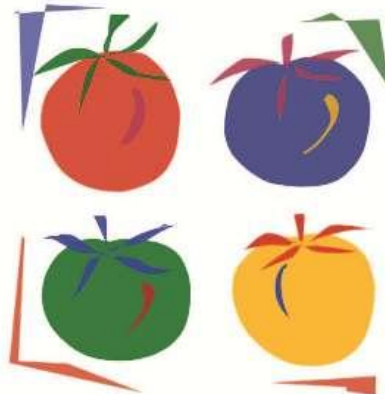


EUCARPIA TOMATO



Avignon 2014

BOOK OF ABSTRACTS

XVIIITH EUCARPIA MEETING, VEGETABLE SECTION, TOMATO WORKING GROUP

22ND – 25TH APRIL 2014

AVIGNON

FRANCE



SCIENTIFIC AND SOCIAL PROGRAMME

TUESDAY 22ND APRIL

- 14.00 – 17.00 Registration and Poster set up at the **Université d'Avignon (rue Louis Pasteur, 'J4' on the map)**
- 18.00 – 20.00 Welcome Reception at the Avignon **Town Hall (Place de l'Horloge, 'G4' on the map)**.

WEDNESDAY 23RD APRIL

- 08.30 – 09.15 Registration and Poster set up at the **Université d'Avignon**
- 09.15 – 09.30 Opening

SESSION I : MANAGEMENT, DIVERSITY AND CHARACTERIZATION OF GENETIC RESOURCES

Chairperson : José Blanca (COMAV Valencia, Spain), Christopher Sauvage (INRA GAFL, France)

- 09.30 – 10:00 **José Blanca** : Tomato variation, origin and domestication
- 10.00 – 10.20 **Jaime Prohens** : Conventional and phenomic tools for typification and enhancement of local tomato varieties
- 10.20 – 10.40 **Irina Balashova** : The heritability analysis of main tomato traits for the special hydroponic technology (on narrow benches)
- 10.40 – 11.20 *Coffee Break*
- 11.20 – 11.40 **Christopher Sauvage** : The study of domestication reveals a loss of diversity, signature of selection and transcriptional modifications in tomato
- 11.40 – 12:00 **Niels Müller** : Natural variation in circadian rhythms suggests an effect of domestication on the circadian clock of tomato
- 12.00 – 14.00 *Lunch at Université d'Avignon et des Pays de Vaucluse (UAPV)*

SESSION II : GENETIC BASIS FOR SELECTION AND ADAPTATION TO ABIOTIC STRESS

Chairperson : Francisco Perez-Alfocea (CEBA-CSIC, Murcia, Spain), Rebecca Stevens (INRA GAFL, France)

- 14.00 – 14.30 **Francisco Perez-Alfocea** : Rootstock-mediated alleviation of abiotic stress in tomato: what's beyond vigour?
- 14.30 – 14.50 **Eliezer Lifschitz** : The making of a sympodial plant: the role of florigen as a regulator and the great communicator of the shoot system in tomato
- 14.50 – 15.10 **Dina Saint-Clair** : Genetic and genomic dissection of traits associated with water stress tolerance in near-isogenic lines derived from wild tomato
- 15.10 – 15.30 **Vincent Truffault** : Metabolic profiling of transgenic lines silenced for an MDHAR gene and impact on ascorbate redox state and carbon metabolism
- 15.30 – 15.50 **José Jimenez-Gomez** : Analysis of natural variation in canalization at the transcriptional level
- 15.50 – 17.00 **POSTER SESSION and drinks**
- 17.00 – 18.00 **Lukas Mueller** : The SOL Genomics Network Workshop
- 18.00 - ... *Free Evening*

THURSDAY 24TH APRIL

SESSION III : GENETIC BASIS AND SELECTION FOR RESISTANCE TO DISEASES

Chairperson : Laura Rose (Heinrich Heine Univ., Düsseldorf, Germany), Jean-Luc Gallois (INRA GAFL, France)

- 08.30 – 09.00 **Laura Rose** : Evolutionary genetics of disease resistance in wild tomatoes

- 09.00 – 09.20 **Mara R. Ercolano** : Genomic approach for identifying and selecting disease resistance candidate genes in tomato
- 09.20 – 09.40 **Yuling Bai** : Fine mapping of the tomato yellow leaf curl virus resistance gene Ty-2 on chromosome 11 of tomato
- 09.40 – 10.00 **Liliana Stamova** : Resistance to fusarium crown and root rot
- 10.00 – 10.30 *Coffee Break*
- 10.30 – 10.50 **Sophie Rommel** : Resistance-shaping mirnas in wild tomatoes
- 10.50 – 11.10 **Camille Gauffier** : The Tomato-Potyvirus system, a comparative system for natural and induced resistances associated with the initiation factors 4E
- 11.10 – 11.30 **Henryk Czosnek** : Discovery of gene networks sustaining resistance of tomato to TYLCV: lessons from transcriptome, metabolome and reverse genetic analyses
- 11.30 – 11.50 **Rafael Fernández-Muñoz** : Glandular trichomes from *S. pimpinellifolium* that confer resistance to whiteflies and reduce tomato yellow leaf curl disease
- 12.00 – 13.30 *Lunch at Université d'Avignon et des Pays de Vaucluse (UAPV)*
- 13.15 – 18.00 **Technical Visit**
- 18.45 – 23.00 **Visit of the Pope's Palace followed by the gala dinner**

FRIDAY 25TH APRIL

SESSION IV : IMPROVING FRUIT QUALITY

Chairperson : Arnaud Bovy (PRI Wageningen, Netherlands), Mathilde Causse (INRA GAFL, France)

- 09.00 – 09.30 **Arnaud Bovy** : The genetics of fruit quality traits in tomato
- 09.30 – 09.50 **Céline Bournonville** : Deciphering ascorbic acid regulation in tomato fruit ripening
- 09.50 – 10.10 **James Stroud** : Quantifying the strength of resistance to *Phytophthora infestans* conferred by different ph- gene combinations
- 10.10 – 10.30 **Hiroshi Matsunaga** : Tomato breeding for high yield and high sugar content under year-round hydroponic cultivation
- 10.30 – 11.00 *Coffee Break*
- 11.00 – 11.20 **Guillaume Bauchet** : GWAS reveals candidate regions for metabolic traits in tomato, *Solanum lycopersicum*
- 11.20 – 11.40 **Christophe Rothan** : EMS mutants for discovery of tomato genes and allelic variants through tilling and NGS-mapping
- 11.40 – 12.00 **Silvana Grandillo** : RNAseq transcriptome analysis of fruit pericarp in a set of *Solanum habrochaites* LA1777 ILs
- 12.00 – 14.00 *Lunch at Université d'Avignon et des Pays de Vaucluse (UAPV)*

SESSION V : GENOMICS AND SELECTION TOOLS

Chairperson : David Francis (Ohio State Univ. Wooster, USA), Rafael Fernández-Muñoz (IHSM Malaga, Spain)

- 14.00 – 14.30 **David Francis** : Genome assisted selection for tomato fruit nutritional quality, yield components, and disease resistance emphasizes a need for multi-trait indices
- 14.30 – 14.50 **Laura Pascual-Bañuls** : Deciphering the genetic control of quantitative traits with magic
- 14.50 – 15.10 **Akio Ohyama** : QTL analysis in recombinant inbred lines derived from a cross of two F1 hybrids of tomato
- 15.10 – 15.30 Closing Remarks

LIST OF TALKS

SESSION I : MANAGEMENT, DIVERSITY AND CHARACTERIZATION OF GENETIC RESOURCES

T001 – José Blanca

Tomato variation, origin and domestication

Blanca, J. (1), Montero-Pau, J. (1), Sauvage, C. (2), Bauchet, G. (2), Illa, E. (3), Díez, M. J. (1), Francis, D. (3), Causse, M. (2), van der Knaap, E. (3), Cañizares, J. (1)

- (1) Institute for the Conservation and Improvement of Agricultural Biodiversity (COMAV), Polytechnic University of Valencia, Spain
- (2) INRA, Unité de Génétique et Amélioration des Fruits et Légumes, Montfavet, France
- (3) Department of Horticulture and Crop Science, The Ohio State University/Ohio Agricultural Research and Development Center, USA

A collection of 949 accessions composed by *S. lycopersicum* var. *lycopersicum* (SLL; 514), *S. l.* var. *cerasiforme* (SLC; 291), *S. pimpinellifolium* (SP; 128), *S. galapagense* (SG;3), *S. neorickii* (SN; 1), *S. chmielewski* (Schm; 1) and 11 crosses between SP, SLC, SLL and *S. pennellii* was analyzed. The genotyping was carried out using the 7720 markers of the SolCAP genotyping platform and some key genes involved in the domestication process (CNR/FW2.2, SIKLUH/FW3.2, LC, FAS, OVATE and SUN). Using Principal Component Analyses (PCAs) based on the genetic data a new genetic-based classification divided the accessions in SP (121), SLC (236), SLL (492) and mixture (100). Within SP and Andean SLC a complex geographical structure correlated with the climate was found. Ecuadorian SP was remarkably differentiated from the rest of SP. In non-Andean SLC the structure found was not as marked. In a neighbor phylogenetic network SLC appeared intermediate between SP and SLC. Thus SLC was found to be a semi-domesticated with a key role in the tomato domestication. Andean SLC had a diversity comparable to SP while the non-Andean SLC had a lower diversity comparable to vintage SLL, indicating that the main bottleneck in the tomato domestication happened in the migration from the Andean region to Mesoamerica. Most SLC accessions were reported to have cherry-sized fruit, but interestingly some South American SLC were collected in markets as full-sized vintage tomatoes. Thus the vintage tomato in South America included SLC as well as SLL materials.

T002 – Jaime Prohens

Conventional and phenomic tools for typification and enhancement of local tomato varieties

Figàs, M. R. (1), Soler, S. (1), Raigón, M. D. (2), Fernández-de-Córdova, P. (1), Prohens, J. (1)

- (1) Institut de Conservació i Millora de l'Agrodiversitat Valencia, Universitat Politècnica de Valencia, Spain
- (2) Departament de Química, Universidad Politècnica de València, Spain

Local varieties of tomato (*Solanum lycopersicum* L.) are having an increasing demand in specialty markets, which demand fruits with a "flavour of the past" (Brugarolas et al., 2009). Enhancement of local varieties can be achieved through morphological characterization in order to identify the distinctive characteristics of local varieties, which is a first step for their typification and protection with a recognized status (mark of quality, protected designation of origin, etc.). The region of València (Spain) has a long horticultural tradition and several recognized varietal types exist. Among them, the Valenciana (prominently pointed heart-shaped), Penjar (long-term storage), and Borseta (pear-shaped) are very popular. Other more widely known varietal types, including the Cherry, Cor (heart-shaped), Plana (flattened), Pruna (canning), or Redona (round) are also common. We characterized 69 local accessions from the region of Valencia corresponding to these eight varietal types with 102 descriptors, including conventional descriptors (IPGRI, 1996) and fruit morphology traits evaluated with the phenomics software tool Tomato Analyzer (Rodríguez et al., 2010). A large morphological diversity has been found in the collection, in particular for fruit traits. Differences for many traits were also found among most varietal groups, except for Cor, Redona and Valenciana for conventional descriptors and Cor and Redona for Tomato Analyzer traits. However, multivariate principal components analysis including both conventional descriptors and Tomato Analyzer traits allowed a good separation of all varietal groups. The results show that the combination of conventional descriptors and phenomic tools like Tomato Analyzer are a powerful tool for the detailed characterization of local tomato accessions. The information is useful for the delimitation of varietal groups of the most demanded local traditional varieties, like the Valenciana group, which is a necessary information for their typification, protection, and enhancement.

References

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Keywords: diversity, local varieties, *Solanum lycopersicum*, Tomato Analyzer, varietal group

T003 – Irina Balashova

The heritability analysis of main tomato traits for the special hydroponic technology (on narrow benches)

Balashova, I. (1), Sirota, S. (1), Balashova, N. (1), Kozar, E. (1), Pinchuk, E. (1)

(1) Res. Inst. of Veget. Breed., Russia

Division of gametophyte and sporophyte cycles in *Telomophytae* evolution ensured significant advantages for adaptation of higher plants in environment. But that division conditioned limitations connected with separate selection of gametes. On the other hand, separate selection of gametes can be used in the breeding of adaptive forms in gametes selection technologies. Effect of gametes selection is conditioned of correlation links between the level of necessary traits expression on gamete stage and the level of such traits expression on the stage of zygote (mature plant). So, determination of such links is the first and necessary step of gametes selection. The idea of the special mother plants selection has been used for the improvement of tomato gametes selection. The goal of the study - to obtain new tomato forms could be cultivated under special hydroponic conditions (on narrow benches). We haven't enough varieties and hybrids for this technology now, because the model of tomato for this purpose must have many advantages. Main of its are: the short height of the stem (shorter than 50 cm), the high productivity, the early ripening and the resistance to main diseases of greenhouses. We studied the heritability of these traits in F_1 hybrid progenies of tomato with help the coefficient of heritability. It can be calculated with correlation and dispersion analyses. Using different types of mothers forms from the collection of marker mutants (with *d*-gene and without it), F_1 hybrids, obtaining with the only male form (v. Talalikhin), and correlation analysis, we show, that such traits of productivity as "the mass of one fruit" ($h^2=0.99$) and "the quantity of fruits on the plant" ($h^2=0.96$) could be inherited by mother's line. Heritability of "the short stem" and "the early ripening" has been studied with 2-factorial dispersion complex. It has been established, that such traits as "the short stem" ($h^2=0.83$) and "the early ripening" ($h^2=0.60$) could be inherited by father's line. So, we bred 9 mothers and 7 fathers forms from more than 1000 samples of tomato. Its have been used for crossings and F_1 hybrid progeny has been obtained.

Keywords: tomato, gametes selection, mother's forms

T004 - Christopher Sauvage

The study of domestication reveals a loss of diversity, signature of selection and transcriptional modifications in tomato (*Solanum lycopersicum*)

Sauvage, C. (1), Nabholz, B. (2), Gautier, S. (3), Ruiz, M. (3), Causse, M. (1), David, J. (4), Glémin, S. (5)

- (1) INRA, Génétique et Amélioration des Fruits et Légumes, Montfavet, France
- (2) Institut des Sciences de l'Evolution. Université Montpellier 2, France
- (3) CIRAD, UMR AGAP, Montferrier-sur-Lez, France
- (4) Montpellier SupAgro, Amélioration Génétique et Adaptation des Plantes Méditerranéennes et Tropicales, France
- (5) Institut des Sciences de l'Evolution de Montpellier, Centre National de la Recherche Scientifique, Université Montpellier 2, France

Over the last 13,000 years, plant domestication has led to considerable phenotypic modifications through selective breeding. Though changes in key traits such as fruit morphology have been well documented, little is known about the underlying molecular consequences. The comparative study of cultivated and current wild type crop populations should measure the effects of domestication, as this phenomenon is intense, recent and accompanied by selective signatures. Up to now, there have been a limited number of large-scale screens to detect selection in crops using high-throughput Sequencing (HTS). Using the autogamous crop *Solanum lycopersicum* (tomato) and its wild relative (*S. pimpinellifolium*), we investigated the genome-wide signature of domestication at the molecular level.

The HTS of 10 wild and 10 cultivated transcriptomes revealed that 12168/34727 (35%) of the genes experienced a 33% averaged loss of nucleotide diversity following domestication. Regions associated to a loss of diversity, representing candidate regions selected during domestication, were identified across the genome. Tajima's D statistic supported these observations and revealed that 4.4% of the genes were under positive or balancing selection. Average π_N / π_S ratios were estimated to 0.252 and 0.291 in cultivated and wild types, respectively. Moreover, 1.87% and 3.01% of the genes showed ratios over 1, suggesting a signature of positive selection. The analysis of site frequency spectrum across the genome revealed at least two regions showing hard selective sweep. At the transcriptional level, domestication significantly affected 3.39% of the genes with fold changes ranging from -8.99x to +7.85x.

Overall, we evidenced that domestication in tomato has led to loss of nucleotide diversity detectable at the scale of the genome. Moreover, high deviations from neutrality and transcriptional modifications were identified, providing new insights into the domestication process of tomato as well as valuable markers for breeding.

Keywords: genomic, domestication, tomato, diversity

T005 - Niels Müller

Natural variation in circadian rhythms suggests an effect of domestication on the circadian clock of tomato

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(1) Max Planck Institute for Plant Breeding Research, Germany

(2) INRA, Institut Jean-Pierre Bourgin, France

The circadian clock controls many important aspects of plant physiology and development, including several traits of agronomical significance in crop plants. In addition, natural variation in circadian rhythms appears to be important for adaptation to specific environments. Still, quantitative differences in circadian rhythms due to artificial selection have not yet been reported. By monitoring circadian leaf movements and transcript rhythms, we found that cultivated tomato (*Solanum lycopersicum*) exhibits different rhythms than its wild relatives, indicating that domestication or early breeding has had an effect on the tomato circadian clock. Quantitative trait locus (QTL) analysis in two populations derived from crosses between cultivated and wild tomato accessions identified one locus controlling differences in circadian rhythms in both populations suggesting that a single event that occurred in cultivated tomato is responsible for those differences. Fine mapping of this locus resulted in a region containing 13 candidate genes. Near isogenic lines differing in those 13 genes exhibit differences in seedling growth in the greenhouse indicating that variation in circadian rhythms could be changing plant growth under diurnal conditions. In conclusion, our work suggests that humans may have selected for altered circadian rhythms during tomato domestication or early breeding to adapt the species to agricultural environments.

Keywords: circadian rhythms, natural variation, QTL, domestication

SESSION II : GENETIC BASIS FOR SELECTION AND ADAPTATION TO ABIOTIC STRESS

T006 - Francisco Perez-Alfocea

Rootstock-mediated alleviation of abiotic stress in tomato: what's beyond vigour?

Martínez-Andújar, C. (1), Albacete, A. (1), Ghanem, M. E. (1,2), Pérez-Alfocea F.(1)

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(2) Present address: ICARDA, Rabat Instituts, Rabat, Morocco

Securing food production for the growing population will require closing the gap between potential crop productivity under optimal conditions and the yield captured by farmers under a changing environment, which is termed as 'agronomical stability'. One major area that has been comparatively neglected is the role of the plant root system in maximising water and nutrients capture and sensing and adjusting to environmental stresses in the soil to control plant growth and shoot physiology, and ultimately agricultural productivity. Grafting is a surgical technique that allows a precise assessment of the effect of altering root traits on crop performance, regardless of any shoot traits, since the scion (shoot) is constant. Indeed, grafting can be considered as an efficient alternative to breeding that allows the rapid exploitation of root-mediated traits from wild species that are usually complex through genetic crosses. Genetic markers of those traits can be identified from recombinant inbred populations through rootstock phenotyping, QTL and fine mapping analyses. Although, in general, the scion genotype has a dominant effect on most agronomic traits, rootstock introduces new sources of genotypic and phenotypic variability due to rootstock x scion x environment interactions. Those interactions are essentially based on hydraulic and chemical signals through the xylem (root-to-shoot) and the phloem (shoot-to-root). Even though maximizing crop yield depends on the leaves receiving an optimal supply of water and mineral nutrients from the root system via the xylem, the integrated plasticity probably involves long-distance communication (small organic molecules, proteins, RNA,...) between different organs with hormones playing an essential role. Abiotic stresses as soil drying and salinization alter these xylem fluxes, and modern -omics techniques offer unparalleled opportunities to understand the complexity of these responses. Indeed, by using high throughput U-HPLC-MS technology, about 800 metabolites in a range of m/z 90-500 have been detected in the root xylem sap of grafted tomato plants, identifying rootstock-genotype specific metabolomic fingerprinting. The number of those metabolites with a biological effect in the scion needs to be addressed, not only in response to biotic and abiotic constraints, but also those with a potential toxicological/nutraceutical/organoleptic influence on fruit quality. Moreover, functional and reciprocal grafting studies have demonstrated that the rootstock can influence shoot physiology by altering the hormonal status, although a regulatory feedback mechanism of recirculation between xylem and phloem also exists. From the point of view of rootstock breeding, the selection for vigour has been the major contribution so far, since resistance to many soilborne pathogens is mainly due to 'escape' mechanisms based on induced vigour. A second question now is to find an adequate vigour balance among rootstock, the scion variety and the growing environment to avoid negative effects. This is the main reason why most current commercial rootstocks are derived from interspecific crosses between *Solanum lycopersicum* x *S. habrochaites*, while just a few are intraspecific from *S. lycopersicum* or derived from closer wild relatives such as *S. pimpinellifolium*. Although this vigour is usually behind the rootstock mediated improvement under both biotic and abiotic conditions, some stress-specific rootstocks and traits beyond the vigour are being discovered. In this regard, the potentiality of vegetable grafting is only at the tip of the iceberg, and it is as broad as genetic variability enabling to cross the barriers between rootstocks and scions. Root-targeted breeding or biotechnology may allow grafting onto

rootstocks with altered hormone metabolism (cytokinins, ACC, abscisic acid, gibberellins, strigolactones, auxins, brassinosteroids,...) to define the role of root-derived hormones in mediating crop performance. Therefore, the use of -omics techniques to underpin rootstock-mediated plant improvement along with root system engineering and breeding provides new opportunities to maintain sustainable crop production under changing environmental conditions, while minimising the demand for new resources.

References

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Keywords: xylomics, phytohormones, salinity, drought, yield stability

T007 - Eliezer Lifschitz

The making of a sympodial plant: the role of florigen as a regulator and the great communicator of the shoot system in tomato

Lifschitz, E. (1), Eshed, Y. (2), Shalit-Kaneh, A. (1), Eviatar-Ribak, T. (1)

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(2) Dept. of Plant Sciences, Weizmann Institute of Science, Israel

The introduction of the recessive *SP* gene into cultivars revolutionized the agrotechnical landscape of the tomato industry. Our laboratories investigate the developmental genetics of the 'determinate' and 'indeterminate' growth habits and the global regulatory mechanisms that coordinate the architecture of the shoot system. *SP* is one of the founder genes of the CETS family (Pnueli et al., 1998, 2001) that function as an antagonist of *SFT* the gene encoding the systemic FLORIGEN (Lifschitz et al., 2006, Lifschitz and Eshed 2006). Gene-dosage analyses in tomato indicated that in inducing transition to flowering the mobile florigen modifies existing meristem-specific ratios, as opposed to absolute levels, between *SINGLE FLOWER TRUSS (SFT)* the gene generating florigen and *SELF PRUNING (SP)*. By the same token the florigen-mediated *SFT/SP* ratio regulates also the morphogenesis of the inflorescences, the stems and the compound leaves. Florigen therefore functions as generic growth hormone and boosting flowering reflects its fundamental function of as a modifier of growth and termination across the shoot system in all flowering plants (Shalit et al., 2009).

We will describe how new genetic interactions between **7** members of the CETS family may facilitate their potential exploitation in breeding programs and basic developmental research. We will proceed to describe the molecular properties of the CETS proteins and of their interacting partners. Finally, we will classify, based on deep RNA sequencing, the developmental circuits that response, in an organ and context-specific manners, to endogenous and systemic florigen.

Keywords: florigen, determinate, indeterminate, growth habit, CETS genes

T008 - Dina Saint-Clair

Genetic and genomic dissection of traits associated with water stress tolerance in near-isogenic lines derived from wild tomato

St.Clair, D. (1), Arms, E. (1), Lounsbery, J. (1), Bloom, A. (1)

(1) University of California-Davis, USA

Increasingly limited fresh-water resources and global climate change pose significant challenges to agricultural production systems worldwide. Breeding for tolerance to water stress and increased water use efficiency (WUE) would improve the sustainability of crop production by requiring less water for a given level of yield. A wild tomato (*S. habrochaites*) that originates from the Peruvian Andes exhibits tolerance to abiotic stresses, including limited water and chilling temperatures. Rapid-onset water stress is induced by root chilling (6°C), reducing water movement from roots to shoots. *S. habrochaites* responds to root chilling by closing stomata and maintaining shoot turgor, while cultivated tomato (*S. lycopersicum*) fails to close stomata and wilts. This trait (shoot turgor maintenance under root chilling) is controlled by a major QTL (*stm9*) on chromosome 9. We high-resolution mapped *stm9* to a 0.32 cM region using a set of recombinant sub-near-isogenic lines (sub-NILs) for chromosome 9 from *S. habrochaites*. To determine if other traits indicative of (or potentially associated with) water stress tolerance map to this region, we evaluated this same set of sub-NILs in replicated field experiments in 2012 and 2013 under severely restricted irrigation (i.e., slow-onset water stress). Traits measured included total fruit yield, shoot dry weight (biomass accumulation), maturity, specific leaf area, leaf phenolic content, and mature leaf delta-13C, a measure of carbon isotope discrimination (13C:12C). Delta-13C is correlated with WUE. Trait data obtained for the sub-NILs was used to map QTL, determine QTL linkage relationships and detect QTL x E interactions. Almost all traits measured in the field experiments segregated within the introgressed region from *S. habrochaites* that also contains *stm9*. The majority of trait QTLs were closely linked to (but not coincidental with) *stm9*. Several QTLs for delta-13C were detected and were conferred by alleles from both the cultivated parent and *S. habrochaites*. In the *S. lycopersicum* reference genome, this chromosome 9 region is gene-rich. We are in the process of sequencing and assembling *S. habrochaites* genomic BACs for chromosome 9 to facilitate identification of QTL candidate genes for subsequent functional testing and enable structural genomic comparisons to the cultivated tomato genome. Our evidence to date suggests that genetic elements within this chromosome 9 region in *S. habrochaites* play important roles in plant responses to water stress and chilling temperatures. Our research findings will aid crop improvement efforts in tomato and help address challenges imposed by limited fresh-water resources.

Keywords: water stress tolerance, wild tomato, QTL

T009 - Vincent Truffault

Metabolic profiling of transgenic lines silenced for an MDHAR gene and impact on ascorbate redox state and carbon metabolism.

Truffault, V. (1,2); Costagliola, G. (2); Gautier, H. (2); Stevens, R. (1)

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(2) INRA, Plantes et Systèmes de Culture Horticole, Avignon, France

Ascorbate is a key molecule involved in numerous cellular processes in plants and is an essential antioxidant protecting the cell from reactive oxygen species (ROS) damage in case of environmental stress. The cellular concentration of ascorbate depends on its biosynthesis, recycling and degradation: these are under genetic control and closely related to environmental conditions.

The recycling pathway controls the redox state of the ascorbate pool and is especially important during stress responses. Recycling of oxidized ascorbate to the reduced form occurs via two enzymes monodehydroascorbate reductase (MDHAR) and dehydroascorbate reductase (DHAR). Transgenic lines silenced for an MDHAR gene in a cherry tomato cultivar have been generated. Our previous results have shown that the activity of MDHAR is correlated with reduced ascorbate and fruit firmness in tomatoes. The objective of this study was to determine whether limiting turnover of the ascorbate pool may affect central metabolism of tomato plantlets and fruits.

MDHAR modified plantlets showed a slower early growth and fruit size was also affected. Silencing MDHAR induced a modification of carbohydrate metabolism affecting particularly sugar concentrations. The ascorbate pool and ascorbate degradation products were also modified. Transgenic lines showed other strong variations of metabolites compared to wild type.

Ascorbate concentration was measured by spectrophotometry. Metabolite profiling of transgenic lines was provided by a GC-MS based survey of tomato metabolites. Results are discussed and shed light on the impact of a modified recycling pathway on metabolism in plantlets and fruits of tomato.

Keywords: ascorbate-recycling, metabolic profiling, redox state

T010 - José Jimenez-Gomez

Analysis of natural variation in canalization at the transcriptional level

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The term “canalization” describes genetic buffering that has evolved under natural selection to maintain phenotypic stability despite genetic and environmental perturbations. Increased canalization is of interest for crops, where phenotypic stability is required across years and conditions. On the other hand, increasing phenotypic variability under stress conditions is advantageous in the wild, and allows plant populations to explore different strategies for stress survival and adaptation. To understand canalization at the molecular level we measured differences in stochastic noise of mRNA abundance in response to stress. For this we performed RNA-seq in cultivated tomato and its wild relative *Solanum pennellii* under control and drought conditions. Our results show how transcripts change expression variability in response to drought and describe the differences in the responses from each genotype. We then perform RNA-seq in a F1 hybrid between these two species to identify the genetic basis of these differences through allele specific expression. In sum, our work helps understanding the mechanisms that regulate phenotypic stability in the light of global climatic changes and increasing water deficit.

Keywords: natural variation, RNA-seq, eQTL, allele specific expression, canalization

T011 - Lukas Mueller

The SOL Genomics Network Workshop

SESSION III : GENETIC BASIS AND SELECTION FOR RESISTANCE TO DISEASES

T012 - Laura Rose

Evolutionary genetics of disease resistance in wild tomatoes

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Resistance genes typically show evidence of rapid protein evolution in natural host populations. Variability at these loci may be the direct result of a co-evolutionary arms race between host and pathogen. However, the prevalence of pseudogenes also segregating in natural populations at well-known resistance genes indicates that the strength of natural selection likely fluctuates and these genes may be subject to bouts of relaxed constraint as well. In my lab, we investigate the strength and target of natural selection within resistance genes present in natural host plant populations of *Arabidopsis* and wild tomatoes. Using a combination of population genetic studies of well-characterized pathogen resistance genes and functional studies involving protein expression, enzymatic assays, plant transformation and pathogen infection studies, we have determined how naturally occurring protein variation in host populations affects defense to pathogens at the individual level. I will describe our recent results from wild tomato species and how we are expanding these studies to include population genomic analyses to understand how resistance genes evolve as a result of host-parasite interactions.

Keywords: evolution, resistance

T013 - Mara R. Ercolano

Genomic approach for identifying and selecting disease resistance candidate genes in tomato

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In last years, achievements in tomato breeding for disease resistance have been successfully obtained through both genetic and molecular approaches. An unprecedented challenge is now to capitalize on the genetic and genomic attempts made in this species. In this study, we investigated the physical arrangement of candidate pathogen recognition genes in the tomato genome and explored pathogen recognition gene cluster composition. Investigation of the functional and spatial arrangement of pathogen recognition genes in the tomato genome can be useful in reconstructing the history of chromosomal rearrangements that shaped their genome architecture. We also deciphered the evolutionary history of pathogen recognition gene loci associated with documented resistance function. Our findings suggested that the reconstruction of evolution events can help to understand mechanisms of adaptive selection and speed up the identification of new sources of resistance. Finally, we demonstrated that information on the tomato genome can be used predictively to link resistance function with specific sequences. An integrated genomic approach for identifying new resistance (R) gene candidates was developed. An R gene functional map was created by co-localization of candidate pathogen recognition genes and anchoring molecular markers associated with resistance phenotypes and an in-depth characterization of identified pathogen recognition genes was performed. Puzzling information were collected and combined in order to obtain a synergy between different approaches. Our strategy was developed in order to reduce the time required for R genes identification, a critical step towards modern genome breeding. In many cases, a predicted protein was narrowed down in a small region, allowing the identification of one or few candidates, now available for exploiting their specific function. Our attempt captured fundamental aspects of data integration contributing to pinpoint key steps in genetic, genomic, and phenotypic data synthesis for a better R gene identification. Accumulated knowledge of disease resistance should facilitate rapid advancement of disease resistance breeding.

Keywords: *Solanum lycopersicum*, R loci, physical map, predicted proteins, genomic approach

T014 - Yuling Bai

Fine mapping of the tomato yellow leaf curl virus resistance gene Ty-2 on chromosome 11 of tomato

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Resistances to begomoviruses, including bipartite tomato mottle virus (ToMoV) and monopartite tomato yellow leaf curl virus (TYLCV), have been introgressed to cultivated tomato (*Solanum lycopersicum*) from wild tomato accessions. We have shown that chromosomal rearrangements have hampered mapping and breeding of the widely used *Ty-1* and *Ty-3* genes derived from *S. chilense*. A similar situation may have occurred to another major TYLCV resistance gene, *Ty-2* from *S. habrochaites* f. *glabratum* accession 'B6013'. The *Ty-2* gene was previously mapped to a 19 cM region on the long arm of chromosome 11. In our recent study, approximately 11,000 plants were screened and nearly 157 recombination events were identified between the flanking molecular markers C2_At1g07960 (82.5 cM, physical distance 51.39 Mb) and T0302 (89 cM, 51.88 Mb). Molecular marker analysis of recombinants and TYLCV evaluation of progeny from these recombinants, localized *Ty-2* to an approximately 300,000 bp interval between markers UP8 (51.35 Mb) and M1 (51.65 Mb). No recombinants were identified between TG36 and C2_At3g52090, a region of at least 115 kb, indicating severe recombination suppression in this region. Due to the small interval, Fluorescence in situ hybridization (FISH) analysis failed to clarify whether recombination suppression is caused by chromosomal rearrangements. Candidate genes predicted based on tomato genome annotation were analyzed by RT-PCR and virus induced gene silencing. Results indicate that the NBS gene family present in the *Ty-2* region is likely not responsible for the *Ty-2*-conferred resistance and that a combination of function of two genes might play a role in *Ty-2*-conferred resistance. Several markers very tightly linked to the *Ty-2* locus are presented and useful for marker assisted selection in breeding programs to introgress *Ty-2* for begomovirus resistance.

Keywords: breeding, resistance, tomato, Tomato yellow leaf curling virus (TYLCV), virus-induced gene silencing

T015 - Liliana Stamova

Resistance to fusarium crown and root rot

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Fusarium Crown and Root Rot of tomato incited by *Fusarium oxysporum* f.sp. *radicis-lycopersici* (FORL) is an important disease with the potential to limit yields of greenhouse and field crops. First found in 1971, FORL is now distributed worldwide. The only gene used in breeding programs – gene *Frl*, has been derived from *S. peruvianum*. In 1995 we reported additional sources of resistance found in Bulgarian tomato lines. Because isolates able to overcome gene *Frl* have been reported in France and Canada, it is worth looking for new genetic materials resistant to FORL. In this report we present our results on the screening of 110 cultivated type tomato lines against three California isolates of FORL, which vary in virulence. The lines included in the screening represent late generations of interspecific crosses with the following wild species: *S. pimpinellifolium*, *S. pennellii*, *S. habrochaites*, *S. chilense* and *S. peruvianum*. The isolates of FORL were collected in commercial fields in Yolo County, California, USA. Root-dip assay technique was used to inoculate the genetic materials. Plant symptoms were evaluated two weeks after inoculation. The lines with gene *Frl* (IRB 301-31, Momor and Mogeor) developed severe (isolate 14X97) or mild (isolates 226 and 52-2) symptoms; therefore, isolates of FORL from California may differ in virulence from isolates of FORL used in the original discovery of gene *Frl*. Several lines (Forly lines) with *S. pimpinellifolium* in their pedigree showed high levels of resistance to isolates 226 and 52-2 and very mild symptoms when challenged with the virulent isolate 14X97. Their strong ability to develop additional roots helped tolerate infection with isolate 14X97. Two lines from crosses with *S. pennellii* also showed resistance to the three California isolates. In addition, several lines obtained from crosses with *S. chilense* and *S. habrochaites* as well as lines having two wild species in their genetic constitution were selected for their resistance to FORL. We observed segregation ratios of resistant to susceptible plants close to 3R:1S in F1 (Forly x Moneymaker) and 15R:1S in F2 (Forly x IRB 301-31). These results suggest that the resistance to FORL in Forly lines is controlled by a single dominant gene that is different from gene *Frl*.

Keywords: tomato, resistance, Fusarium Crown, FORL

T016 - Sophie Rommel

Resistance-shaping mirnas in wild tomatoes

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Introgression of resistance genes (*R* genes) is commonly used to introduce pathogen resistance from wild tomatoes into the cultivated tomato. Therefore, wild tomatoes play a crucial role in breeding. Identification of novel *R* genes and resistance specificities in wild tomatoes is the aim of several labs worldwide, while less focus has been given to the study of endogenous plant mechanisms controlling transcription and translation of resistance genes and how this influences pathogen resistance in the host. Many plant *R* genes encode for proteins containing nucleotide binding site-leucine rich repeat (NBS-LRR) motifs. Recently novel microRNAs (miRNAs) have been described in *Solanum lycopersicum* that contribute to *NBS-LRR* post-transcriptional regulation. In our study, we will investigate how levels of miRNA expression affect disease resistance. We will analyse the evolutionary history of the miRNA superfamily miR482/2118 that regulates *NBS-LRR* genes in the nightshade family (Solanaceae). Our preliminary analyses reveal that the gene-family size has varied over evolutionary time, but appears to be more stable within the Solanaceae as compared with other flowering plants. Using phylogenetic methods, we detected an excess of lineage-specific losses relative to gains. Currently we are investigating the rate of sequence evolution of members of this gene family in wild tomatoes and other close relatives in the Solanaceae. This will indicate how variation is distributed across the domains of the miRNA genes and which regions are potential targets of natural selection. An ultimate goal of our research is to understand the link between naturally occurring sequence variation in members of this gene family and host plant defense. Upregulation of these miRNA genes is predicted to negatively regulate their targets: *NBS-LRRs*. Therefore, host disease resistance may be perturbed through the action of these genes. An effective pathogen would specifically up-regulate host miRNA to suppress levels of *NBS-LRRs*. However, many pathogens do the opposite, and thereby potentially activate host resistance inadvertently. Using the oomycete pathogen *Phytophthora infestans* as a model, we will investigate how levels of miRNAs and *NBS-LRRs* are affected during the infection process. Our preliminary inoculations with a panel of different strains of *Solanum*-specific *P. infestans* indicate that levels of host miRNA do differs depending of which pathogen strain is used. With this system in hand, we aim to determine how levels and target specificity of these miRNA gene family members contribute to host plant resistance in wild and cultivated tomato.

Keywords: NBS-LRRs, wild tomatoes, miRNA, *Phytophthora infestans*

T017 - Camille Gauffier

The Tomato-Potyvirus system, a comparative system for natural and induced resistances associated with the initiation factors 4E

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Tomato (*Solanum lycopersicum*) is one of the most cultivated vegetable in the world, but suffers from important yield losses caused by viral diseases. Therefore, the development and use of cultivars that are genetically resistant to viruses has become a critical factor of competitiveness for both breeders and producers and one of the key stakes for sustainable agriculture. In this context, generating new resistance alleles using biotechnological approaches (e.g., TILLING) appears as a powerful tool to diversify host targets to promote resistance against viruses.

Most characterized recessive resistances to potyviruses so far are natural variant of the translational initiation factor eIF4E. Those variants often encode functional eIF4E proteins but have lost the ability to interact with the viral protein VPg. In tomato, a broad-spectrum resistance to Potyvirus is associated with the natural resistance allele *pot1 - eIF4E1* from *Solanum habrochaites* PI247087. More recently null *eIF4E* alleles were obtained by TILLING but strikingly, the resistance spectrum associated with the null *eif4e1* allele is considerably narrower than the one associated with the natural resistance allele *pot1-eIF4E1*. Understanding the apparent discrepancies between those two resistances -natural and induced- could be important to help developing more efficient TILLING-based resistances to pathogens.

Therefore, we are investigating the differences between the lines harbouring those two alleles, as well as the implication of the others tomato *eIF4E* genes – *eIF4E2* or *eIFiso4E*. As a first step, genetic effects are investigated by swapping the genetic backgrounds between the natural and induced *eIF4E1* resistance alleles but also by combining KO mutations affecting several *eIF4E* genes to see whether this will impact on the plant resistance spectrum to potyviruses. Secondly, the type of mutation affecting *eIF4E1* (AA changes vs KO mutant) is also investigated and especially the incidence that a null mutation may have on the protein level accumulation of the other *eIF4E*.

Besides its applications on crop breeding, these experiments could shed new light on the Tomato/potyvirus pathosystem and redundancy among *eIF4E* genes.

Keywords: *S.lycopersicum*, *S.habrochaites*, eIF4E, potyvirus, TILLING

T018 - Henryk Czosnek

Discovery of gene networks sustaining resistance of tomato to TYLCV: lessons from transcriptome, metabolome and reverse genetic analyses

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Tomato yellow leaf curl virus (TYLCV), a whitefly transmitted geminivirus, threatens tomato cultivation worldwide. A tomato line resistant to TYLCV (coined R) has been developed by classical breeding, using the wild tomato species *Solanum habrochaites* as resistance source [1]; a susceptible line (coined S) was generated during the same program. To identify the genes and defense signaling pathways underlying resistance to TYLCV infection, we postulated that: 1) genes for TYLCV resistance have been introgressed from the wild tomato species; 2) resistance is sustained by networks of genes that interrelate by positive and negative signals with known stress response pathways; 3) genes preferentially expressed in the R line are likely to be part of this network, silencing of genes located at important nodes of the network should lead to the collapse of resistance; 4) primary and secondary metabolites may serve as signaling regulatory molecules. Subtraction cDNA libraries of R and S lines, before and after inoculation, indicated that ~70 transcripts represented genes preferentially expressed in R plants [2]. Silencing of genes of R plants encoding proteins that may be involved in signal transduction such as *Permease I*, *Hexose transporter LeHT1*, *Lipocalin* and in water management such as *Tonoplast-intrinsic aquaporins TIP1;1* and *TIP2;2*, all induced the collapse of resistance upon virus infection. Since sugars play a role as chemical chaperons, the effect of *LeHT1* silencing on the R plant transcriptome was studied using a home-designed oligonucleotide microarray and the effect on mono- and disaccharide concentrations by GCMS. The transcriptional pattern of *LeHT1*-silenced infected R plants was closer to that of infected R than infected S plants. In the silenced R plants, the sugar profile was similar to that of S plants; the concentration of monosaccharides decreased, hexoses were not internalized into the cell in order to act as defense signaling molecules, and resistance collapsed [3]. Silencing *TIP* genes in R line and in *Arabidopsis* dramatically decreased the resistance of these plants to virus infection. Constitutive expression of *TIP2;2* in transgenic *Arabidopsis* and tomato plants was correlated with increased transpiration and photosynthesis, enhanced TYLCV resistance and with decreased ABA and increased SA levels. Sequential gene silencing allowed to determine a first draft of the gene hierarchy in the silencing network: Aquaporin Tip1;1 - Invertase LIN6 - hexose transporter LeHt1 - Lipocalin ; Permease I is not part of this path.

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Keywords: gene networks, metabolome, transcriptome, reverse genetics, resistance

T019 - Rafael Fernández-Muñoz

Glandular trichomes from *S. pimpinellifolium* that confer resistance to whiteflies and reduce tomato yellow leaf curl disease

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The accession TO-937 from *S. pimpinellifolium* shows acylsucrose-producing type IV glandular trichomes that are absent in the cultivated species and that confer resistance to a number of arthropod pests such as the spider mites *Tetranychus urticae* and *T. evansii*. The whitefly *Bemisia tabaci* is a threat for tomato cultivation worldwide as it is vector of a number of viral diseases among which tomato yellow leaf curl disease caused by TYLCV is the main crop-limiting factor in Mediterranean countries. TO-937 was used as donor parent in a backcross program to introgress pest resistance into the cultivated tomato and a number of advanced backcross lines were obtained. One of these, the BC₃ line ABL 14-8 was studied for resistance to whitefly *Bemisia tabaci* biotype Q and a more advanced line assayed against the criniviruses-transmitting whitefly *Trialeurodes vaporariorum*. The improved lines showed antixenosis effects on the whiteflies in preference bioassays and slight antibiosis effects. The feeding behavior of *B. tabaci* on leaves of ABL 14-8 was studied and difficulties for probing and reaching the phloem derived from non-acceptance of epicuticular secretions were revealed. Then, incidence of the phloem-limited virus TYLCV in healthy/infected ABL 14-8 plants was studied in small-scale greenhouse experiments using viruliferous/healthy whiteflies. The superficial type of resistance observed in ABL 14-8 against *B. tabaci* probing significantly reduced primary and secondary spread of TYLCV. However, these results were not so promising for young plantlets in which sufficient acylsucrose production is still not achieved. As a way to accomplish early expression of acylsucrose secretion and enhanced whitefly and virus protection, application of the volatile precursor of induced defense hormone jasmonic acid, methyl-jasmonate (MeJA), was assayed. MeJA application resulted in elevated glandular trichomes densities and acylsucrose and phenolic compounds secretions in young plantlets of an advanced backcross line in BC₅ derived from ABL 14-8. MeJA-treated BC₅ line plantlets showed higher repellence to *B. tabaci* and reduced primary spread of TYLCV to levels comparable to those of adult plants. To study inheritance of those traits, a RIL population derived from the cross *S. lycopersicum* cv. MoneyMaker x *S. pimpinellifolium* acc. TO-937 was obtained and a saturated linkage map was constructed. QTLs for trichome density and total acylsucrose production were located on chromosome 2, co-localizing with QTLs for spider mite and *B. tabaci* resistances while QTLs for several types of acylsucroses revealed by thin layer chromatography were located on chromosomes 1 and 11. Studies on transcriptomics of trichome formation and acylsucrose production in advanced nearly-isogenic lines. Finally, we are also in the way to combine the trichome-borne whitefly resistance with plant resistance to the virus.

SESSION IV : IMPROVING FRUIT QUALITY

T020 - Arnaud Bovy

The genetics of fruit quality traits in tomato

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Traditionally the most important targets in fruit and vegetable breeding are producer traits, such as yield, disease resistance and shelf life. In the past decade, however, the importance of consumer-driven quality traits, such as flavour, nutritional value and appearance have been increasingly recognised. This is not only due to growing consumer demands for more healthy food with an excellent quality, but also to the availability of various omics tools, which make it possible to study complex, multifactorial traits, such as flavour and health. These quality traits are to a large extent determined by the phytochemical composition of our food products. In order to improve the consumer-quality of food crops, we aim to explore and utilise the natural diversity for flavour- and health-related metabolites. Our general strategy involves the elucidation of biochemical pathways underlying important quality traits using a genetical genomics approach and the isolation of the responsible genes underlying quality-related mQTLs.

Tomato is one of the most important vegetable crops worldwide and an important model for fleshy fruits. Cultivated tomato has a very narrow genetic base, due to severe genetic bottlenecks experienced during domestication. In contrast, a lot of genetic diversity can be found in wild relatives of tomato, which are therefore used as novel sources of important agronomic and quality traits. We analysed a diverse collection of both cultivated and wild tomato germplasm for variation in colour-, health- and flavour-related metabolites, in order to elucidate the biochemical and molecular mechanisms underlying these quality traits. In this lecture, several examples of this approach will be discussed.

This research was funded by the Dutch NWO genomics initiative Centre for Biosystems Genomics and the EU project EUSOL (PL-016214).

T021 - Céline Bournonville

Deciphering ascorbic acid regulation in tomato fruit ripening

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The ascorbic acid (AsA) is an essential antioxidant in both plants and humans. Plant-derived AsA is the major source of vitamin C in the human diet. In addition to its effect on tomato nutritional value, increasing tomato AsA content would likely affect postharvest storage and resistance to fungal pathogens of the fruit. While AsA metabolism is well characterized, the mechanisms involved in its regulation remain poorly understood. Recent studies in *Arabidopsis* indicate that this pathway is highly regulated at transcriptional level by single regulatory proteins. In order to investigate the regulation of AsA in tomato (*Solanum lycopersicum*) fruit, we undertook two different strategies to identify regulatory factors controlling fruit AsA content.

First strategy is based on the screening of an EMS tomato mutant population in the miniature cultivar Micro-Tom for identifying mutant lines with AsA-deficient or AsA-enriched fruits. Among the 500 M2 mutant families screened for AsA content, four mutant lines with increases in fruit AsA content ranging from 2.5 to 4 fold and 2 mutant lines with reduced AsA-content ranging from 0.2 to 0.4 fold were selected. These lines are currently studied for pathogen resistance. In addition, the isolation of the causal mutations has been undertaken by a BSA-NGS mapping approach for two of the mutant lines with AsA-enriched fruit. Second strategy is based on the exploitation of genomics data resulting from comparative analysis of four tomato lines silenced for key enzymes of the AsA biosynthetic and recycling pathways (Garcia *et al.* 2009). Correlation analyses between fruit AsA content and transcript levels identified a large number of transcription factors (TF) putatively regulated by AsA. Among them, fifteen candidate genes displaying high correlation ($R^2 \geq 0.95$) with AsA content have been selected and are currently studied for their implication in the regulation of fruit AsA content using various approaches. Current results confirm that several of the TFs studied are indeed linked to AsA, thus providing new targets for studying AsA accumulation in the fruit. Strategies, tools and current results will be presented and discussed.

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Keywords: ascorbic acid, EMS mutant, tomato fruit

T022 - James Stroud

Quantifying the strength of resistance to *Phytophthora infestans* conferred by different *Ph*- gene combinations - Results from field trials and detached leaflet assay

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Late-blight disease, caused by the Oomycete pathogen *Phytophthora infestans*, is one of the main factors limiting outdoor cultivation of tomato in cool, wet climates such as Northern Europe. Whilst most commercial tomato production in Northern Europe takes place in glasshouses or polytunnels, there is a great demand for resistant cultivars for outdoor growing by amateur gardeners. Several major genes conferring resistance to *P. infestans* have been discovered in tomato, of which two, *Ph-2* and *Ph-3*, are widely used in contemporary resistance breeding programmes. Field trials of tomato cultivars and breeding lines were conducted in 2012 and 2013 as part of a tomato breeding project at Bangor University, and molecular markers were used to establish the *Ph-2* and *Ph-3* genotypes of the material in the trial. As expected, the results broadly support the commonly held view that *Ph-2* and *Ph-3* confer stronger resistance to *P. infestans* when homozygous and in combination. Additionally, the results suggest that the presence of *Ph-3* in the homozygous state confers particularly strong resistance to fruit infection. Planned experiments in Spring 2014 will use detached-leaflet tests to quantify the strength of resistance conferred by differing resistance gene combinations against several different *P. infestans* isolates. The results of these experiments will be presented.

Keywords: *Phytophthora infestans*, tomato, *Ph-2*, *Ph-3*, detached-leaflet test

T023 - Hiroshi Matsunaga

Tomato breeding for high yield and high sugar content under year-round hydroponic cultivation

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The use of hydroponic culture for tomato production has recently increased in Japan. The highest tomato yields achieved under hydroponic cultivation in Japan are 25–30 kg/m², whereas yields in Europe and North America have already reached more than 60 kg/m². Although various factors contribute to this difference, the major factor is considered to be the yield potential of the cultivars grown in each region. To improve the yield of Japanese cultivars, we created two new breeding lines, Tomato Ano Kou 8 Gou and Tomato Ano Kou 9 Gou, selected for high yield and high sugar content under hydroponic cultivation. Each line is an F₁ hybrid between inbred lines selected from the progeny of a cross between ‘Geronimo’ (De Ruiters Seeds, the Netherlands), a Dutch high-yield F₁ cultivar, and ‘Momotaro 8’ (Takii & Co. Ltd., Japan), a Japanese high-sugar-content F₁ cultivar. We performed a tomato cultivar trial under year-round hydroponic cultivation using the two new F₁ breeding lines, two Dutch high-yield F₁ cultivars and four Japanese F₁ cultivars, which include high-sugar-content cultivars. All plants were grown hydroponically with a high-wire system in a greenhouse at the NARO Institute of Vegetable and Tea Science. On 18 July 2012, seeds were sown, and on 7 August 2012, seedlings were transplanted to rockwool slabs. On 23 May 2013, plants were decapitated at two leaves above the flowering truss, and individual mature fruits were harvested once or twice a week from 5 October 2012 to 11 July 2013. To promote fruiting, a solution containing 4-chlorophenoxy acetate (Tomato Tone, Ishihara Sangyo Kaisha, Ltd., Osaka, Japan) was sprayed on the young flowering trusses. The total and commercial fruit yields per plant of the breeding lines were similar to those of the Dutch cultivars and greater than those of the Japanese cultivars. The soluble solids content of the fruit of the breeding lines was similar to that of the Japanese cultivars and greater than that of the Dutch cultivars. The fruits of the breeding lines were similar in size to those of the Dutch cultivars and larger than those of the Japanese cultivars. The breeding lines had a lower percentage of small fruits and blossom-end-rot fruits than the Japanese cultivars, but a higher percentage of cracking fruit than any of the other cultivars. These results suggest that Tomato Ano Kou 8 Gou and Tomato Ano Kou 9 Gou are promising candidates for high-yield and high-sugar-content tomato cultivars suitable for hydroponic culture. Moreover, these breeding lines are expected to improve the yield of Japanese hydroponic tomato production.

Keywords: hydroponic cultivation, yield, sugar content, year-round cultivation.

T024 - Guillaume Bauchet

GWAS reveals candidate regions for metabolic traits in tomato, *Solanum lycopersicum*

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Modern breeding can impact crop cultivars volatile fruit content, inducing low volatile profile and therefore low organoleptic quality (Klee and Tieman 2013). Genome-wide association studies have been successful in identifying genes involved in polygenic traits notably in tomato, and can be a useful way towards fruit quality improvement. Genotyping arrays (Sim, Durstewitz et al. 2012; Viquez-Zamora, Vosman et al. 2013) are available and enable GWAS for traits of interest. We present an association panel including 70 wild relative accessions (*S. pimpinellifolium*), 170 admixed accessions (*S.l. cerasiforme*) and 50 domesticated accessions (*S. lycopersicum*). GWAS analysis was conducted using single and multi-locus analysis (Yu et al., 2006; Segura et al., 2012) with 12000 SNP markers and a set of sugar-related, amino-acids as well as a broad range of volatiles. The present study is the first one in tomato reporting associations for a large set of volatiles at the genome scale. We found significant associations for 84 loci with a total of 17 traits including glucose, malate or phenylacetaldehyde. Identified loci were also concordant with previously published quantitative trait loci (guaiacol), while new loci were identified (phenylacetaldehyde). These results (1) provide a list of candidate loci to be functionally validated and (2) provide a powerful approach for finding genetic variants that can be directly used for fruit quality improvement and deciphering the genetic architecture of complex traits.

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Keywords: tomato, GWAS, metabolome, volatiles

T025 - Christophe Rothan

EMS mutants for discovery of tomato genes and allelic variants through Tilling and NGS-mapping

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Tomato is currently the model plant for fleshy fruit development and for *Solanaceae* species. Recent genomic approaches including transcriptome, proteome and metabolome analyses and genetic mapping have produced a wealth of candidate genes whose function needs to be assessed. The recent development in model and crop plants of TILLING (Targeting Local Lesions Induced IN Genomes), which reveals allelic series corresponding to several independent point mutations, and the current availability of deep sequencing tools further increase the interest of generating induced genetic diversity in tomato. We describe here the generation and use of EMS tomato mutants in the miniature cv. Micro-Tom and provide examples of the discovery by TILLING of allelic variants of target genes implicated in fruit quality traits (vitamin C, vitamin E, carotenoids). We further show that mutant collection can now be used for rapid discovery of genes underlying remarkable traits (fruit size, cuticle, color) in tomato through forward genetics approach including (i) screening mutant collection for phenotypic variation, (ii) mapping the mutation responsible for trait variation and/or (iii) identify the causal mutation by next generation sequencing (NGS) mapping.

Key words: tomato, EMS mutants, fruit, TILLING, NGS-mapping

T026 - Silvana Grandillo

RNAseq transcriptome analysis of fruit pericarp in a set of *Solanum habrochaites* LA1777 ILs

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The tomato fruit is an ideal and well-studied model system for investigation of fruit development, flavor and nutrition. Exceptional variation in these traits is found within germplasm of the tomato clade and numerous genetic, molecular and germplasm resources enable efficient research in the tomato system. Populations of marker-defined introgression lines (ILs) originating from interspecific crosses represent powerful tools to harness the genetic potential stored in unadapted germplasm for both fundamental biology studies and to improve the agricultural performance of modern crop varieties. Recent developments of 'omics' platforms have further enhanced the power of such congenic and permanent resources.

In order to gain further insights into the networks regulating key fruit metabolic pathways underlying flavor and nutritional quality, we have undertaken a systems approach that leverages natural variation stored in a new set of *Solanum habrochaites* (LA1777) ILs in the genetic background of the *S. lycopersicum* processing tomato cv. E6203. Towards this end, we have performed RNA-seq analysis from red ripe fruit pericarps of 30 *S. habrochaites* ILs. Single nucleotide polymorphisms (SNPs) and indel profiles were identified for each IL based on the RNA-Seq data. This high-density genotyping allows precise definition of the boundaries of each IL facilitating use of this resource for breeding of nutrient, flavor in addition to any other trait of interest. In several cases, the high resolution of introgression definition achieved through RNA-seq SNP analysis allowed the identification of small additional *S. habrochaites* introgressions missed in lower resolution genotyping studies based on RFLP or PCR markers. Replication of RNA-seq analysis of these and additional lines is under way and the status of this developing expression resource will be presented.

Keywords: RNAseq, tomato, introgression lines (ILs), *S. habrochaites* LA1777, fruit quality

SESSION V – GENOMICS AND SELECTION TOOLS

T027 - David Francis

Genome assisted selection for tomato fruit nutritional quality, yield components, and disease resistance emphasizes a need for multi-trait indices

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Next generation sequencing technologies and high-throughput genotyping platforms are driving crop improvement toward progressively more sophisticated and data-driven approaches. In tomato, SNP based marker density now exceeds necessary requirements for genome assisted improvement. If traditional horticultural crops are to benefit from changing technology, there will be a need to shift breeding strategies with an emphasis on the collection of objective phenotypic data. Current approaches to the design and implementation of genome wide selection (GWS) strategies leave little flexibility for selecting diverse traits with data that are not quantitative or lack a clear economic value. I will discuss the integration of tomato fruit nutritional quality into a processing tomato breeding program as an illustration of both the potential and pitfalls of GWS. High beta-carotene content is, in part, conferred by variation in the 5' untranscribed portion of the chromoplast-specific lycopene cyclase gene (*CrtL-b*) known classically as Beta (β). Alleles of β from *S. pennellii*, *S. habrochaites*, *S. chilense* and *S. galapogense* have been bred into cultivated tomato over the last fifty years. Sequencing reveals significant 5' variation and association analysis using germplasm from the SolCAP collection suggests two SNPs as candidates for the high beta-carotene phenotype. We used an accelerated backcrossing program with positive selection for β and SNPs applied to background genome selection. Alleles of β from Jaune Flamme, *S. pennellii*, and *S. galapogense* were introduced into the OH8245 genetic background. We were able to select families with ~96.5-97.8% OH8245 (equivalent to BC₄) following two rounds of backcrossing (BC₂). Field evaluation in multi-location trials suggests that alleles of β differentially modulate the levels of beta-carotene in the fruit, providing genetic variation to customize products. Genome assisted breeding has provided tools for rapid selection of a single locus. Integrating these alleles for improved nutritional quality with yield and disease resistance using approaches developed for dairy cattle and grain crops may prove problematic as the horticultural industry rarely values traits on a uniform scale and may not collect objective data for relevant traits. The development and measurement of multi-trait selection indices that match market needs is therefore a priority if the promise of genomics is to be realized.

Keywords: *Solanum lycopersicum*, genomics, plant breeding

T028 - Laura Pascual-Bañuls

Deciphering the genetic control of quantitative traits with magic

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Identification of the polymorphism controlling the variation of quantitative traits is still a challenge for plant geneticists. Traditionally, linkage mapping in bi-parental populations or association studies is used for quantitative trait locus (QTL) mapping. However, the results might be limited by the low variation between two parental lines, and by population structure in association studies. These limitations may be overcome by more complex experimental populations. To increase the number of recombinations the development of advanced intercross lines (AILs) was proposed. To increase the genetic variation analyzed, nested association mapping (NAM) populations were developed from a diverse set of parental lines that are crossed with a reference line. However, such populations do not allow the QTL analysis in different genetic backgrounds. To overcome this limitation AILs methodology was extended to multiple parent populations. Multi-parent advanced generation inter-cross (MAGIC) populations allow the screening of a wider genetic diversity by mixing the genomes of a greater number of parental lines and increase the precision of QTL mapping by including more rounds of informative meiosis. Following this assumption, a MAGIC population consisting in 1000 progenies developed from 8 founder lines should be able to map QTLs with a sub-centimorgan resolution.

In this study, we present the construction of the first tomato MAGIC population. The population (397 lines) was obtained by crossing 8 tomato lines, selected to include a wide range of genetic diversity of the species. These eight founder lines have been deeply characterized following a systems biology approach (Pascual *et al.* 2013) and their whole genomes were resequenced allowing the identification of more than 4 millions SNPs (Causse *et al.* 2013). Polymorphism information was employed to select a subset of 1486 markers specially designed to analyse the MAGIC population. The selected markers were employed to develop the first intra-specific saturated map in tomato. The linkage map covered 2156 cM, and showed an 87% increase in recombination frequencies compared to bi-parental populations. The map allowed the prediction of the haplotype origin for an average of 89% of the MAGIC line genomes. The MAGIC population was characterized under greenhouse conditions and QTLs related with fruit quality traits were detected. We demonstrated the power and precision of the MAGIC population for QTL mapping on fruit weight. We then highlighted the power of such population when coupled with whole genome sequencing to restrict the number of putative causal polymorphisms underlying the QTLs and conduct positional cloning. A strategy to fine map QTLs and identify the causal polymorphisms is proposed. In conclusion, we have established a population of genetically diverse lines that could be analyzed for any quantitative trait under different environments, providing a fine resource to analyze the basis of phenotypic plasticity.

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Keywords: MAGIC, QTL, SNPs, fruit quality, mapping

T029 - Akio Ohyama

QTL analysis in recombinant inbred lines derived from a cross of two F₁ hybrids of tomato

Ohyama, A. (1), Hayashi, T. (2), Matsunaga, H. (1), Iwata, H. (3), Negoro, S. (1), Miyatake, K. (1), Yamaguchi, H. (1), Nunome, T. (1), and Fukuoka, H. (1)

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Tomato breeding for the fresh produce market in Japan has focused on high quality (e.g., high sugar content), but the yields of Japanese cultivars are generally lower than those of high-yield cultivars developed for mass-production hydroponic culture systems in Europe. Furthermore, unusual weather conditions, such as high temperature, are making it difficult for Japanese producers to obtain a steady yield of fruit. Therefore, improvements in the yield as well as the quality are important themes in tomato production and breeding in Japan. To identify the genetic factors that contribute to the yield and quality of tomato, we selected two F₁ cultivars as parents and crossed them to establish the experimental populations. One parent was the Japanese F₁ cultivar 'Momotaro 8' (Takii & Company, Ltd., Japan), which is characterized by high sugar content, and the other was the high-yield Dutch F₁ cultivar 'Geronimo' (De Ruiters Seeds, the Netherlands). This cross corresponded to a four-way cross, as the four parental lines of the two F₁ cultivars were considered to be the founders. We generated an F₁ population consisting of 240 plants by crossing the two cultivars, from which generations were advanced by repeated cycles of selfing with single-seed descent to generate recombinant inbred lines (RILs). More than 200 of RILs were obtained and used for QTL mapping of various agricultural traits related to yield and quality. A linkage map consisting 12 linkage groups was constructed by genotypes of RILs using 197 genome-wide simple sequence repeat (SSR) markers. In QTL mapping, haplotype analysis was performed at F₁ generation since genotypes of founders of the parental cultivars were unknown. In parallel, virtual markers (putative QTLs) were located evenly spaced (e.g. 5cM) apart on the whole genome and then significant QTLs were identified by the Bayesian estimation for the marker effects. As a consequence, we found various QTLs for those traits, including soluble solids content and single fruit fresh weight. The program used for QTL mapping can also predict genetic breeding values (GBVs) of RILs. The present results will be helpful for the genetic improvement of both Japanese and European tomato cultivars.

Acknowledgements. This work was supported by grants from the Ministry of Agriculture, Forestry and Fisheries of Japan (Genomics for Agricultural Innovation, SGE-1002; Genomics-based Technology for Agricultural Improvement, NGB-2005).

Keywords: four-way cross, genetic breeding value, QTL analysis, RILs, SSR markers

LIST OF POSTERS

SESSION I : MANAGEMENT, DIVERSITY AND CHARACTERIZATION OF GENETIC RESOURCES

P001 - Reddy Aparna

Population genomics of wild tomatoes: Evolutionary inference from NGS

Aparna, R. (1)

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We are studying the evolution and natural history of two species of wild tomato: *Solanum peruvianum* and *S.chilense*. These closely related sibling species are obligate outcrossers and endemic to the west coast of Peru and Chile. Previous population genetic analyses based on a handful of genes indicate that these species have diverged from one another extremely recently, although strong barriers to interbreeding between these species exist. Due to the recentness of their divergence, these species show high levels of shared polymorphism - genetically they are almost impossible to distinguish, although morphologically they can be distinguished easily.

In our study, we have combined the power (and comfort) of next generation sequencing, with the abundant genomic information available for cultivated tomatoes and close relatives. Multiple individuals from each host species were sampled from different locations across the species' range and a single individual per location was sequenced. Total RNA from young leaves was extracted and prepared for sequencing. Each paired-end library is comprised of a minimum of 40 million reads and these reads were mapped onto the genome of the cultivated tomato.

The large number of SNP markers provides the foundation for ongoing analyses to determine the time of speciation between these species, the size of the populations, the amount of gene flow between populations and between species, and how the genetic variation is distributed on a geographic scale. Further long-term goals are to investigate the strength and targets of natural selection in the tomato genome. However, such analyses must rest upon a solid understand of population demography, as set out as the main goal of this project.

Keywords: evolution, divergence, polymorphism, speciation, interbreeding

P002 - Maria Ercolano

Omics approaches for the identification of genes underlying relevant quality traits in San Marzano and Vesuviano tomato

D'Esposito, D. (1), Ferriello, F. (1), Sacco A. (1), Dal Molin A. (2), Venturini L. (2), Ferrarini, A (2). Barone, A. (2), Delle Donne M. (2), Frusciante L. (1), Ercolano M.R. (1)

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Tomato is one of the most economically important vegetable crops worldwide. Great diversification of the crop has taken place in Italy giving rise to a rich collection of varietal types. The "Campania" is one of Italian regions with the greatest biodiversity in traditional tomato varieties. In particular, San Marzano and Vesuviano varieties, showing high fruit quality, were adapted to local conditions by an extensive selection work performed by farmers. Owing to their economic and social importance is crucial to understand the genetic basis of their unique traits. Vesuviano and San Marzano genomes were sequenced at 40x genome coverage using the Illumina platform. Insertions, deletions and SNP variants were carefully measured. Distinctive variations in key genes and transcription/regulation factors related to fruit quality have been revealed for both cultivars. In an attempt to further understand if the tomato fruit quality is related to its local habitat, the transcriptome sequencing of San Marzano, Vesuviano and Heinz1706 fruits, sampled in two localities (Acerra and Sarno), was also performed. The transcriptome of each variety was reconstructed on the basis of the respective genome. The analysis of differentially expressed genes between the two localities within each genotypes (Acerra vs. Sarno) resulted in 71, 338, 160 genes uniquely up-regulated in Heinz1706, San Marzano and Vesuviano, respectively and 153, 91 and 324 genes specifically down-regulated in Heinz1706, San Marzano and Vesuviano, respectively. The GO (GeneOntology) enrichment analysis carried out on the up-regulated genes, for the three comparisons above reported, showed 9 and 1 significant GO categories uniquely enriched in San Marzano and Vesuviano. No significant GO categories were identified specifically enriched in Heinz1706. The same analysis performed on the down-regulated genes showed 5, 1 and 3 significant GO categories uniquely enriched in Heinz1706, San Marzano and Vesuviano, respectively. These results indicate that different patterns of gene expression exist between the two localities in the different genotypes examined. A deep analysis of the members of the enriched GO categories as well as a further investigation of differentially expressed genes with respect to the pathways they belong is in progress. The effort performed will help to dissect the path from sequence variants to phenotype.

Keywords: differential gene expression, fruit quality, gene variants, NGS sequencing, tomato transcriptome

P003 - Jaime Prohens

Characterization of Solanum lycopersicum var. cerasiforme accessions from Bolivia

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Solanum lycopersicum var. *cerasiforme* is commonly found in weedy or semi wild state in the Eastern Andean slopes (Rick and Holle, 1990). One of the countries in which *Solanum lycopersicum* var. *cerasiforme* is commonly found is Bolivia. However, compared to other countries of the region like Peru or Ecuador, there have been fewer studies on the diversity of Bolivian *Solanum lycopersicum* var. *cerasiforme*. We performed a morphological characterization of fruits of 79 accessions of *S. lycopersicum* var. *cerasiforme* from Bolivia using 34 IPGRI (1996) descriptors. A wide diversity was observed for most traits, in particular for fruit shape. Seven out of the eight IPGRI (1996) descriptor categories (with the exception of Heart-shaped) for the predominant fruit shape were found in the collection. The most common shapes were Slightly flattened (34) and Rounded (23), followed by Flattened (9), High rounded (4), Cylindrical (4), Pyriform (3), and Slightly flattened (2). This suggests that genetic differences for the most relevant fruit shape genes must exist in the collection (Rodríguez et al., 2011). Most of the accessions (62) produced red fruits, but also an important number of accessions (15) had yellow fruits; two other accessions had pink fruits. Fruit weight was also very variable with an average fruit weight of 11.7 g and a range from 2.3 g to 37.0 g. A multivariate principal components analysis separates accessions basically according to their fruit size (positive correlation with the first component), and to the number of locules (positive correlation with the second component) and intensity of exterior colour and presence of green shoulders (negative correlation with the second component). The results show that Bolivian *Solanum lycopersicon* var. *cerasiforme* presents a wide morphological diversity and therefore represent a valuable genetic resource for tomato breeding. Our results may also support the hypothesis of Rodríguez et al. (2011) that some of the most relevant fruit shape mutations arose prior to domestication. Further molecular diversity studies in this collection will provide complementary information.

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Keywords: descriptors, diversity, fruit shape, morphology, *Solanum lycopersicon* var. *cerasiforme*

P004 - Tatiana Tereshonkova

Appearance of an attribute "semideterminate growth habit" in depends on genotype and growing conditions

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Semideterminate growth habit in tomato is a special case of sp (self-pruning) gene expression and manifests itself as a quantitative trait. We studied 36 hybrids under conditions of Moscow (central region) and Rostov- on-Don (southern Russia) and revealed a variation of the attribute "type of growth". In Rostov 8 hybrids showed themselves as semideterminate while in Moscow, as a determinate. Semideterminate hybrids have a tendency to increase the number of inflorescence on the main stem in condition of high density planting. Analysis of the results of crosses (top -cross method) revealed that semideterminate growth habit manifested in 5 variants of combination of parental lines that different on the "growth habit".

Keywords: semi-determinate growth habit, growing conditions

SESSION II : GENETIC BASIS FOR SELECTION AND ADAPTATION TO ABIOTIC STRESS

P005 - Diederik Smilde

Marker assisted DUS testing for disease resistance. Case study about Verticillium resistance

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Naktuinbouw is a major player for the official registration of tomato varieties for listing and Plant Breeder's Rights (PBR). Every year, Naktuinbouw performs several hundreds of trials for distinctness, uniformity and stability (DUS), as defined by CPVO and UPOV. Listing of varieties is obligatory, and PBR help breeders to protect their varieties. The possibilities for supporting the official registration of tomato varieties by developing molecular markers for specific cloned resistance genes are ever increasing. Naktuinbouw is actively searching the most suitable platform for implementation of molecular markers. In the course of this work, we sometimes encounter difficulties that could be interesting for a wider audience. We will present a case study of *Verticillium* resistance in tomato varieties in relation to DNA markers for Ve1 and Ve2.

Keywords: tomato, *Verticillium*, markers, DUS

P006 - Julie Ripoll

Promote adaptation to drought and fruit quality by alternating increased intensity of deficit irrigation and recovery period

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Under natural conditions, plants experience repeated cycles of water deficit and recovery period, which could negatively impact on plant growth and yield. However, deficit irrigation is known to improve fruit quality under certain conditions of application. Understanding the genetic variability in plant response and adaptation to such conditions is crucial for breeders. The 8 parents of the Multi-Parent Advanced Generation Inter-Cross population of tomato, with the largest genetic variability observed in tomato, were selected for this study. Three 15-day periods of water deficit (-38%, -45% and -55% of water supply compared to the well-irrigated control) interrupted by two 15-day periods of recovery, were applied from the reproductive stage. Stress intensity applied to each genotype was assessed based on the predawn water potential. Adaptation to drought was evaluated by plant growth (estimated leaf surface area), fruit size and weight, and fruit quality (appraised by soluble sugar and organic acid contents). According to the predawn water potentials, significant differences of stress perception among genotypes appeared only during the third period of water deficit. Plant growth and fruit caliber were not significantly affected, although slightly reduced, by the alternating treatments. Fruits were divided into two batches according to the stage of development (cell division, cell expansion and ripening) affected by treatments. Results showed a significant increase in sucrose and low increase in glucose and fructose in fruits as well as a decrease in citric acid regardless of the fruit development stage affected. On their whole, results indicated that the alternation between deficit irrigation and recovery periods permit to maintain plant growth and fruit caliber and to slightly improve fruit taste.

Keywords: *S. lycopersicum*, Magic population, water deficit, recovery, fruit quality

P007 – Rebecca Stevens

Regulation of tomato ascorbate levels: a candidate gene involved in fruit nutritional value and stress tolerance

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Ascorbate is a powerful antioxidant in plants, and its levels are an important quality criterion in commercial species. Factors influencing these levels include environmental variations, particularly light, and the genetic control of its biosynthesis, recycling and degradation. Our previous work has identified QTL for fruit vitamin C content and a candidate gene, encoding a monodehydroascorbate reductase (MDHAR) isoform, an enzyme catalysing reduction of the oxidized radical of ascorbate, monodehydroascorbate, to ascorbate. The activity of this enzyme is also correlated with fruit tolerance to post-harvest chilling stress in introgression lines containing the vitamin C QTL. In order to understand the role of this gene in fruit physiology and metabolism we produced transgenic lines in different genetic backgrounds. We show that the isoform encodes a cytosolic and peroxisomal MDHAR, and that this enzyme negatively regulates ascorbate levels in certain genotypes of *Solanum lycopersicum* (tomato). Transgenic lines overexpressing MDHAR show a decrease in ascorbate levels in leaves, whereas lines where MDHAR is silenced show an increase in these levels in both fruit and leaves. Furthermore, the intensity of these differences is light-dependent. The unexpected effect of this MDHAR on ascorbate levels cannot be explained by changes in the expression of Smirnoff-Wheeler pathway genes, or the activity of enzymes involved in degradation (ascorbate peroxidase) or recycling of ascorbate (dehydroascorbate reductase and glutathione reductase) suggesting a previously unidentified mechanism regulating ascorbate levels. The impact of under-expression of this gene in processing tomato on fruit post-harvest physiology has also been examined.

Keywords: ascorbate, post-harvest, QTL, redox

P008 - Svetlana Ignatova

Development and use of abiotic stress tolerance coefficient in tomato breeding

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At present the problem of tomato tolerance to drought, heat, cold and high salt concentration in the solutions used in small-scale production technologies remains highly topical. Over the last 5-7 years Russia and Europe have experienced temperature perturbations. This study used methods of rapid evaluation of the breeding material at various ontogenetic stages: evaluation of seeds and evaluation of the mature pollen. In both cases the evaluation was made with respect to two parameters: the number of germinated seeds or pollen grains and the length of the seedlings. We combined those parameters into one value (K_{str}) and applied a ratio, which characterizes stress tolerance of any variety. $K_{str} = (A \times B - a \times b) / (A \times B)$, where A is the length of seedlings in the control group, B is the number of seedlings used in control, a is the length of seedlings in the experimental group, b is the number of seedlings used in the experiment.

Evaluation of tolerance to low (6°C and 10°C) and high (35°C, 45°C and 53°C) temperatures was made following heating of the mature pollen and seedlings. The control temperature was 25°C. It was found that tolerance to low and high temperatures is inherited independently and is regulated by one gene or a block of genes in each specific case. We have identified groups tolerant to either low or high temperatures or to both low and high temperatures or intolerant either to one or other factor. Based on the groups identified we developed F1 hybrids, which demonstrated high tolerance to temperature stresses in the 2010 extreme weather conditions. During 2.5 months evaluation that year (from 15th June to 30th August) the greenhouse temperature range was 35°C to 50°C during the day and 25 °C and above at night. Under such extreme conditions male and female gametophytes were formed, so heat resistance came into full play. Only 8 % of 140 hybrids demonstrated 100 % fruit formation, 26.1 % generated 80 % of fruit and formed a group of heat resistant hybrids. One of them (F1 Forsage) was proven to have high heat resistance in greenhouses in the south of Russia: as of 01.09.2011, the crop was 52.4 kg/m², which is 36.7 % higher than the established standard or crop of other hybrids used. In June through August the crop was 11.2 - 10.5 - 9.5 kg/m² respectively.

Tolerance to physiological drought and salt stress was assessed following seed germination in the 7.4-8.8 % solution of polyethylene glycol 6000 (PEG 6000) and 0.80-0.88 % solution of NaCl. We determined the best concentrations and identified the best hybrids. Percentage of hybrids tolerant to both factors was found to be 65%. As for stress tolerance ratio of the best hybrids, it has a negative value. Results of the tomato hybrids evaluation, which were obtained using differential concentrations (PEG 6000, 8.2%, and NaCl, 0.83%), demonstrated high correlation with each other ($r = 0.83$).

Keywords: hybrids, stress, high and low temperature, salt stress, physiological drought, tomato

SESSION III : GENETIC BASIS AND SELECTION FOR RESISTANCE TO DISEASES

P009 - James Stroud

Exploring Phytophthora infestans diversity on tomato in Great Britain. Evidence of host specialisation.

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Late Blight disease, caused by the oomycete *Phytophthora infestans*, is one of the most economically important diseases of tomato and potato. *P. infestans* normally reproduces asexually and populations frequently include many clonally reproducing genotypes. Evidence exists that genotypes differ in their virulence or aggressiveness towards different host species and the frequency of *P. infestans* genotypes varies from one host species to another in many areas. Whilst the genotypic composition of *P. infestans* populations on potato has been well studied in Great Britain, this is not the case with *P. infestans* populations on tomato. This study attempted to determine whether a subset of tomato-adapted *P. infestans* genotypes exists in Great Britain by collecting *P. infestans* samples from throughout the country from 2011 to 2013 and genotyping them using SSR markers. Samples collected in 2011 and 2012 suggest that genotype 23_A1, which is rarely found on potato crops, is far more common on tomato, whilst genotypes 13_A2 and 6_A1, (which made up most of the samples collected from potato crops in recent years) were much less common on tomato. 23_A1 was absent from the 2013 sample, but even so, clear differences exist between the potato and tomato populations, with a higher diversity of unique isolates on tomato. Understanding the genotypic composition of *P. infestans* populations will help breeders to develop tomato cultivars with resistance to Late Blight.

Keywords: *Phytophthora infestans*, late-blight, tomato, potato, Britain

P010 - Liliana Stamova

Screening for resistance to Phytophthora capsici in tomatoes.

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Two decades ago *Phytophthora* Root Rot was reported to be a major disease on tomatoes in California. The disease is caused by *Phytophthora capsici* and *Phytophthora parasitica*. In 2011 *Phytophthora capsici* infection was found in several tomato fields. This disease is expected to become a growing problem in tomato crops in California in the future. Until now no commercial tomato cultivars resistant to *Phytophthora capsici* have been reported. We present the results from screening eighty cultivated type tomato lines for resistance to a California isolate of the pathogen. The lines have different wild species (*S. peruvianum*, *S. chilense*, *S.habrochaites* and *S.pennellii*) in their pedigree. Twenty lines represent complex crosses involving two wild species. Three different methods of inoculation were used: 1) Trays of seedlings at the two-leaf-stage with holes in the bottom were placed on trays (without holes) containing one liter of inoculum of *P. capsici*; 2) Plants with 2-4 leaves were dipped for 10 min. in inoculum; and 3) The inoculum was applied in furrows made between rows of seedlings grown in trays. Ten days after inoculation root and stem symptoms were visually rated on a scale of 0 – 4, where 0 = no symptoms and 4 = seedlings killed. The results revealed the existence of different levels of resistance in the genetic materials screened. Twenty lines showed susceptibility equal to that of the susceptible control - leaves wilted almost to the very top of the plant, root systems with brown-colored lesions and no secondary root generation. Forty lines segregated with different ratios of resistant (high or moderate) and susceptible plants. Twenty lines showed very high levels of resistance (rated 0 or 1) when challenged with the California isolate. One month after inoculation the plants were vigorous, with healthy, well developed roots without brown lesions. The level of resistance of the best lines was much higher than that described by H. Bolkan (1985) as “an ability of the genotype to generate additional roots”. Results obtained after screening F1 and F2 generations of the cross (Ph.c.25 x MoneyMaker) suggest the action of a single dominant gene responsible for resistance to *P. capsici*. Development of cultivars resistant to *P. capsici* will be a valuable contribution for tomato growers.

Keywords: tomato, resistance, *Phytophthora capsici*

P011 - Liliana Stamova

Additional sources for resistance for resistance to Verticillium dahliae race-2 and Pyrenochaeta lycopersici

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Both Verticillium wilt caused by *Verticillium dahliae* race 2 and Corky root evoked by *Pyrenochaeta lycopersici* are economically important tomato diseases in California. Race 2 of *Verticillium dahliae* detected as early as in 1957, is now well spread all over the world. Recently our line Veda has shown high level of resistance when challenged with isolates from California, France, Brazil and Morocco. Given the variability of the world population of the pathogen we keep searching for additional sources for resistance. Seventy progenies of interspecific crosses were screened with the California isolate SW-5, obtained from Dr. R. M. Davis (University of California, Davis). A standard root-dip method was used to inoculate the plant materials. The evaluation of disease symptoms were made weekly for the period of two month. Fifteen lines with *L.chilense* in their pedigree as well as 10 late generations coming from complex crosses with two wild species were selected for their high level of resistance to race 2 which was equal or higher to that of line Veda. With Corky root, the only gene used in the tomato breeding programs for resistance is the recessive gene *py*. Recently we reported about line Pirelly that carries a dominant factor for resistance to *Pyrenochaeta lycopersici*. The evidence of variability in the pathogen population, as well as the existence of two types –T1 and T2 of the pathogen, make searching for new resistance sources well motivated. A large number of genetic materials were inoculated with T1 and T2 California isolates obtained from Prof. L. Epstein (University of California, Davis). The evaluation took place two months after inoculation. The plants were rated based on their symptoms on the main root and with special attention to the damages of the feeding roots. We found high level of resistance in several lines having two wild species (*S. peruvianum* and *S. chilense*) in their genetic background. The lines have good fruit-setting and mid-sized fruits.

Keywords: tomato, resistance, *Vert2*, *Pyrenochaeta lycopersici*

P012 - Sophie Rommel

Resistance shaping miRNAs in wild tomatoes

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Introgression of resistance genes (*R* genes) is commonly used to introduce pathogen resistance from wild tomatoes into the cultivated tomato. Therefore, wild tomatoes play a crucial role in breeding. Identification of novel *R* genes and resistance specificities in wild tomatoes is the aim of several labs worldwide, while less focus has been given to the study of endogenous plant mechanisms controlling transcription and translation of resistance genes and how this influences pathogen resistance in the host. Many plant *R* genes encode for proteins containing nucleotide binding site-leucine rich repeat (NBS-LRR) motifs. Recently novel microRNAs (miRNAs) have been described in *Solanum lycopersicum* that contribute to *NBS-LRR* post-transcriptional regulation. In our study, we will investigate how levels of miRNA expression affect disease resistance. We will analyse the evolutionary history of the miRNA superfamily miR482/2118 that regulates *NBS-LRR* genes in the nightshade family (Solanaceae). Our preliminary analyses reveal that the gene-family size has varied over evolutionary time, but appears to be more stable within the Solanaceae as compared with other flowering plants. Using phylogenetic methods, we detected an excess of lineage-specific losses relative to gains. Currently we are investigating the rate of sequence evolution of members of this gene family in wild tomatoes and other close relatives in the Solanaceae. This will indicate how variation is distributed across the domains of the miRNA genes and which regions are potential targets of natural selection. An ultimate goal of our research is to understand the link between naturally occurring sequence variation in members of this gene family and host plant defense. Upregulation of these miRNA genes is predicted to negatively regulate their targets: *NBS-LRRs*. Therefore, host disease resistance may be perturbed through the action of these genes. An effective pathogen would specifically up-regulate host miRNA to suppress levels of *NBS-LRRs*. However, many pathogens do the opposite, and thereby potentially activate host resistance inadvertently. Using the oomycete pathogen *Phytophthora infestans* as a model, we will investigate how levels of miRNAs and *NBS-LRRs* are affected during the infection process. Our preliminary inoculations with a panel of different strains of *Solanum*-specific *P. infestans* indicate that levels of host miRNA do differs depending of which pathogen strain is used. With this system in hand, we aim to determine how levels and target specificity of these miRNA gene family members contribute to host plant resistance in wild and cultivated tomato.

Keywords: NBS-LRRs, wild tomatoes, miRNA, *Phytophthora infestans*

P013: Tatiana Tereshonkova

Tomato powdery mildew: maintaining the pathogen (Oidium neolycopersici) on plants in vitro

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Pathogen of a dangerous tomato disease in greenhouses - *Oidium neolycopersici* is an obligate parasite and it cannot grow on artificial media. The very important moment in tomato breeding for resistance to powdery mildew is to save different strains of pathogens in the offseason. The purpose of research was to explore the possibility of storage and cultivation of different isolates of the fungus on tomato plants *in vitro*. As a result, we picked up the artificial media, the composition of hormones, methods of decontamination and infection of plants, sterilization and cultivation mode. There were many problems with decontamination of infected plant before transferring to the artificial media. To obtain the sterile explants with symptoms of powdery mildew of tomato *in vitro*, the segments of plants infected with powdery mildew *in vivo*, should be rinsed with distilled water and transferred to a nutrient medium. After development of powdery mildew we take conidia from them and inoculate new sterile explants till obtaining the plants with symptoms of powdery mildew without signs of other infections. We can maintain fungus *in vitro* during 1-1.5 month by such method.

Keywords: *Oidium neolycopersici*, plants *in vitro*, artificial inoculation

SESSION IV : IMPROVING FRUIT QUALITY

P014 - Bistra Atanassova

Antioxidant capacity of tomato genotypes differing in fruit colour

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The present study was designed to investigate the antioxidant capacity of ripe tomato fruits possessing different colors. Six tomato genotypes were included in the study: 1. L. At 16 homozygous for gene *Anthocyanin fruit (Aft)*, red fruits, lycopene content 5.7 mg%, b-carotene content 0.85 mg%, reducing sugars content 3.4 mg%; 2. Rozalina Rossa F1 – pink fruits, lycopene content 4.8 mg%, b-carotene content 0.98 mg%, reducing sugars content 4.2 mg%; 3. Reyana F1 – red fruits, lycopene content 6.0 mg%, b-carotene content 0.72 mg%, reducing sugars content 3.8 mg%; 4. Zlatista – yellow fruits, b-carotene content 0.5 mg%, reducing sugars content 5.0; 5. Paco orange – orange fruits, b-carotene content 6.0 mg%, reducing sugars content 4.3; 6. Bendida – orange fruits, b-carotene content 3.2 mg%, reducing sugars content 4.0 mg%. Three series of investigations were carried out: on fruits immediately after harvest, three and six days after fruits storage under 4°C in the dark. The following methods were used for determination of the antioxidant capacity: the Ferric reducing antioxidant power (FRAP) and Trolox equivalent antioxidant capacity (TEAC) assays after two-step procedure of processing and extraction of water-soluble and lipo-soluble antioxidants. It was found that L. At 16 (*Aft*) possessed the highest TEAC and FRAP activity (respectively expressed as micromoles of Trolox equivalents per gram and as micromoles Fe²⁺ per gram) in all three series of experiments, while Zlatista was characterized by the lowest values concerning these characters. The highest activity in FRAP assay (both water- and lipo- soluble antioxidants) was detected three days after harvest in all variants while in TEAC assay the highest activity (both water- and lipo- soluble antioxidants) was detected six days after harvest of the fruits.

Keywords: tomato, antioxidant capacity, FRAP, TEAC

P015 – Christopher Sauvage

A multilocus mixed model for GWAS reveals associations for metabolic traits in the tomato, *Solanum lycopersicum*

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Genome-wide association studies have been successful in identifying genes involved in polygenic traits notably in crops, and can be a useful way towards crop improvement. We have applied in a major crop, tomato, a recently developed multi-locus mixed model as a general method for mapping complex traits in structured populations (Segura et al., 2012). Among major crops worldwide, tomato (*Solanum lycopersicum*) is a highly valuable fruit with excellent nutritional value (Causse et al., 2010). SNP beadchips (Hamilton et al., 2012) are available and enable GWAS for traits of interest. Recently, a pilot study defined the optimal conditions for GWAS by using cherry tomato accessions (Ranc et al., 2012). In our study, we examined a core collection of 180 tomato varieties composed of 20 wild accessions (*S. pimpinellifolium*), 130 admixed accessions (*S. cerasiforme*) and 30 domesticated accessions (*S. lycopersicum*). Multi-locus GWAS analysis was conducted using the MLMM package (Segura et al., 2012) with 7700 SNP markers and a set of sugar-related, vitamin C-related and morphological traits as well as a broad range of metabolites involved in central carbon metabolism. The present study is the first one in tomato reporting associations for a large set of traits at the genome scale. We found significant associations for 89 loci with a total of 19 traits including fresh weight, sucrose, ascorbate, malate or citrate notably. Identified loci were also concordant with published quantitative trait loci (eg malate), while new loci were identified (for tocopherol). Moreover, several related metabolites, such as citrate and malate (both involved in the Krebs cycle) displayed two identical associations. These results (1) provide a list of candidate loci to be functionally validated and (2) provide a powerful analytical approach for finding genetic variants that can be directly used for crop improvement and deciphering the genetic architecture of complex traits.

P016 – David Liscombe

Breeding tastier tomatoes-on-the-vine for Canadian greenhouse growers

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Greenhouse tomato production is a major industry in Canada, worth approximately CDN \$500M (€330M) annually at the farm gate. More than half of the Canadian greenhouse tomatoes are grown in Southern Ontario, a region that experiences hot, humid summers and cold winters with short days. Despite these unique growing conditions, tomatoes-on-the-vine (TOV) varieties currently grown in Canadian greenhouses have been developed outside of Canada, often in regions with very different climates. Growing conditions can have significant effects on tomato flavour by modulating the biosynthesis and accumulation of sugars, acids, and more than 30 volatile chemicals that ultimately determine the organoleptic quality of the fruit. Vineland has partnered with the Ontario Greenhouse Vegetable Growers (OGVG) to establish a breeding program in Ontario focused on developing more flavourful, locally-adapted TOV varieties. This year, we evaluated 368 varieties comprising old and current hybrids as well as heirloom types. Genetic relatedness was determined by genotyping-by-sequencing, and flavour-related chemicals including 38 volatiles, sugars (glucose, fructose) and acids (citrate, malate) were quantified to select for sources of genetic diversity and desirable flavour chemistry. Sensory evaluation and consumer profiling will be integrated with genetic and chemical data to guide the breeding program toward tastier TOVs.

P017 – Lucie Fernandez

Micro-tom mutants for identification of target genes controlling fruit growth in tomato

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Mechanisms involved in fruit size/growth are difficult to unravel because of complex interactions between cell specification, polarity, asymmetric division, rearrangement and growth. In the recent years, genomic approaches including transcriptome, proteome and metabolome analyses have produced a wealth of candidate genes possibly involved in the control of biological processes or traits of interest, such as fruit size. Following their discovery, the relationships between the candidate genes identified and the processes or traits of interest must be confirmed, and gene function needs to be assessed. Forward genetics appears as the most powerful approach for the identification of new gene functions and mutant collections offer invaluable resources for discovering new phenotypes and new allelic variants. Thanks to the recent availability of tomato genomic sequence and the current availability of deep sequencing tools, linking genotypic variations to associated phenotypic changes is now more accessible.

We generated highly-mutagenized tomato EMS mutant resources using the miniature tomato cultivar Micro-Tom for genetics approaches in tomato. This collection has been used, in part, for identifying novel fruit size mutants in tomato and, last, give some hints on how current deep sequencing technologies may be used for identifying unknown mutations responsible for phenotypic changes in Micro-Tom mutants. Towards this end, a Micro-Tom EMS mutant collection of 3500 phenotyped lines available at INRA Bordeaux was screened for fruit size (small or large fruit) and pericarp thickness (thick or thin) mutants. Among these, 36 mutant lines were selected (21 fruit size mutants, 15 pericarp thickness mutants). Following confirmation of the phenotype, 15 mutant lines were submitted to detailed fruit developmental analysis in view of identifying fruit growth processes altered in these mutants. The results illustrate how screening mutant collections, which is relatively straightforward once the initial characterization of the mutant collection and the construction of the database have been done, can successfully contribute to the isolation of a large number of new fruit size/growth mutants and thus provide new genetic material for deciphering the mechanisms involved in the control of fruit growth and size.

Keywords: fruit growth, tomato, EMS mutant, forward genetics

P018 - Raphaël Tisiot

Heirloom tomatoes. Characterization of new varieties

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Within the diversified market segments of tomato, heirloom types are expanding. The Ctifl set up characterization trials of this market segment aiming to assess the new varieties of this type present on the market. Therefore 11 varieties of the Marmande, Coeur de Boeuf and Albenga types grown in the south were studied according to their agronomic, physico-chemical and sensoriel qualities. The assessments were carried out in April and June allowing the changes in qualitative potential of these products to be characterized. This trial has revealed high variability within the heirloom segment, essentially linked to the different subtypes.

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Keywords: heirloom tomatoes, sensorial qualities

P019 - Sanders Junglee

Mercury induced water deficit stimulates antioxidant defence systems in tomato fruits (*Solanum lycopersicum* L., cv. Microtom) through nitric oxide, abscissic acid and hydrogen peroxide

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Drought is one of the most important environmental factors that influences fruit growth and nutritional quality. Indeed, decreased water availability has an immediate impact on water status and affects fruit growth and maturation via negative effects on water absorption, photosynthesis and the metabolism of the fruit. It is accepted that antioxidant system plays an important role in the development and the maturation of tomato fruit. Thus, water stress could interfere with the antioxidant status of fruit depending when the stress occurred. The effects of different levels of water deficit on oxidative parameters, ascorbate (AsA) content and activities of antioxidant enzymes were studied in the fruit of tomato (*Solanum lycopersicum* L. cv. Micro-Tom). Plants bearing fruits at different developmental stages were subjected to different levels of water deficit. Changes in H₂O₂ and MDA contents indicated that water shortage induced oxidative stress in fruits. The concentrations of AsA and dehydroascorbate were generally modified with water stress treatments. Moreover, evolution in SOD and CAT activities and DHAR, MDHAR, APX and GR activities were dependent on the fruit age and water stress intensity. In addition, epifluorescence microscopy using NO₅₅₀ probe revealed overproduction of the signalling molecule, nitric oxide (NO), in fruits subjected to water deficit. Taken together our results suggest that water deficit positively influences fruit quality through the modification of transcriptomic profile and the activation of antioxidant systems through NO.

Keywords: water deficit, oxidative stress, antioxidant enzymes, nitric oxide

SESSION V : GENOMICS AND SELECTION TOOLS

P020 - Cécile Bres

TILLING identification of point mutations in target genes using a tomato EMS mutant population in the miniature cultivar Micro-Tom

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Tomato is currently the model species for fleshy fruit development and for *Solanaceae* species. The recent completion of a high quality genome sequence of the inbred tomato (*Solanum lycopersicum*) cultivar 'Heinz 1706' allowed the prediction and *in silico* annotation of ca 35000 genes whose functions are in most cases still unknown. Assigning a biological function to these genes is among the priorities of the tomato community, especially for genes contributing to fleshy fruit development and quality, and to other major agronomical traits in tomato and *Solanaceae*.

Reverse genetics approaches aiming at defining the function of a candidate gene are mostly based in model plant species on the use of insertional mutant populations. Until recently, large scale analysis of the biological function of candidate genes in tomato using this strategy has been hardly feasible, due to the large number of mutants required and to the limitations of genetic transformation system in tomato. Alternative approach well-adapted to tomato has been the development of EMS mutant populations and detection of unknown mutations in target genes through the Targeting Induced Local Lesions In Genomes (TILLING) technology. TILLING combines the induction of a high number of random mutations in tomato genome with mutagens like ethylmethane sulfonate (EMS) and the use of high throughput screening systems to discover induced lesions in DNA target sequences.

We have developed at INRA Bordeaux a tomato population of 8000 EMS-highly mutagenized lines in the miniature cultivar 'Micro-Tom' and used TILLING for the identification of mutations in genes of interest. Classical TILLING involves the screening of DNA samples using a mismatch-specific endonuclease that cleaves only those PCR products that carry a mutation. Infrared-based LI-COR system is then used to visualise the mutations in any gene of interest. To date, we have used TILLING to identify 330 mutations in 33 different genes in our Micro-Tom EMS mutant population. This reflects a mutation frequency ranging from 1 mutation/160 kb to 1 mutation/1460 kb (depending on the locus) with an average of 1 confirmed mutation/523 kb. We are currently evaluating the potential use of the powerful next-generation sequencing technology for the identification of EMS mutations in our Micro-Tom EMS mutant population.

Keywords: reverse genetics, TILLING, tomato

P021 – Daniel Chambonnet

In situ induction of haploid gynogenesis in tomato

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Tomato (*S. lycopersicum*) gynogenesis can be induced by allo-pollination with *Solanum sisymbriifolium*. One day after pollination, microscope observations under fluorescence show that *S. sisymbriifolium* pollen is unable to germinate and to fertilize tomato ovules. However parthenocarpic fruits develop and four weeks after pollination, these fruits contain albumen-deprived seeds that include variable proportions of embryos of very particular shapes, globular, heart-shaped, torpedo-shaped, or very hairy. Generally of a small size, these induced embryos are located in the central part of the seeds whereas control zygotic embryos of the same age, of a much bigger size, are located along the longest seed curvature side, their milky albumen occupying most of the seed volume. The globular, heart-shaped and torpedo-shaped induced embryos are unable to develop into plantlets when cultivated *in vitro* on various synthetic media. On the other hand, when transferred from their albumen-deprived seed into the albumen of a nurse seed manually deprived of its own zygotic embryo, most if not all of the induced embryos develop into plantlets, displaying abnormalities or developing slower than zygotic embryos. All macroscopic observations are consistent with a haploid origin of the plantlets obtained.

SESSION VI : MISCELLANEOUS

P022 - Aleksandra Lewandowska

The role of red and far red light in the regulation of storage material degradation in germinating tomato seeds

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Starch is the main storage material in plants and its degradation in seeds is one of the key metabolic processes, determining the course of germination and seedling development. Until now, not much information has been gathered about storage starch degradation pathways in dicotyledonous seeds. Numerous plant species also accumulate lipids apart from starch. In many seeds triacylglycerols are the main storage material. However, in order to become a source of energy for the growing seedling, fat must be converted into carbohydrates via the glyoxylate cycle. Fat degradation has been studied mostly in oil-storing seeds and information about the interplay between starch and lipid metabolism in seeds containing both types of storage material is very scarce. The aim of this study was to analyze the degradation of starch and lipids in germinating wild type tomato (*Solanum lycopersicum*) seeds, which contain both types of storage materials, and to check whether these processes are regulated by light via phytochromes. Starch and lipids were quantified using biochemical assays in germinating seeds and seedlings during the first 5 days of growth. The distribution of storage materials in different seedling parts (root, hypocotyl, cotyledon, seed coat) was also determined after 5 days of growth. In order to investigate the process of fat-carbohydrate transformation, the levels of sucrose and the activities of glyoxylate cycle enzymes were measured. It is well known that the germination of many plant species, including tomato, is regulated by phytochromes, photoreceptors sensitive to red and far-red light. Phytochromes also control many other physiological processes, acting through several different pathways. To investigate the role of phytochromes in storage material degradation, imbibed seeds were subjected to red or far red light treatment, which respectively induces or inhibits germination. Seeds germinating in darkness were used as a control in these experiments. The results confirm that far red light inhibits tomato seed germination, while consecutive irradiation with red light overcomes this inhibition and induces germination. Additionally, during the first five days of growth a decrease in lipid content and increases in starch and sucrose contents have been observed. The decrease in lipid content during the 5th day suggests that they are converted to carbohydrates and used as energetic substrates later in the process. This was confirmed by the increased activity of glyoxylate cycle enzymes at this time point. The changes in metabolite content exhibit similar patterns in seeds subjected to different light treatments. This may suggest that these processes are controlled by different mechanisms than germination itself. Only in the case of starch content a significant difference between red- and far red-irradiated seedlings could be observed which may point to a role of phytochromes in storage starch metabolism.

Keywords: phytochrome, storage material degradation, glyoxylate cycle, seed germination

P023 - Tsukasa Nunonne

Map based cloning of tomato parthenocarpic fruit-2 (pat-2) gene

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The parthenocarpic fruit 2 (*pat-2*) gene of tomato is a recessive mutation conferring parthenocarpy, which is the capability of a plant to set seedless fruits in the absence of pollination and fertilization. Parthenocarpic mutants offer a useful method to regulate fruit production. To confirm the chromosomal location of the *pat-2* locus, F₂ population segregating from a cross between parthenocarpic line and tomato cultivar were grown and classified as parthenocarpic or wild-type. Through genetic analysis, we constructed linkage map based on SSR markers and the *pat-2* locus was assigned to chromosome 4, located on 6.4 cM region between 2 SSR markers. Using a total of 2,672 F₃ individuals, we refined the candidate region to a ~300kb interval. Comparing the sequence of this region between the parthenocarpic line and tomato cultivar, a 1034-base pair (bp) deletion was observed in parthenocarpic line in a predicted gene. The predicted gene is comprised of 2 exons and 1 intron, and the deletion in *pat-2* located on the first exon and intron. It is estimated that the *pat-2* gene is deficient functionally and translated into a truncated protein. We introduced a construct containing inverted repeat of the truncated *pat-2* candidate gene into non-parthenocarpic tomato cultivar via *Agrobacterium*-mediated transformation. Independent RNAi transformants showed parthenocarpy. We also introduced a construct containing the full-length *pat-2* candidate gene into parthenocarpic line. Independent OX transformants showed non-parthenocarpy. The candidate gene encodes zinc finger-homeodomain protein that functions as transcription factors.

Keywords: parthenocarpy, *pat-2*, tomato

P024 – Atilla Ata

Evaluation of some tomato lines on high temperature conditions

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Tomato is one of the most popular and widely consumed vegetables grown worldwide and it was growing broad area in the world. Tomato is sensitive to high temperature that may have adverse crop production especially 35 °C and above. In high temperatures tomatoes set less fruit and weight loss in fruit. In order to produce durable tomatoes to abiotic stress conditions, breeders aim to develop new tomato varieties in recent years. In this study, 250 indeterminate tomato genotypes planted in grow chamber. Then 90 genotypes selected and planted in open field in June in Şanlıurfa Province which one of the hottest place in summer in Turkey. It was taken amount of yield from setting of tomatoes fruits between 15 July and 15 August. It was obtained number of seeds in these fruits. As a result yield varied between 0- 5999.33 kg/plot (12 plants). The highest total yield was obtained from the Alata 372-2-3 genotypes. Latter, 24 genotypes that were tolerant to high temperature selected.

Keywords: tomato, high temperature, breeding

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CITY MAP



SUPPLEMENTARY INFORMATION

WHERE TO EAT ?

Name of the Restaurant	Budget (Main course)	Phone	Address	Booking	Comments
Chez Mimmo	12 €	04 90 82 42 73	19 rue du Chapeau Rouge (G5)	Mandatory	Pizza, Risotto, Pasta (+veggie)
Le Pili	12 €	04 90 27 39 53	34 place des Corps Saints (F7)	Advised	Pizza, Pasta (+veggie)
Macadam	15 €	04 86 19 82 91	Place des Corps Saints (F7)	Advised	Traditional (+veggie)
Chez Ginette et Marcel	15 €	04 90 85 58 70	25 place des Corps Saints (F7)	Not Necessary	Selection of slices of bread with topping (cheese, salmon)
Chez Ripert	20 €	04 90 27 37 97	28 rue Bonneterie (F5)	Advised	Traditional (+veggie)
La Vache à Carreaux	23 €	04 90 80 09 05	14 rue Peyrollerie (F4)	Advised	Every meal is cooked with cheese, nice selection of wine
Le Riad	25 €	04 90 82 10 85	17 rue Galante (E6)	Advised	Typical Moroccan meals
Chez Lulu	27 €	04 90 85 69 44	06 place des Chataignes (F4)	Mandatory	Fine Traditional (+veggie)
L'épicerie	27 €	04 90 82 74 22	10 place Saint Pierre (F5)	Mandatory	Fine Traditional (+veggie)
Au Tout Petit	27 €	04 90 82 38 86	04 rue d'Amphoux (G5)	Mandatory	Fine Traditional (+veggie)
Les 5 sens	35 €	04 90 85 26 51	18 rue Joseph Vernet (D4)	Mandatory	Fine Traditional (+veggie)

WHERE TO DRINK ?

Name of the place	Address	Comments
PubZ	58 rue Bonneterie (G6)	Beer and Rum
O'Neills	38 cours Jean-Jaurès (E7)	Irish Pub (and food)
Red Sky	Place Pie (G5)	Beer
La Cave des pas sages	41 rue des Teinturiers (H7)	Beer, Wine
Chez Marie	04 rue Louis Pasteur (H4)	Wine (and food)
Avitus	11 rue du Vieux Sextier (F5)	Fine Wine

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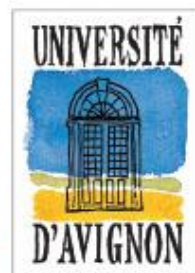
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