (GWAS) are widely used in diploid species to discover causal variants in diversity and breeding populations, but GWAS software tailored to autopolyploids is lacking. The objectives of this research were to (1) develop an R package based on the Q+K mixed model for use with bi-allelic, co-dominant markers in autopolyploids, and (2) apply the software to a diversity panel of autotetraploid potato lines. A unique feature of the R package, called GWASpoly, is its ability to model different types of polyploid gene action, including additive, simplex dominant, and duplex dominant. Using a simulated tetraploid population, we confirmed our hypothesis that statistical power is higher when the assumed gene action in the GWAS model matches the gene action at unobserved QTL. Thirteen traits were analyzed in the SolCAP potato diversity panel, and, consistent with previous studies, highly significant QTL for tuber shape and eye depth co-localized on chromosome 10. For the other traits, only marginally significant QTL were detected, most likely due to insufficient statistical power: for simulated traits with a heritability of 0.3, the median genome-wide power was only 0.01. Our results indicate that both marker density and population size were limiting factors for GWAS with the SolCAP panel.

P11

Marker assisted organic breeding for late blight resistance

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Biolmpuls is a potato breeding program aiming at the selection of new varieties well adapted to organic farming. Therefore all selection stages are conducted under organic growing conditions. Resistance to late blight is a very important target in this breeding program. Therefore, at least 1 major late blight resistance (*Rpi*-) gene is present in one of the parents of every cross combination. True seeds are sown in trays filled with organic soil, and true seedlings are transplanted directly into the field at an organic farm. From selected seedlings a single tuber is harvested to be planted as a single hill first year clone. From selected first year clones six-hill plots are grown as seed potatoes in the second year, and from selected second year clones multiple tubers are used for seed potato production, yield and late blight trials in the next year. Each year the selection of true seedlings is postponed until natural late blight infection occurs, enabling the breeders to select in an early breeding stage for late blight resistant genotypes. In years of early natural late blight occurrence, selection for resistance can also be conducted in the first and second year clones. In 2015 the selected second and third year clones were screened with molecular markers developed to detect the presence of *R8*, *R9*, *Rpi-blb2*, *Rpi-chc1*, *Rpi-ber* and *Rpi-edn2*. Resistance of third year clones was also estimated on an artificially inoculated (IPO-C) late blight field trial. Results and comparisons between marker and field results will be shown.

P12

Integrated genomic approaches for investigating Solanaceae defence system

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Multiple omics approaches lead to new insights into plant-pathogen interactions, owing to the availability of high throughput biological data and computational tools able to extract important meanings. A Plant Resistance Genes database, (<http://www.prgdb.org>) was established in order to provide a comprehensive overview of plant resistance genes (R-genes). It is community database for plant scientists who could in turn contribute to this public resource through a WIKI-like system. The inferred cross-link between genomic and phenotypic information allows to access to a large body of information to find answers to several biological questions. Our database offers a range of querying and mining tools, including Solanaceae metaspecies section. As proof of concept, tomato, potato and pepper putative pathogen recognition genes were annotated with our specific predictor and characterized with respect to structural diversity, phylogenetic relationships and chromosomal distribution. A first genome-wide comparative analysis of candidate pathogen recognition genes in the Solanaceae was conducted underlying mechanisms of molecular adaptive selection at *Solanum* R loci. Analysis of main R-gene clusters allowed to reconstruct their evolution history.

P13 **InnoPlant, a combined research program between Inra and French seed potato industry: promoting potato breeding for sustainable resistance to pathogens**

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InnoPlant Joint Technological Unit was established in 2012 by the FN3PT / RD3PT (French Federation of seed potato growers) and INRA (French National Institute for Agricultural Research) as a tool to promote innovation and to increase the competitiveness of the French seed potato industry. The partnership is supported by the French Ministry of Agriculture. GNIS (French Interprofessional body for seeds) and ACVNPT (Association of potato breeders) are associated partners for some activities.

InnoPlant has a scientific program that combines research, development, dissemination and transfer on four major themes: health of seed potatoes, breeding for sustainable genetic resistance to different pathogens, integrated pathogens management and innovative strategies to increase the competitiveness of the sector.

This poster focuses on the breeding component of the program. In a context of reducing pesticide use, it is necessary to exploit resistant genetic resources and to combine them with efficient crop practices. To achieve this objective, the following actions are carried out:

Characterization of multi-resistant breeding lines i.e. that combine resistance to different pathogens including PVY virus, foliage late blight, *Globodera pallida* and *Meloidogyne incognita*

Assessment of resistance to emerging or newly identified pathogens. An example is to evaluate INRA breeding lines that are resistant to the bacterium *Pectobacterium atrosepticum* against the recently identified pectinolytic species belonging to *Pectobacterium* and *Dickeya* associated to potato blackleg and soft rot

Development of knowledge on the genetic structure and the phenotypic value of the genetic resources which are used in breeding programs, that include genetic material maintained in the INRA BrACySol Biological Resource Center (BRC) and improved breeding material maintained in collections held by French breeding companies

Identification of the best panel of SNP and best genotyping technology depending on the objective: to develop association mapping strategies or marker-assisted selection

Implementation of a long term cryopreservation method.

<http://www.umt-innoplant.fr>

P14 **Expression of key NUE genes at different nitrogen levels in five potato varieties**

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Doubling of agricultural food production worldwide over the past 40 years has been associated - beside other factors - with a 7-fold increase in the use of nitrogen fertilizers. Genotype - nitrogen level interactions reflecting to differences in responsiveness have been observed in several species in numerous studies. Developing N-efficient cultivars which take up N and/or utilize N efficiently provides an environmental-friendly strategy. Significant variation in nitrogen use efficiency (NUE) was reported among commercial potato cultivars. In this study we performed a NUE survey of potato cultivars by analyzing the expression of key NUE genes at different nitrogen levels. In the experiments four potato cultivars of our institute (White Lady, Katica, Hópehely, Chipke) and the S440 breeding line have been tested at three nitrate supply levels (in form of modified half strength standard Hoagland nutrient solution at 7.50, 3.00 and 0.75 mmol NO₃ concentrations) in a greenhouse pot experiment in 2013. We analyzed the expression patterns of nitrate reductase, nitrite reductase, ammonium transporter and asparagine synthase genes with RT-qPCR using leaf samples those have been collected at 0, 7, 14, 28 and 42 days after treatment. The complex pattern of the results will be shown in the presentation.

P15 Development of symptoms in diploid and tetraploid potato after inoculation with highly aggressive strain of *Dickeya solani*

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Pectinolytic bacteria *Dickeya solani* is spreading in potato crops in Europe. The highly aggressive strain of *D. solani*, IFB0099 (syn. IPO2276), has been used for inoculation of potato tubers and potato plants. The plant material consisted of cultivars and diploid interspecific hybrids of *Solanum*, originated beside *Solanum tuberosum* from wild and primitive cultivated potato species. The temperature 26° C was chosen, for testing tuber resistance, based on the results of tuber maceration of four cultivars, Irys, Glada, Gandawa, Sonda, in four different temperatures: 20° C, 23° C, 26° C and 30°C. The tuber resistance of 24 diploid clones did not differ significantly from the highly resistant standard of resistance to *Pectobacterium*, the somatic hybrid of *S. brevidens* (+) *S. tuberosum*, USA 249, and was significantly higher than the resistance of cvs Glada, and Irys, medium resistant and susceptible to *Pectobacterium* spp., respectively. The best cultivar in this experiment was cv. Mieszko, originated from the double backcross of the highly resistant to *Pectobacterium atrosepticum* diploid clone DG 88-9. The plant resistance was evaluated after inoculation with bacteria of the bases of stems of potato pot-plants grown in a greenhouse. Plants were covered with transparent foil after inoculation to provide high humidity. Due to high temperature in a greenhouse the symptoms of the stem base rotting were observed after few days post inoculation. In the repeated experiment the symptoms were observed mostly as leaf necroses and wilting than the rotting of stems. In general, potato cultivars showed symptoms of infection with bacteria *D. solani* on higher number of plants than the diploid clones. Thus the diploid hybrids might be used for improving potato resistance to bacteria *D. solani*.

P16 Use of the reference potato genome sequence to improve tetraploid genetic maps of chromosome IX

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The reference potato sequence is a useful resource to develop new markers in regions of interest. Using genetic mapping, we identified, a region of chromosome IX responsible for a significant variation of late blight resistance in two tetraploid mapping families. Our aim was to improve the accuracy of the maps. We used SolCAP SNP then developed SSR markers in the 7,1 Mbp region of the V4.03 reference genome previously determined using Blast of primers genetically mapped in a ~25 cM interval.

Three full-sib tetraploid families (150 or 280 individuals) were used to test then map the markers. 12/138 SolCAP SNP located in the region were chosen on the basis of their distribution along the pseudomolecule, position into gene related to resistance pathways and costs estimates. They were revealed using KASPar technology (LGC Genomics) 48 primers pairs were selected in 3434 unilocus primers pairs designed with Primer3 around the 5940 microsatellite motifs detected in the region using a local tool. The selection criteria were: 4bp repeat length, ~150kbp between the markers, molecular weight between 200 and 400bp. Before mapping the entire families, the informativeness of the primers was checked using both parents and 8 individuals of three full-sib families. PCR products were separated using an ABI PRISM® 3100 Genetic Analyzer.

7/12 SNP showed polymorphism in the 3 families. 4 SNP were polymorphic in only 2 families and the remaining SNP was polymorphic in only one family. Level of polymorphism varied from 2 to 5 clusters depending on the dosage and the parental configurations.

43/48 markers amplified 1 to 5 alleles. 43 to 50 alleles were polymorphic in each family, among them, 15 alleles were common to all. 13 markers were chosen on the basis of the origin of the polymorphism, the number of alleles revealed and their intensity.

Mapping on the entire families is underway.

P17 Understanding genetic control of tuber dormancy and sprouting in potato

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Potato tubers undergo a phase of dormancy after their maturation and their inherent dormancy phenotype is one of the key determinants of tuber postharvest life. Premature dormancy release (sprouting) in potato tuber during storage is accompanied by significant deterioration in quality. Use of chemical inhibitors for controlling sprouting (e.g. chlorpropham and maleic hydrazide) is under threat and tuber storage under temperatures lower than currently practiced is also discouraged because of consequential increase in energy use and adverse impacts on the environment. Besides the need to develop new storage strategies for potato, breeding potato cultivars with extended dormancy is desirable. Towards achieving this goal, we are dissecting the genetic components underlying tuber dormancy using a large diploid Phureja-Tuberosum mapping population (06H1) that exhibits significant genetic variation in tuber dormancy characteristics. The 06H1 population was genotyped using the Illumina Infinium potato 8k SNP array and a linkage map has been constructed. Field-grown tubers have been assessed for dormancy-release and sprout growth in storage at nine time-points over two consecutive growing seasons. Analysis of this data has revealed the presence of two major as well as other smaller effect QTLs on several potato chromosomes. Candidate genes located in the vicinity of these QTLs have been identified and work to develop transgenic lines has been initiated. Insights from this study, including some preliminary results from bulk segregant analysis using exome capture-based sequencing of low and high dormancy clone pools, will be presented.

P18 The co-expression of chitinase and RIP genes enhances resistance to *Rhizoctonia solani* in potato plants (*Solanum tuberosum* L.)

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Potato (*Solanum tuberosum* L.), one of the most important food crops in the world, is susceptible to many fungal pathogens including *Rhizoctonia solani* which is the causal organism of *Rhizoctonia* disease complex in potato, resulting in two different symptomologies of the disease, namely stems canker and black scurf. In the present study, the tetraploide potato cultivar Desirée was transformed via *Agrobacterium tumefaciens* strain LBA4404 containing the plasmid pBIN19 which harbor the Ribosome Inactivating Protein (*rip30* gene) and strain GV3101 containing the binary plasmid pGJ132 harboring both the chitinase gene (*chiA*) and *rip30* gene. The potato leaf disc was used as explants for transformation. PCR, Southern blot and Western blot were used for characterization of the transgenic plants. In this study it was shown that the regenerating callus, the developed shoots and the percentage of transgenic plants were influenced by the type of plasmid vector used for the transformation. Also, not all the plants developed in selective medium were positive for the corresponding gene using the PCR technique. Southern blot analysis confirmed that transgenic plants integrated 2-3 copies of *chiA* and *rip30* genes respectively into their genome. The expression of the CHIA and RIP30 proteins was confirmed in the leaf extracts of the transgenic clones by Western blot analysis. Greenhouse assay was carried out to evaluate resistance to the pathogen. Potato plants expressing *rip30* gene showed only partial resistance to *R. solani*. However, higher levels of resistance were obtained when *rip30* was associated to *chiA* genes. Transgenic potato plants generated by *Agrobacterium*-mediated transformation and co-expressing *Serratia marcescens* chitinase (*chiA*) and barley Ribosome Inactivating Protein (*rip30*) was much more effective in preventing development of the disease and inhibited fungal growth better than transgenic potato expressing either *chiA* or *rip30*, suggesting a synergistic effect.

P19 Scaling up more nutritious potatoes for food security and nutrition in Colombia

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National University of Colombia and McGill University worked with farmer communities in potato breeding and in laboratory, using genomics, metabolomics and nutritional analysis to develop new potato varieties. Three yellow potato varieties were selected, with enhanced resistance to late blight, improved yield and better nutritional quality, contain nearly 20% more iron and zinc than the most cultivated Colombian variety, *Criolla Colombia* and offer major health benefits. The potatoes have also high taste acceptability and results from a clinical study found 10.6% less iron deficiency in children who consumed the improved potatoes compared to those who consumed current commercial varieties. A scaling up plan of new varieties is based on education as a sustainable change principle, oriented to build farmers' autonomy. Training Schools for Family Agriculture combined with Rural Entrepreneurial Nuclei (REN) and deepened food security and nutrition (FSN) interventions through home and school gardens will lead to increased income and improved health and nutrition. The model for the adoption of new varieties includes production of good quality seed, good agricultural practices, entrepreneurial training for marketing and stimuli to consumption. The scaling up the new potato varieties looks for achieving objectives on FSN: increased yield family income, availability of more diverse and nutritious foods, increased micronutrients in the diet, and nutritional and sanitary education. Besides, there will be training for dialogue among authorities and communities, and promotion of women roles in the family and community decision-making. Farmer groups organized in REN will use existing channels to access markets. The potatoes are expected to have high marketability acceptance and their sale and consumption will not require changes in dietary patterns or consumers' behavior and culture. This project is funded by International Development Research Centre and Department of Foreign Affairs, Trade and Development from Canada through the Canadian International Food Security Research Fund.

P20 Identification of interactions between *Phytophthora infestans* RxLR effectors and *Solanum tuberosum* host proteins by yeast two-hybrid approach

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Potato (*Solanum tuberosum* L.) is the third most important food crop just after wheat and rice and as each crop it is challenged by various pathogens. Lacking an adaptive immune system, plants including potato have developed a monitoring system to detect and divert pathogen invasions. Defense responses can be initiated on the host cell surface by so called pattern recognition receptors which detect particular conserved molecular signatures of pathogens or apoplastic effector proteins. In turn, pathogens deliver other effector proteins that operate within host cells and interfere with pattern recognition receptor immunity in order to promote disease progression. Effectors can be recognized by plant disease resistance (R) proteins but little is known about the identities of the host proteins that are targeted by oomycete effectors and even less is known about the roles those targets play in plant immunity. One of the most devastating potato diseases is the late blight caused by the oomycete *Phytophthora infestans*. According to genome information, the pathogen secretes more than 500 RxLR effectors that are translocated inside the host cells.

The goal of our study is to gain a better insight into the function of *P. infestans* RxLR proteins. For that purpose, yeast 2-hybrid (Y2H) approach was applied to identify candidate host targets of *P. infestans* RxLR effectors. Y2H cDNA library from potato infected with the pathogen was constructed and screened with candidate RxLR molecules. Our experiments aim to learn more about the regulation of expression and the delivery system of RxLR-effectors and to identify their molecular targets in the host.

In conclusion, a more in depth knowledge of RxLR biology is central for understanding and ultimately controlling diseases caused by *Phytophthora*.

P21 Bulk segregant QTL analysis of chipping quality in elite potato cultivars

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In recent years genetic markers has been developed and utilized to optimize crops in terms of yield and reducing the use of pesticides and fungicides in the production, storage and processing of the crops. The potato has not kept pace with this development, despite the status as the world's fourth most important crop. The scientific community has suggested genetic markers, but few have been embraced by the breeders, partly because only a few have been tested in an industrially relevant population.

After the publication of the Potato Genome (Spud DB Genome Browser, <http://potato.plantbiology.msu.edu/cgi-bin/gbrowse/potato/>) a possibility has emerged to search for candidate genes under and near QTL regions. Combined with the Bulk Segregant Analysis, to find novel and known QTL's for specific traits, and ultra high-throughput Next Generation Sequencing, to find mutations specific for the bulks, this opens up for the discovery of new genetic markers for the industry.

We designed a large industrially relevant population based on 18 elite cultivars used by the Danish breeding company LKF Vandel and utilized the three methods mentioned above. The population was phenotyped for chipping quality after storage at 6 degrees Celsius, and two bulks of 96 phenotypically extreme performers were defined and sequenced with Next generation Sequencing. We then analyzed the resulting data for regions of the genome with greater density of mutations, which was different between the two bulks. These regions are putative QTL regions with association to chipping quality. In at least one case, genes were found in these regions, with properties that have previously been found to associate with chipping quality.

P22 Research on the potato resistance to various pathotypes of *Synchytrium endobioticum*

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Synchytrium endobioticum (Schilberszky) Percival, the causal agent of potato wart disease is the most important quarantine pathogen infecting potato. The widespread pathotype of the pathogen is 1 (D1), but in Europe about 40 virulent pathotypes were also identified. The phenomenon of resistance to virulent pathotypes of *S. endobioticum* is poorly understood and number of cultivars resistant to such pathotypes is limited. In the Plant Breeding and Acclimatization Institute – National Research Institute studies on potato resistance to *S. endobioticum* have been realized for several years, and recently a highly effective source of resistance was identified in diploid interspecific hybrids. This source of resistance is of a special importance, since it ensures high resistance to broad spectrum of *S. endobioticum* pathotypes. The aim of presented study was to determine the inheritance of resistance against seven virulent 2(G1), 2(Ch1), 3(M1), 6(O1), 8(F1), 18(T1), 39(P1) and a common pathotype 1(D1) of *S. endobioticum* in this source of resistance. The donor of resistance was diploid potato clone DG 97-264, extremely resistant (score 1) to the all above mentioned pathotypes of *S. endobioticum*. This clone was crossed with a diploid clone DG 97-1805 (resistant to 1(D1) but susceptible to seven virulent pathotypes) and F1 progeny (SEN 12-01) was obtained. Among 180 tested F1 progeny clones 81 were resistant to all eight examined pathotypes of the wart pathogen. The remained 99 clones, in vast majority, were susceptible to seven virulent pathotypes. So far 56 of these clones have been tested also for resistance to 1(D1), and 24 of them were resistant and 32 susceptible to this pathotype. Such observations suggest that this newly identified source of resistance provide simultaneous protection against one common and at least seven virulent pathotypes of *S. endobioticum*, and is inherited as a quantitative trait. Research is in progress.

P23 The yield of different potato varieties under the conditions of Western Serbia

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The study presents results of a two-year experiment of yields of different potato varieties in Western Serbia: Cleopatra, Anushka and Presto (early), Kuroda, Omega and Dita (medium-early) and Desiree, Roko and Jelly (medium-late). The research was conducted during 2008 and 2009. The four-replicate field trials were set up using standard methodology according to the random block design.

The analysis of variance suggest that number above ground stems per plant, number of tubers per plant and total tuber yield were significantly fluctuating depending on genotype (G) and year (Y).

In the two-year period average, the highest total yield was recorded in the variety Desiree (28.30 t ha⁻¹), followed by Anushka variety (26.60 t ha⁻¹), while the lowest average yield recorded was in the variety Omega (13.35 t ha⁻¹).

Obtained results show that the highest yields under western Serbia were recorded in early varieties (Cleopatra, Anushka and Presto) and medium late varieties (Desiree) that expressed good resistance to high air temperatures and stress caused by drought.

Keywords: Potato, variety, tuber, yield

P24 An integrated genomics and effectoromics approach for potato wart disease resistance breeding

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Long persistent spores and lack of chemical control make the potato wart disease, caused by the obligate fungus *Synchytrium endobioticum*, an important quarantine disease of cultivated potato (*Solanum tuberosum* L.). The cultivation of varieties resistant to a wider spectrum of pathotypes may be decisive to reduce the propagation of the disease. However, there is a lack of information about where various *R* genes are located in the genome and what pathotypes they recognise. An integrated approach of effector genomics, QTL mapping and genome wide association studies (GWAS) will be followed to tag the loci involved in race specific wart disease resistance in potato. An identification of resistance QTLs for multiple pathotypes will be carried out in different F1 populations. Complementary to these F1 population studies, a set of 200 different potato varieties was added to perform the GWAS. Fine mapping of the genomic regions highlighted in QTL mapping and GWAS will be pursued. In this respect, the work will greatly benefit from the progress made in a parallel study in which *S. endobioticum* Avr genes will be identified. An assay based on recognition of the Avr protein will provide a highly simplified phenotyping method. The results will be used to develop molecular genetic tools or SNP assays for marker-assisted-selection as well as for the screening of potato cultivars and accessions to identify resistance sources. Moreover, it will contribute to understand the inheritance of wart disease resistance, and to understand the interactions and coevolution between the pathogen and its host.

P25 Estimation of genetic parameters for tuber dormancy in populations of diploid potato

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Considered to be greatly important and challenging for potato agronomy, features associated to tuber dormancy have been object of extensive quantitative genetic analyses. The aim of this study was to estimate tuber dormancy period heritability and determine its genetic advance by the methods of selection between families, selection between and within families, and stratified and simple mass selection, in a full-sib diploid population. Tuber dormancy was measured as days elapsed since harvest until the moment the tuber started sprouting. Showing statistically significant differences among families, this parameter ranged from 15,9 days for the control Criolla Colombia to 74,7 days for family 09-004. The heritability value for the different methods of selection ranged from 0,87 to 0,93, showing a strong genetic additive effect. The population showed a marked bias towards short dormant periods, confirming that the Phureja group contributes dominant genes associated to the lack of tuber dormancy, with a well-marked effect on the F1 generation. Using a selection ratio of 10%, the expected reduction in the dormant period is 17,6; 18,2 and 25 for individual, kin and mass selection, respectively. This is a fairly interesting possibility, as far as using the Phureja group in potato breeding is likely to significantly reduce the tuber dormancy period at the diploid and tetraploid levels.

P26 Potato breeding and plant biotechnology in Estonian Crop Research Institute

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Potato Breeding has been performed at the Estonian Crop Research Institute since 1920. Forty-one varieties have been released since. Seven of them are currently in EU variety list; others are stored *in vitro* in the gene bank. The main goals of potato breeding in Estonia are: disease resistance medium and medium late high yielding external tuber and culinary quality table potato varieties, suitable for organic farming.

The potato is crossed in greenhouses in 50 to 80 variations per year, which yield 25,000 to 30,000 seedlings per year. After 5-year selection the cardinal test contains 15-18 perspective breeding lines (10-15 genotypes), among which the new varieties are chosen if suitable.

The cooperation between potato breeders and the Division of Plant Biotechnology has lasted for 30 years. The varieties used in cross-fertilisation, perspective breeds and the initial material for seed growing are through virus-eradication procedure using the technology including the meristem culture and are preserved *in vitro*. In cooperation the somaclonal variations evolved in meristem culture have been researched. A good quality initial material can be obtained and it can be preserved long term *in vitro* and multiplied for seed production or breeding.

Our experience has shown that the variability of meristem clones can serve as a valuable tool in improving the yield and other properties of potato varieties and breeding lines. We have reason to presume that such kind of deviation was affected by longer thermotherapy period and higher concentration of growth hormone in medium (Rosenberg et al 2010).

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P27 Susceptibility of the potato cultivars to a second growth of the progeny tubers consecutive to severe hot and dry climatic conditions

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Potato is susceptible to various environmental factors during the phase of progeny tubers growth. A long period (>20 days) of drought and heat (>30°C), results in a slowdown of the growth of the progeny tubers. After rain return and the decrease of the temperature, the growth of the progeny tubers can restart. For some varieties, the growth of the tubers does not restart normally and physiological disorders appears usually named as “second growth” of the progeny tubers. The progeny tubers can produce outgrowth leading to the development of misshaping tubers (pointed, knobby and dumbbell tubers). The progeny tubers can also start sprouting and produce a second generation of progeny tubers (tuber strings). Such disorders will have a negative impact of tubers quality. The outgrowth of the progeny tubers are feeding from the tuber and transforming starch in sugar. As a result, the progeny tubers can become glassy or with a poor or irregular dry mater content and a high rate of reducing sugars. The commercial consequence is that those tubers are not appropriate for the fresh market due to the deformities and also not adequate for processing due to the high sugar content responsible for blackening after frying. The growing season 2015 in Switzerland was characterized by 1.5 month of dry and very hot conditions (no rain and maximum of temperature close to 40°C), favorable for the development of sprouting and misshapen tubers. It was decided to take advantage of this extreme climatic event to evaluate the susceptibility of 47 cultivars to these physiological disorders. It appears that the cultivars Bintje, Agria, Victoria, Ditta, Annabelle and Erika are very susceptible to regrowth and that the cultivars Agata, Alexandra, Antonia, Celtiane, Esmeralda, Gourmandine, Gwenne, Challenger, Lady Rosetta and Osira are tolerant to extreme hot and dry growing conditions.

P28 New ware potatoes with combined resistances to nematodes (*Globodera pallida* Pa3, *Globodera rostochiensis* Ro1-5) and PVY developed by breeding with protoplast fusion and marker-assisted selection

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Modern potato breeding combines conventional breeding methods and biotechnological strategies. Especially molecular marker techniques facilitate the selection of valuable progenies at an early stage. One of our most important targets is the development of resistant and adapted plants. For this purpose, several genetic markers linked to important breeding traits like resistances to cyst nematodes (HC: *Globodera pallida* Pa3, Sattarzadeh et al. 2006; Gro1: *Globodera rostochiensis* Ro1-5, Paal et al. 2004), *Phytophthora infestans* (Rpi-bulb 1: Fadina et al., 2013; Rpi-bulb 3: Zhu 2013, Rpi-sto1: Zhu et al. 2012) and extreme resistance to Potato Virus Y (YES3: Song & Schwarzfischer, 2008) are available. We apply them in our breeding program in order to create multiresistant cultivars. In particular, also somatic hybrids with already accumulated resistance genes generated by electrofusion of protoplasts were used as breeding parents. Around 25.000 seedlings from over 200 crossings were grown in sterile culture and analysed by a multiplex PCR-System. Only seedlings with combined resistances were propagated *in vitro* and planted in the greenhouse. Resistances to nematodes were proved by biotest. In the poster, we will summarize all data from the marker analysis as well as the further selection process. With this strategy we developed numerous ware potato clones with combined resistances to Pa3, Ro1-5 and PVY in one plant. Until now, no cultivar with the combination of these three resistances exists in this section. The additional introgression of Rpi genes is in progress.

P29 Cytoplasmic genetic diversity in potato

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Different plastid DNA markers have been used to investigate genetic diversity within cultivated and wild potato species. Correlation of distinct cytoplasmic genomes with complex agronomic characters was recently demonstrated (Sanetomo, Gebhardt, 2015). Using five PCR-based markers six cytoplasm types (A, D, M, P, T, W) have been identified in potatoes; three of them (D, T, W/-) are associated with male sterility (Hosaka, Sanetomo, 2012, 2014).

Using set of PCR-based markers of Hosaka, Sanetomo (2012) we performed a large scale screening of 463 accessions of 30 South American species and 89 accessions of 14 Mexican species to identify their cytoplasm types. Five cytoplasm types (A, M, P, T, W) have been identified in South American species and two (D, W) – in Mexican species.

Of 187 South American cultivated species (aboriginal varieties) 64% possessed by the P cytoplasm type (mostly Andigenum Group diploids), 10% – by the A type (mostly Andigenum Group tetra- and triploids), 4% - by the M type (*S. juzepczukii*, *S. ajanhuiri*, few accessions of Andigenum Group), 4% - by the W type (landraces of different ploidy levels) and 18% had the T type (mostly Chilotanum Group). These data were compared with our results of cpSSR analysis (Gavrilenko et al. 2013). The cytoplasm types A and T match well to the cpSSR haplotypes II and III, correspondingly. The type P corresponds to cpSSR haplotypes I and VIII. High polymorphic cpSSRs differentiated the M and W types into many different haplotypes.

The most frequent cytoplasm type in Russian breeding varieties is the T type (72%), the W and the D types were detected in 17% and 16% correspondingly. Based on the cpSSR analysis the W type of varieties was divided into several haplotypes. Our future plans directed on continuation of cytoplasmic diversity research in breeding varieties to study their maternal lineage.

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P30 Evaluation of potato genotype yield and starch content stability

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One of the aims in potato breeding programme is to develop varieties with high yield potential; essential for successful variety introduction into production additionally is yield stability in changing growing conditions, especially in organic farming system. As well as evaluation quality traits stability (starch content) become a relevant task. The aim of research is to evaluate breeding clones for trait stability for selection purposes. Dynamic concepts were used for evaluation yield stability: regression coefficient (b1 and sd2), ecovalence (E) and nonparametric method – yield value ranging. The b1 was applied also for starch stability assessment as well genotype environmental variance (s2) from ANOVA as static concept. The trial was carried out in two regions of Latvia using fields of integrated and organic farming systems during 2013 - 2014, totally six growing environments. Yield of eleven potato genotypes were evaluated, including set of standard varieties. Regression coefficient characterise that mostly genotypes tuber yield was flexible depending on growing environment. For one genotype b1 > 1 significantly, so this genotype yield reflected environment acceptance. Comparatively high E was observed for high yielding genotypes, so yield stability using this parameter was detected as low. Ranging of varieties for yield value in each environment did not characterize traits stability, but could be used for detection of genotypes suitability to most appropriate environment. Environmental variance comparatively characterise starch content stability of potato genotypes. The genotypes starch content stability evaluation using regression (b1 and sd2) differed comparing with evaluation using environmental variance (s2).

Keywords: potato genotype, trait stability, yield, starch content

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P31

Assessment of late blight resistance: two methods

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Improving potato resistance to *Phytophthora infestans* and new varieties release to the Belgian potato catalogue are the aims of the potato breeding program established at the CRA-W (Belgium).

Resistance of the clones to foliage blight is assessed following two methods. On one hand, once a sufficient quantity of seed potatoes is obtained, field trial takes place. The objective of the trial is to evaluate the plant material resistance in natural conditions. Clones are compared to control varieties without fungicide application. The disease progression is measured following a scale corresponding to the percentage of foliage affected by the disease (Eucablight scale). Disease score starts when the first symptoms of late blight appears, it is carried out every 3 to 4 days and it stops once the susceptible genotypes score 100%. The rAUDPC (relative Area Under Disease Progress Curve) value is calculated and a resistance score on a scale from 1 to 9 is given to each clone.

On the other hand, since 2014, detached leaf tests are applied to assess foliage blight resistance. Three strains of *Phytophthora infestans* genotyped by SSR markers have been chosen for the trials. The choice of the strains is based on genotypic analysis on strains collected in Wallonia during 2013 (Euroblight project). The selected strains are: Blue-13 (13_A2), Pink-6 (6_A1) and 1_A1. So far, three tests have been carried out: the first one on varieties used as genitors in our breeding program, the second one on the progeny of our crossings at an early stage (seedlings from the true potato seeds) and the third one on plantlets from wild *Solanum* species showing at least one *RPI* gene.

P32

Identification of potato varieties with a panel of SNP markers

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Rapid technological developments in DNA marker techniques will sooner or later cause a shift from PCR fragment analysis into SNP marker analysis. At this moment the identification of potato varieties is achieved with nine SSR markers (STM0019, STM2005, STM2028, STM3009, STM3012, STM3023, STM5136, STM5148 and SSR1) which across all loci display the absence or presence of 95 SSR alleles. We have analyzed 539 varieties with a 20K SNP array which resulted in 14530 SNPs. This dataset was used to deduce the minimal number of SNP markers to allow a robust identification of potato varieties. This resolving power is highly influenced by the population allele frequency of the SNPs and the method of data capture, where the assay method would generate absence / presence values only, or also allele dosage estimates.

Besides of the ability to identify cultivars with existing SNPs it is also conceived that it should be possible to substitute each SSR allele with a SNP marker that specifies the same haplotype. This would require a haplotype specific SNP allele (hs-SNP) in complete linkage disequilibrium for each SSR allele. A pilot using SSR1, which resides in the promotor region of the *GBSSI* gene (PGSC0003DMG400012111), shows that SNPs may even improve the current resolution. It appeared here that SSR alleles with equal fragment length are based on more than one haplotype. It was observed that every haplotype specified by a specific SSR length also has hs-SNPs. The utility of SNP panels for cultivar identification is not limited to issues with protected varieties with breeders' rights, but allows the sector to validate the identity of their working collection on an annual basis at reasonable costs.

P33 Taming the wild potato: cytogenetic and genomic characterization of the *Solanum commersonii* germplasm for introgressive hybridization breeding

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Solanum commersonii (2x=24, 1EBN), a wild species from Uruguay, is a valuable source of germplasm for potato breeding because of its resistance to bacterial wilt (*Ralstonia solanacearum*) and frost tolerance. As a first stage in pre-breeding we prospected the genetic variability available in natural populations. To circumvent postzygotic incompatibility we exploited the production of unreduced gametes and elucidated their formation mechanisms. We also studied pairing and structural variation between chromosomes from *S. commersonii* and their homoeologs from *S. tuberosum*. Genetic variation in natural populations was assessed through microsatellite markers. A detailed description of meiosis was made to evaluate unreduced gamete production. We studied homoeologous pairing behaviour at diakinesis of 3x hybrids (*S. commersonii* x *S. tuberosum* Group Phureja). Fluorescent *in situ* Hybridization (FISH) using chromosome-specific BAC probes was applied to pachytene complements of *S. commersonii* to analyze collinearity with *S. tuberosum*. Genetic variation and variation for resistance was observed among genotypes, allowing selection of genotypes to start the introgression schemes. Unreduced pollen with high viability was found in natural populations and there was correlation between parallel spindles and triad formation, from which two unreduced gametes are formed in a mechanism genetically equivalent to first division restitution (FDR). Triploid hybrids showed nearly-autotriploid meiotic behaviour, forming up to 12 trivalents. The position of BAC signals indicates high colinearity between the two genomes but loop formation in pachytene trivalents revealed small-scale rearrangements. Our results point to *S. commersonii* as an outstanding candidate for introgressive hybridization. *Solanum commersonii* possesses diversity to broaden the genetic base of potato. It displays variability among accessions in resistance and has spontaneous production of viable unreduced gametes by FDR, thus retaining maximum heterozygosity. Lack of genome differentiation facilitates homeologous pairing and recombination, but small-scale rearrangements may prevent introgression of specific chromosome regions. Evaluation of microsynteny will help anticipate potential drawbacks like linkage drag.

P34 Combined use of COSII-associated SNPs and HRM to assess chromosome dosage in potato F1 hybrids

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An important potato resource for potato breeding is represented by 2x1EBN *S. commersonii*. A breeding scheme based on the production of triploid bridges was developed to introgress its genes into the cultivated gene pool. In this research, triploids obtained from this breeding scheme were used to assess a chromosome dosage PCR-based approach. The method we propose estimate chromosome parental origin through a) the identification of SNPs among single-copy conserved orthologs sequences (COSII markers), and b) the application of HRM to rapidly and quantitatively differentiate each chromosome. Screening of 45 COSII marker sequences allowed the identification of *S. commersonii* specific SNPs for all 12 chromosomes. Combining HRM with the establishment of synthetic DNA hybrids, SNP markers were successfully used to predict the expected parental chromosome ratio of five interspecific triploid hybrids. The HRM profile similarity between the reference (synthetic DNA mixtures) and the triploid hybrids was based on a percentage of similarity score, called Genotype Confidence Percentages (GCPs). A GCP value >90 was detected for all the comparisons between the triploid HRM profile and the *S. commersonii* : *S. tuberosum* 2:1 reference sample. This indicated that for all 12-chromosome triplets the triploid hybrids harbor two *S. commersonii* homologues and one *S. tuberosum* homeologous, as expected. These results demonstrated the ability of this strategy to distinguish from each other diverged genomes, and to estimate chromosome dosage.

P35

Advances in breeding for resistance to potato bacterial wilt (*Ralstonia solanacearum*)

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Potato bacterial wilt (BW) caused by *Ralstonia solanacearum* is a very damaging disease worldwide. Some cultivated (Phureja) and wild species sources of resistance, have been identified earlier. The instability of resistance, pathogen latency in tubers and/or limited adaptation to specific growing regions has prevented adoption of resistant varieties. At The Sainsbury Laboratory (UK), one EFR receptor (a PRR from *Arabidopsis thaliana*) providing a broad plant defense response to bacterias has been cloned. Accessions of *Solanum commersonii* and *Solanum chacoense*, wild potato species collected in Uruguay, have shown high resistance level to BW, quantitatively inherited. Disease resistance screening is regularly performed under controlled experimental conditions, using an aggressive pathogen strain and root damaging. Disease latency evaluation in asymptomatic plants is performed through high sensitivity PCR test. Introgression of this resistance to cultivated potatoes has been achieved through sexual polyploidization (2n gametes). In contrast to *S. chacoense*, *S. commersonii* requires the utilization of Phureja group as a bridge species for initial crossings. Several back-crosses to Tuberosum have been obtained after each generation of selection. At BC2, several 4x genotypes developed at CIP for BW resistance were included in crossings. During F1 and BC1 generations, only selection for resistance is applied. Thereafter, BC2 and further progenies are field planted and selected for agronomic traits before disease resistance screening. Large differences in combining ability for yield and selectable traits were observed. Improved germplasm has been developed with high levels of BW resistance up to BC3. Furthermore, 23 transgenic events of the susceptible cultivar 'INIA Iporá'-EFR and 12 events of the BC2 resistant clone 09509.6-EFR have been developed at The Sainsbury Lab. Preliminary results show that several of this BC2 transformed genotypes achieved the highest level of resistance and reduced latency. Complementing various sources might prove very useful for developing valuable resistant germplasm widely adaptable.

P36

Impact of peat elixir and vermikompost extract on the yield and quality of potato tubers in conventional cultivation system

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Modern agriculture may be characterized by the narrow specialization of farms, therefore many farms have no animal-origin organic fertilizers available. Researches about the use of organic products in agriculture to restrict the use of pesticides and artificial fertilizers have been performed for a long time. Nowadays different organic preparations are produced that are acquired as a result of complex processes, ensuring ready-to-use biologically active substances in them and also activating their properties. One of the purposes of the research was to evaluate the impact of extracts from organic products on the yield of potato tubers (*Solanum tuberosum*) in the conventional cultivation system. A field experiment using the potato cultivars 'Borodyansky Rozovij' (early) and 'Lenora' (medium-early) was arranged in the State Stende Cereals Breeding Institute in 2011, 2012 and 2013. Peat elixir and vermikompost extract obtained at different temperatures: +45 °C and +95 °C, as well as a mixture of these extracts were used for treatment of tubers and plants. In total for every cultivar, the research included 24 treatments, and in addition a control variant (without treatment) and a variant using a standard potato cultivation technology. Tubers were treated immediately before planting, but plants were treated three times during the vegetation period. Potato cultivars 'Borodyansky Rozovij' and 'Lenora' differently reacted on extracts from organic products and treatment variants. Average three-years research results proved that the use of organic product extracts significantly ($p < 0.05$) affected the yield of tubers in different treatment versions for both cultivars – 'Borodyansky Rozovij' and 'Lenora'. The starch content in tubers has changed as the result of application of organic product extracts in all treatments, but the variation was not significant ($p > 0.05$) if compared to control. Nitrate content in tubers did not exceed the permissible limit in any variant. It was found that nitrate content in tubers tended to decrease in variants, where the extracts was applied three times during the growing season.

Keywords: potatoes, peat elixir, vermikompost extract, nitrates, starch

P37 Leaf transcriptional profiling of two potato varieties exhibiting high anthocyanin content

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Anthocyanins are bioactive compounds that can significantly affect leaf response to environmental stresses. The presence of high levels of anthocyanins in tissues exposed to stress conditions could be an important advantage for plant resistance to different pathogens and insects. In potato, studies mainly focused on the identification of genes regulating phenylpropanoid production in tuber tissues. Conversely, the molecular mechanisms related to anthocyanin production and accumulation in other vegetative tissues still remain unclear. In light of this, the aim of our research was to identify the main factors affecting the activation of anthocyanin pathway in leaves. This was pursued through the transcriptional profiling of two varieties ("Double Fun" and "Blue Star") exhibiting a different pigmentation in leaves. RNA was extracted from three biological replicates of fully expanded third and fourth leaves at appearance of first flowering buds and was sequenced by a NGS paired-end strategy at the Genomicx4life. After reads processing, quality check and all the routinely bioinformatic analyses, 15,820 loci were retained for further investigations. Overall, 5,679 DE genes were identified between the two varieties, 2,659 up-regulated and 3,020 down-regulated. Enriched GO terms were identified focusing the attention on those with a FDR value ≤ 0.05 and a higher enrichment score. In the comparison "Double Fun" vs. "Blue Star", 14 down-regulated DE genes were assigned to GO term regulation of anthocyanin metabolic process (GO:0031537). The involvement of these genes, as well as of others related to the flavonoid biosynthetic pathway, is being undertaken to clarify the complex mechanisms regulating anthocyanin biosynthesis and accumulation in potato leaves.

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