Flash Talk - ISPS2019-201

Does size matters? variation among two Hordeum species populations provide insights on the role of genome size in adaptation

Bar Shermeister*[1,2], Guy Atzmon[2], David Schwartz[1,2] and Sariel Hübner[1,2]

[1] Tel-Hai Academic College, Israel

[2] MIGAL Galilee Research Institute, Israel

*Corresponding Author Email: barshmeister@gmail.com

Genome size vary widely across life kingdoms, genus and species. However, it is unclear whether genome size varies also on the intraspecific level. Theoretical models predict that increased genome size could have an adaptive advantage by improving the efficiency of selection when effective population size is low. Yet, controversy evidences for intraspecific variation among plant species have been provided, thus it is unclear what is the essence of genome size variation or the underlying mechanism. To address these questions, natural populations of two Hordeum species (i.e. H. bulbosum and H. spontaneum) collected at a wide range of environments along Israel were screened for genome size variation using flow cytometry protocols. We provide evidences from over 200 genotypes, of each species, for intraspecific variation in genome size and its correlation with environmental gradients. In both species, larger genomes were observed in warm and dry habitats while smaller genomes were observed in cold and wet habitats. Our results provide an empirical support for existing theoretical models although the adaptive mechanisms regulating genome size variation should be further investigated.

Flash Talk - ISPS2019-202

Redundancy in Tomato Gibberellin Receptors Contributes to Phenotypic Stability under Changing Environments

Natanella Eliaz*[1], Hagai Shohat[1], Shula Blum[1], Sivan Livne[1], Dvir Mendelson[1] and David Weiss[1]

[1] Institute of Plant Sciences and Genetics in Agriculture, The Robert H. Smith Faculty of Agriculture, Food, and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

*Corresponding Author Email: david.weiss@mail.huji.ac.il

The pleiotropic and complex gibberellin (GA) response relies on targeted proteolysis of DELLA proteins by a GA-activated GIBBERELLIN-INSENSITIVE DWARF1 (GID1) receptor. The tomato (*Solanum lycopersicum*) genome encodes for a single DELLA protein, PROCERA (PRO), and three receptors, SlGID1a (GID1a), GID1b1 and GID1b2, that may guide specific GA responses. In this work, CRISPR-Cas9-derived *gid1* mutants were generated and their effect on GA responses was studied. The *gid1* triple mutant was extremely dwarf and fully insensitive to GA. Under optimal growth conditions, the three receptors functioned redundantly and the single *gid1* mutants exhibited very mild phenotypic changes. Among the three receptors, GID1a had the strongest effects on germination and growth. Yeast two-hybrid assays suggested that GID1a has the highest affinity to PRO. Analysis of lines with a single active receptor demonstrated a unique role for GID1a in protracted response to GA that was saturated only at very high doses. When the *gid1* mutants were grown in the field under ambient changing environments, they showed phenotypic instability, the high redundancy was lost and *gid1a* exhibited dwarfism that was strongly exacerbated by the loss of another *GID1b* receptor gene. These results suggest that redundancy in GA sensing is required for phenotypic robustness under environmental extremes.

Flash Talk - ISPS2019-203

Local auxin biosynthesis regulates root regeneration

Rotem Matosevich-Lepar*[1], Michela De Martino[1], Itay Cohen[1] and Idan Efroni[1]

[1] Institute of Plant Sciences and Genetics in Agriculture, The Robert H. Smith Faculty of Agriculture, Food, and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

*Corresponding Author Email: rotem.matosevich@mail.huji.ac.il

The root apical meristem (RAM) coordinates root growth throughout the plant life. The RAM structure is maintained by a complex network of transcription factors and phytohormones, the main one being auxin. Remarkably, severe damage to the RAM, such as complete excision of the stem cell niche, triggers rapid regeneration. Auxin is required for this process and auxin response markers are rapidly induced near the excision site. The current model states that auxin distribution in plant tissues is mainly regulated by polar localized PIN-FORMED (PIN) transporters. However, this has not been tested during regeneration, when normal patterns are severely disrupted. We used genetic and chemical perturbations to test the role of PIN-mediate auxin transport during regeneration. Surprisingly, disruption of auxin transport does not inhibit regeneration, however, local inhibition of auxin biosynthesis resulted in aborted regeneration. High resolution transcriptomics and imaging reveals two sources of auxin production in the regenerating root, acting in temporal succession. Using chemical inhibition and artificial miRNA simultaneously targeting 6 YUCCA auxin biosynthesis enzymes, we blocked auxin synthesis at specific tissues and time points. Inhibition of the early auxin source delays regeneration, while inhibition of the later source blocks root reformation. Active biosynthesis is no longer necessary once tissue patterns have been recovered. To further investigate the factors that enable localized auxin accumulation, we focused on early regeneration genes. The ERF115 transcription factor is rapidly induced at the excision site and is required for regeneration competence. We show that ERF115 act by promoting localized auxin response and that ERF115 deficiency can be rescued by auxin application. Taken together, we present a novel mechanism for regulating auxin distribution when tissue patterning is disrupted. We propose that the ability to specify new sources of auxin production following damage is one of the cardinal features that enable plant regeneration.

Flash Talk - ISPS2019-204

Stick-Bend-Grow Dynamics Underlies Root Waving

Zhenwei Zhang[1], Danny Van Open[1], Naftali Lazarovich[1] and Ido Regev*[1]

[1] French Associates Institute of Agriculture and Biotechnology of Drylands, The Institutes for Desert Research, Ben-Gurion University of the Negev, Midreshet Ben-Gurion, Israel

*Corresponding Author Email: regevid@bgu.ac.il

The origin of root waving, the growth of Arabidopsis thaliana roots in a sine wave pattern, when grown on an Agarose plate tilted in 45 degrees with respect to the horizon, is still debated. Specifically, it is not clear if root waving occurs due to an internal, genetic, growth mechanism or due to interaction of the root with the agar. Here we suggest a quantitative mechanical model in which static friction caused by the gravitropic tendency of the root to grow into the agar, limits the growth of the root and leads the growing zone to buckle elastically. The buckling explains why the root tip changes direction during bending and why the root twists concurrently to the bending. We demonstrate that the buckling equations agree with the experimentally observed dynamics and follow by studying waving experimentally at different tilt angles. We observe changes in wavelength and amplitude which are in qualitative agreement with the mechanical model. These results indicate that the origin of the waving phenomenon is primarily mechanical and can only occur in thin roots which are susceptible to buckling.

Flash Talk - ISPS2019-205

The Use of Hairy Roots for Validation of CRISPR/Cas Plasmid Constructs and Genome Functional Analysis in Composite Plants

Bekele A. Tuffa*[1], Michael Kravchik[1], Yulia Shnaider[1], Diana Leibman[1], Yehudit Tam[1], Dalia Woolf[1], Victor Gaba[1] and Amit Gal-On[1]

[1] Department of Plant Pathology and Weed Research, The Volcani Center, Bet Dagan 50250, Israel

*Corresponding Author Email: bekelet@volcani.agri.gov.il

Since the inception of CRISPR/Cas technology for genome engineering, Agrobacterium tumefaciens-mediated pl ant transformation has been frequently employed for developing new crop varieties. Once CRISPR/Cas-sgRNA plasmid constructs are synthesized and before they are being used for plant transformation, validation of their effectiveness is indispensable. So far, sequence analysis sometimes coupled with agro-infiltration procedures are being employed to do the same. However, in recalcitrant crops such as the cucurbitaceae, agro-infiltration procedures are either ineffective or impossible. Alternatively, validation of CRISPR/Cas-sgRNA cassettes and characterization of edited genes in A. rhizogenes induced hair roots seem reliable and time saving. The CRISPR/Cas-sgRNA plasmid constructs were mobilized into ATCC15834 or K599 A. rhizogenes strains and used to infect tomato and potato, or melon and cucumber explants, respectively. Genomic DNA was extracted from in vitro grown kanamycin resistant transgenic roots, amplified by PCR, and the PCR products were analyzed by restriction digested and Sanger sequencing. In most of the cases we observed deletion of DNA sequences located in the sgRNA target regions, upstream of the Proto Spacer Adjacent Motif (PAM). In other cases deletions of nucleotides both upstream and downstream of the PAM or removal of the whole DNA sequence between two sgRNA target sites were observed. Although the same CRISPR/Cas-sgRNA cassette was used in tomato and potato, the types of insertions/deletions (indels) showed significant variations. Validation of CRISPR/Cas-sgRNA plasmid constructs in A. rhizogenes induced hairy roots, before implementing whole plant transformation procedures could be time saving. In vitro grown A. rhizogenes induced hairy roots could efficiently be used for validation of CRISPR/Cas-sgRNA plasmid constructs. Generally, coupling CRISPR/Cas mediated genome engineering with A. rhizogenes induced hairy roots, could provide a better working model for genome functional analysis, especially in recalcitrant composite plants.

Flash Talk - ISPS2019-207

Next-Generation trait mapping in melon

Elad Oren*[1], Tamar Lahav[2], Ravindra Banaudha[2], Galil Tzuri[2], Vitaly Portnoy[2], Ayala Meir[2], Asaf Dafna[2], Adi Faigenboim[3], Joseph Burger[2], Yaakov Tadmor[2], Yonatan Elkind[1], Shiri Freilich[2] and Amit Gur[2]

[1]Institute of Plant Sciences and Genetics in Agriculture, The Robert H. Smith Faculty of Agriculture, Food, and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

[2]Newe-Ya'ar Research Center, ARO, Ramat Yishay, 3009500, Israel

[3]Agricultural Research Organization, Volcani Center, Rishon LeZiyyon, 7505101, Israel

*Corresponding Author Email: elad.oren@mail.huji.ac.il

Trait mapping is a core component in forward genetics and marker assisted breeding. Until recent years, genetic mapping was mostly based on single-copy, low-throughput DNA markers, and whole-genome projects relied on a maximum of several hundreds of markers. Post genomic era, where reference genomes are available and genotyping is moving towards sequence-based methods, is calling for a methodological shift and implementation of suitable tools. In the current project we are calibrating and implementing several approaches to build genetic maps from high-density sequence-based genotyping of melon populations. We demonstrate the challenges and tools to deal with them and generate high-quality dense maps. The importance of relying on several strategies to deal with common problems such as missing genotypic data, sequencing errors, structural variation, and reference assembly errors is discussed. These practices will be used to map traits in multiple melon populations and align all to the reference melon genome sequence (Cm V3.5.1). We expect this multi-allelic comparative approach will improve mapping confidence and resolution. As part of this project, a comprehensive data set is constructed from various populations (currently consists of 38,000 phenotypic data points) to allow mapping of multiple fruit related traits in a comparative framework. Examples for our recent high-resolution mapping results will be presented and a proposed downstream bioinformatics pipeline to prioritize candidate genes within QTL intervals will be described.

Flash Talk - ISPS2019-208

An efficient pipeline for production of new Lisianthus varieties by combining floral dipping and targeted gene knockout

Sonia Ehi-Eromosele[1], Fang Fang[2], Tzahi Arazi[2] and Michal Oren-Shamir*[1]

[1] Institute of Plant Sciences and Genetics in Agriculture, The Robert H. Smith Faculty of Agriculture, Food, and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

[2] Institute of Plant Sciences, The Volcani Center, Agricultural Research Organization, Bet Dagan, 50250, Israel

*Corresponding Author Email: vhshamir@volcani.agri.gov.il

Lisianthus (Eustoma grandiflorum), belonging to the Gentianaceae family is a prominent cut-flower that is rapidly growing in recognition due to its large flowers, long stems, and extended vase life, and was rated sixth among the top 10 cut flowers by Royal Flora Holland in 2016. It is considered the 'next rose' due the rose-like shape of its flowers and the dramatic increase in its sales over the last few years (over 30% increase in sales between 2015 and 2016). To date, lisianthus transformation has been achieved via Agrobacterium transformation of leaf explants, however only few cultivars were successfully transformed using this method, with a very low yield. The Agrobacterium-mediated tissue culture method is laborious, requiring technical ability, and time consuming due to the long regeneration period from explant to T1 seeds. Hence, development of efficient methods are sought after for stable transformation of this plant specie. We demonstrated floral-dipping transformation of a commercial plant, lisianthus Excalibur Pink with fluorescent Agrobacterium containing a GFP marker gene. The establishment of an efficient floral-dip protocol may accelerate the molecular breeding of this cut-flower and related plant species. Many studies have previously been conducted in attempts to alter flower color, flower fragrance and flowering time in lisianthus via tissue culture-mediated transformation. Genetic engineering via gene editing technology offers great potential for the production of ornamental plants with novel flower colors. We are currently establishing an efficient CRISPR/Cas9 system for inducing targeted mutagenesis in Lisianthus using our Agrobacterium-mediated floral dip method. In this study, the CHS (Chalcone synthase) and FLS (Flavone synthase) genes targeted for knockout are key genes in the Lisianthus anthocyanin biosynthesis pathway and are responsible for a wide array of floral colors. We expect to generate mutant lisianthus plants in these target genes, and influence floral colour.

Fang, Fang, Moran Oliva, Sonia Ehi-Eromosele, Michele Zaccai, Tzahi Arazi, and Michal Oren-Shamir. "Successful floral-dipping transformation of post-anthesis lisianthus (Eustoma grandiflorum) flowers." The Plant Journal (2018) 96, 869-879.

Flash Talk - ISPS2019-209

Identification and characterization of Indole-3-carbinol response mutants in Arabidopsis thaliana

Hala Khamesa*[1] and Danny Chamovitz[1]

[1] School of Plant Sciences and Food Security, The George S. Wise Faculty of Life Sciences, Tel Aviv University, Ramat Aviv, Tel Aviv 69978, Israel

*Corresponding Author Email: kh.hala19@gmail.com

Indole-3-carbinol (I3C) is a small molecule produced as a breakdown product of glucosinolates in the Cruciferae family following mechanical damage or insect attack. I3C functions in as a protective agent against foraging insects. It is also implicated in human health with significant anti-cancer properties. In Arabidopsis, exogenously applied I3C inhibits root elongation in a dose dependent manner, acting as an auxin antagonist that competes for the binding site of the auxin receptor Tir1. Autophagy induced following I3C treatment is specific, targeting auxin receptors. Yet, little else is known about I3C perception, signaling and detoxification. This work aims at identifying novel components of the pathways interacting with I3C signaling in Arabidopsis. We employed a genome-scale artificial microRNA (amiRNA) screen, using modified Arabidopsis thaliana miR319a precursor to express amiRNAa targeting protein kinase families, to identify strains with either enhanced or reduced responses to exogenous I3C. We have identified and confirmed two strains where the amiRNA down-regulates distinct protein kinase families. Strain I3CR2 shows significant resistance to I3C-induced root growth inhibition, while strain I3CS1 is hypersensitive to I3C. Our analyses indicate that the kinase families identified intersect I3C signaling through auxin-independent pathways.

ISPS2019-210

To bloom or not to bloom? Exploring tree winter physiology through carbohydrate metabolism and temperature kinetics

Or Sperling*[1]

[1] Gilat Research Center, Department of Horticulture, Agriculture Research Organization (ARO), The Volcani Center, M.P. Negev, 85280, Israel

*Corresponding Author Email: orsperling@gmail.com

Sustainability of perennial species in seasonal climates requires vital and synchronized blooming and plants gauge winter progression to time bud-break. Yet the persistent physiological mechanisms that track environmental changes are still obscured. We postulated that this 'winter memory' of dormant plants incorporates their most important budget, their energetic sugar reserves, and set to associate them to climate and phenology.

ISPS2019-211

Prediction and large-scale analysis of plastid operons reveals unique genetic features in the evolution of chloroplasts

Noam Shahar[1], Iddo Weiner[1] and Iftach Yacoby*[1]

[1] School of Plant Sciences and Food Security, The George S. Wise Faculty of Life Sciences, Tel Aviv University, Ramat Aviv, Tel Aviv 69978, Israel

*Corresponding Author Email: iftachy@post.tau.ac.il

While bacterial operons have been thoroughly studied, few analyses of chloroplast operons exist, limiting the ability to study fundamental elements of these structures and utilize them for synthetic biology. Here, we describe the creation of a plastome-specific operon database achieved by combining experimental tools and predictive modeling. Using a Reverse-Transcription-PCR based method and published data, we determined the transcription-state of 210 gene-pairs originating from four evolutionary distinct chloroplast-containing organisms. By analyzing sequence-based features computed for our data-set, we were able to highlight fundamental characteristics differentiating between operons and monocistrons. These include an interesting tendency towards maintaining similar mRNA-folding profiles in operon gene-pairs, a feature which failed to yield any informative separation in Cyanobacteria, suggesting that it catches unique traits of operon gene expression which have evolved in chloroplasts. Subsequently, we used this feature set to train a random-forest classifier for operon prediction. As our results demonstrate the ability of our predictor to obtain accurate (85%) and robust predictions on unlabeled data-sets, we proceeded to building an operon-map database for all sequenced plastids. The availability of our database may now present new opportunities for promoting the metabolic-engineering field in plastid-containing organisms such as higher-plants and algal species.

ISPS2019-212

Mapping the Arabidopsis Metabolic Landscape by Untargeted Metabolomics at Different Environmental Conditions

Yariv Brotman[1] and Si Wu*[1]

[1] Department of Life Sciences, Ben Gurion University of the Negev, Beer-sheva, Israel

*Corresponding Author Email: brotmany@post.bgu.ac.il

Metabolic genome-wide association studies (mGWAS), whereupon metabolite levels are regarded as traits, can help unravel the genetic basis of metabolic networks. A total of 309 Arabidopsis accessions were grown under two independent environmental conditions (control and stress) and subjected to untargeted LC–MS-based metabolomic profiling; levels of the obtained hydrophilic metabolites were used in GWAS. Our two-condition-based GWAS for more than 3000 semi-polar metabolites resulted in the detection of 123 highly resolved metabolite quantitative trait loci ($p \leq 1.0E-08$), 24.39% of which were environment-specific. Interestingly, differently from natural variation in Arabidopsis primary metabolites, which tends to be controlled by a large number of small-effect loci, we found several major large-effect loci alongside a vast number of small-effect loci controlling variation of secondary metabolites. The two-condition-based GWAS was followed by integration with network-derived metabolite–transcript correlations using a time-course stress experiment. Through this integrative approach, we selected 70 key candidate associations between structural genes and metabolites, and experimentally validated eight novel associations, two of them showing differential genetic regulation in the two environments studied. We demonstrate the power of combining large-scale untargeted metabolomics-based GWAS with time-course-derived networks both performed under different abiotic environments for identifying metabolite–gene associations, providing novel global insights into the metabolic landscape of Arabidopsis.

ISPS2019-213

Adaptation to stress in annual Brachypodium spp. along the aridity gradient in Israel

Shira Penner*[1] and Yuval Sapir[1]

[1] School of Plant Sciences and Food Security, The George S. Wise Faculty of Life Sciences, Tel-Aviv University, Tel Aviv 69978, Israel

*Corresponding Author Email: shirapenner@mail.tau.ac.il

Climate change is troubling for plants facing increased drought. Current knowledge regarding evolutionary potential of plants to mitigate climate change is limited. Therefore, we are utilizing the natural variation in plants growing along the aridity gradient in Israel, from Mediterranean to extreme desert climates, to understand adaptation to future aridity. Three annual species of the grass Brachypodium grow naturally along the aridity gradient in Israel, comprise of three cytotipic species differ by chromosomes number and ploidy levels. Using flow cytometry, we determined ploidy level of plants collected from >20 natural Brachypodium populations along this gradient and found that ploidy level was not associated with aridity. In a common-garden we found that desert populations are more adapted to cope with future aridity, mainly through earlier flowering time, whereas ploidy level does not play a role in this phenotypic adaptation. In order to test for the effects of ecological factors on adaptation to drought in Brachypodium, we performed a controlled watering experiment in the Botanical Garden and a reciprocal experiment in Mediterranean and arid sites, where we also tested for adaptation to inter-specific competition, which assumed to be the major stress in Mediterranean climate. We measured survival and performance of plants from across the gradient, both diploids and polyploids in these experimental plots, and found that adaptation to the original climate, rather than ploidy level, affects fitness, morphology and phenology under the combination of drought and competition stresses. These findings support the results from population survey that suggested no association of ploidy level and phenotype across the gradient. Moreover, the results support the hypothesis that long-term natural selection created pre-adaptation to changes in climatic conditions. Overall, this study proposes that phenotypic pre-adaptation to drought results from local adaptation, and that genome doubling is not necessarily a genomic mechanism underlying drought tolerance.

ISPS2019-214

The Dead Can Nurture: Novel Insights into the Function of Dead Organs Enclosing Embryos

Buzi Raviv*[1], James Godwin[1], Gila Granot[1] and Gideon Grafi[1]

[1] French Associates Institute of Agriculture and Biotechnology of Drylands, The Institutes for Desert Research, Ben-Gurion University of the Negev, Midreshet Ben-Gurion, Israel

*Corresponding Author Email: Buziraviv@gmail.com

Plants have evolved a variety of dispersal units whereby the embryo is enclosed by various dead protective layers derived from maternal organs of the reproductive system including seed coats (integuments), pericarps (ovary wall, e.g., indehiscent dry fruits) as well as floral bracts (e.g., glumes) in grasses. Commonly, dead organs enclosing embryos (DOEEs) are assumed to provide a physical shield for embryo protection and means for dispersal in the ecosystem. In this review article, we highlight recent studies showing that DOEEs of various species across families also have the capability for long-term storage of various substances including active proteins (hydrolases and ROS detoxifying enzymes), nutrients and metabolites that have the potential to support the embryo during storage in the soil and assist in germination and seedling establishment. We discuss a possible role for DOEEs as natural coatings capable of “engineering” the seed micro-environment for the benefit of the embryo, the seedling and the growing plant.

ISPS2019-215

Pre-RNA maturation and respiratory complexes assembly during germination and early seedling development in Arabidopsis thaliana plants

Laure D. Sultan*[1] and Oren Ostersetzer-Biran[1]

[1] Department of Plant and Environmental Sciences, The Alexander Silberman Institute of Life Sciences, The Hebrew University of Jerusalem, Givat Ram, Jerusalem 91904, Israel

*Corresponding Author Email: laure.sultan@gmail.com

The expression of the mtDNA in plants is complex, particularly at the posttranscriptional level. One of the most remarkable features of plant mitochondrial gene-expression and RNA processing involves the splicing of many group II-type introns, which lie within the coding regions of many of the organellar genes. The excision of the introns from the coding regions they interrupt is essential for mtDNA expression, and regulated by various cofactors that belong to a diverse set of protein families. These factors may also link respiratory-mediated functions with environmental or developmental signals [1, 2].

The plant organellar group II introns have likely evolved from maturase (MAT)-encoding RNAs. Yet, these are degenerated in sequence and lost their related intron-encoded ORFs. In angiosperms, only a single MAT (i.e. *matR*, encoded within *nad1* intron4), has retained in the mtDNA of angiosperms. In addition to MatR, angiosperms also encode several MAT-related proteins (nMAT 1 to 4). MatR and the nMATs are expressed during early stages of seed development, but their levels decline considerably following germination [3-7]. Previously we showed that MatR and the nMATs function in the splicing of most of the mitochondrial introns in Arabidopsis, and that their activities are required for the biogenesis of the respiratory machinery and respiratory functions of fully matured plants [3-7].

Why plant mitochondria gene-expression involves such a complex array of distinct transcriptional and post-transcriptional steps? Currently, we cannot provide a definitive explanation, but we speculate that these essential RNA-processing steps may evolved during seed-plants evolution as consequences of a need for tighter regulation of organellar functions by the nucleus, during breaking of seed dormancy. Here, we report our current analyses of the roles of maturase factors on mitochondria biogenesis during imbibition and early seedlings establishment stages in Arabidopsis.

1. Dalby, S. J.; Bonen, L., Impact of low temperature on splicing of atypical group II introns in wheat mitochondria. *Mitochondrion* **2013,** 13, (6), 647-655.

2. Schmitz-Linneweber, C.; Lampe, M.-K.; **Sultan, L. D**.; Ostersetzer-Biran, O., Organellar maturases: A window into the evolution of the spliceosome. *Biochimica et Biophysica Acta (BBA) - Bioenergetics* **2015,** 1847, ((9)), 798-808.

3. Keren, I.; Bezawork-Geleta, A.; Kolton, M.; Maayan, I.; Belausov, E.; Levy, M.; Mett, A.; Gidoni, D.; Shaya, F.; Ostersetzer-Biran, O., AtnMat2, a nuclear-encoded maturase required for splicing of group-II introns in Arabidopsis mitochondria. *RNA* **2009,** 15, (12), 2299-2311.

4. Keren, I.; Tal, L.; Colas des Francs-Small, C.; Araújo, W. L.; Shevtsov, S.; Shaya, F.; Fernie, A. R.; Small, I.; Ostersetzer-Biran, O., nMAT1, a nuclear-encoded maturase involved in the *trans*-splicing of *nad1* intron 1, is essential for mitochondrial complex I assembly and function. *Plant J* **2012,** 71, (3), 413-426.

5. Cohen, S.; Zmudjak, M.; Colas des Francs-Small, C.; Malik, S.; Shaya, F.; Keren, I.; Belausov, E.; Many, Y.; Brown, G. G.; Small, I.; Ostersetzer-Biran, O., nMAT4, a maturase factor required for nad1 pre-mRNA processing and maturation, is essential for holocomplex I biogenesis in Arabidopsis mitochondria. *The Plant Journal* **2014,** 78, (2), 253-268.

6. **Sultan, L. D**.; Mileshina, D.; Grewe, F.; Rolle, K.; Abudraham, S.; Głodowicz, P.; Khan Niazi, A.; keren, I.; Shevtsov, S.; Klipcan, L.; Barciszewski, J.; Mower, J. P.; Dietrich, A.; Ostersetzer, O., The reverse-transcriptase/RNA-maturase protein MatR is required for the splicing of various group II introns in Brassicaceae mitochondria. *The Plant Cell* **2016,** 28, (11), 2805-2829.

7. Zmudjak, M.; Shevtsov, S.; **Sultan, L. D**.; Keren, I.; Ostersetzer-Biran, O., Analysis of the Roles of the Arabidopsis nMAT2 and PMH2 Proteins Provided with New Insights into the Regulation of Group II Intron Splicing in Land-Plant Mitochondria. *International journal of molecular sciences* **2017,** 18, (11).

ISPS2019-216

CO_2 Regulation of Stomatal Movements in the Face of Global Climate Change

Tamar Azoulay-Shemer*[1,2,4], Andisheh Bagheri[2], Cun Wang[2], Aaron Stephan[2], Hans-Henning Kunz[2], Nikki Schwankl[2], Ido Rog[3], Menachem Moshelion[4] and Julian I. Schroeder[2]

[1] Newe Ya'ar Research Center, Agriculture Research Organization, Ramat Yishay, Israel.

[2] Division of Biological Sciences, Cell and Developmental Biology Section, University of California San Diego, La Jolla, CA, USA

[3] The Robert H. Smith Institute of Plant Sciences and Genetics in Agriculture, Faculty of Agricultural, Food & Environmental Quality Sciences, The Hebrew University of Jerusalem, Rehovot, Israel

[4] Department of Plant and Environmental Sciences, Weizmann Institute of Science, Rehovot, Israel

*Corresponding Author Email: shemer.tamar@volcani.agri.gov.il

Stomatal pores are responsible for >95% of plant water loss. CO2 levels in leaves are determined by respiration, photosynthesis, stomatal conductance and atmospheric [CO2]. Increased CO2 levels in leaves cause stomatal closing. Mesophyll and guard cells fix CO2 into carbohydrates, which are then stored in the chloroplasts as starch. The role of starch metabolism in guard-cell and mesophyll cells in stomatal conductance responses is a matter of debate, and genetic approaches are needed.

To study whether starch-metabolism in guard cells and/or in mesophyll cells is rate-limiting for CO2-induced stomatal movement. Stomatal CO2 responses, CO2 assimilation-rates, and starch levels quantification in defined starch-metabolism mutants (AGPase, pPGI, BAM1, BAM3, SEX1) were conducted. Data reveal that starch biosynthesis in guard cells but not mesophyll cells are an essential-process required for proper CO2-induced stomatal closure. TPU-limited photosynthesis is not a direct transducer of CO2-induce stomatal closure. Furthermore, photoperiod length found to dramatically affect stomatal conductance responses to [CO2] via AGPase, which was found to be a rate-limiting for CO2-induced stomatal closure, solely under short-day growth conditions. Surprisingly, our data show that mutations in the starch degradation enzymes BAM1, BAM3 & SEX1 do not disrupt stomatal conductance responses to CO2-shifts.

ISPS2019-217

Transposable elements facilitate the identification and uncovering the underlying mechanisms of large-scale genomic rearrangements in wheat

Inbar Bariah*[1], Danielle Keidar-Friedman[1] and Khalil Kashkush[1]

[1] Department of Life Sciences, Ben-Gurion University of the Negev, Beer-Sheva 8410501, Israel

*Corresponding Author Email: bariah@post.bgu.ac.il

Wheat is a good model to study allopolyploidization and genome evolution, due to multiple allopolyploidization events that occurred during its evolution and the relatively young age of the hexaploid bread wheat. Following allopolyploidization (hybridization and chromosome doubling), the newly formed polyploid genome reacts in a burst of genomic changes, including deletion of transposable elements (TEs) containing sequences. Nevertheless, the mechanisms of DNA elimination following allopolyploidization events remain unknown. In this study, a specific variant of the Fatima LTR retrotransposon was retrieved from the recently available high-quality genome drafts of T. aestivum (genome AABBDD) and wild emmer, T. turgidum ssp. dicoccoides (genome AABB), and used as a genetic marker to identify sequence variations between wheat allopolyploids. Comparative alignment of sequences flanking wild emmer specific Fatima insertions led to the identification of DNA sequences, sized up to millions of bp, that underwent insertions/deletions (InDels) between T. turgidum ssp. dicoccoides and T. aestivum. The identification and characterization of the InDels borders using a chromosome walking approach shed light on possible underlying mechanisms of large structural variation between wheat allopolyploids. The breakpoints of large-scale deletions are consistent with DNA elimination via unequal intra-strand recombination and double-strand break (DSB) repair. Other cases of sequence variation identified between wheat allopolyploids were due to sequence duplication and introgression of novel DNA fragments. Finally, PCR validations using the domesticated form (T. turgidum ssp. durum) of wild emmer wheat revealed some of the identified InDels have already occurred in durum, suggesting that domestication and speciation might have a prominent role in stabilization of the nascent polyploid species through genomic rearrangements.

ISPS2019-218

The Best Defense is Attack: The Viral Protein P0 Induces a Plant ER-Derived Degradation Pathway for the Decay of Membrane-Bound Argonaute 1 (AGO1)

Simon Michaeli*[1], Marion Clavel[1], Corrado Viotti[1], Esther Lechner[1], Thibaut Hacquard[1], Jian Wu[2], Marieke Dubois[1], Gad Galili[2], Hervé Vaucheret[3] and Pascal Genschik*[1]

[1] Institut de Biologie Moléculaire des Plantes (IBMP), CNRS, Strasbourg, France

[2] Department of Plant and Environmental Sciences, Weizmann Institute of Science, Rehovot, Israel

[3] Institut Jean-Pierre Bourgin, INRA Centre de Versailles-Grignon, Versailles, France

*Corresponding Author Email: simon.michaeli@ibmp-cnrs.unistra.fr

Plant viruses are responsible for enormous crop losses worldwide. To defend themselves, plants employ several anti-viral mechanisms, including RNA-silencing. However, during the evolutionary arms race with their hosts, viruses have acquired specialized proteins that counteract RNA-silencing, termed Viral Suppressors of RNA-silencing (VSRs). Plant ARGONAUTE1 (AGO1) is pivotal in RNA-silencing, hence is a major target of VSRs. The VSR P0 from Turnip Yellow virus (TuYV) was formerly shown to trigger AGO1 degradation via an autophagy-like process. However, the identity of host proteins involved and the cellular site at which AGO1 and P0 interact, were unknown. By employing live-cell imaging and electron-microscopy, we show that P0 and AGO1 associate on the Endoplasmic Reticulum (ER), resulting in their loading into ER-associated vesicles that are mobilized to the vacuole in an autophagy-dependent manner. Nevertheless, autophagy deficiency was unable to block P0-dependent AGO1 degradation. We further identified ATG8-Interaccting proteins 1 and 2 (ATI1 and ATI2) as proteins that associate with P0 and interact with AGO1 on the ER up to the vacuole. Notably, ATI1 and ATI2 represent an endogenous degradation pathway of ER-associated AGO1 that is significantly induced following P0 expression. Moreover, ATI1 and ATI2 deficiency renders significant increase in post-transcriptional gene silencing (PTGS) activity. Collectively, we identify ATI1 and ATI2 as representatives of a pathway involved in ER-associated AGO1 turn-over and proper PTGS maintenance and we show that this pathway is manipulated by a single viral protein. This research promotes our understanding of AGO1 post-translational regulation and our knowledge on the manner by which viruses exploit cellular pathways of their hosts to promote infection.

ISPS2019-219

Multi-omics approach provides insights into extreme illumination stress response

Haim Treves*[1], Beata Siemiatkowska[1], Urszula Luzarowska[1], Omer Murik[2], Ute Armbruster[1], Yariv Brotman[1], Stefan Rensing[3], Jedrzej Szymanski[4] and Mark Stitt[1]

[1] Max Planck Institute of Molecular Plant Physiology, 14476 Potsdam-Golm, Germany

[2] Department of Plant and Environmental Sciences, The Hebrew University of Jerusalem, 91904 Jerusalem, Israel.

[3] Faculty of Biology, Philipps-University Marburg, 35032 Marburg, Germany.

[4] Network Analysis and Modelling, IPK, Gatersleben, Germany

*Corresponding Author Email: Trebes1024@mpimp-golm.mpg.de

The unparalleled performance of Chlorella ohadii (Treves et al., 2016), clearly indicated that we lack essential information on the photosynthetic machinery and what sets the upper growth limits. Unlike other photosynthetic organisms, C. ohadii productivity is unaffected by irradiances of twice full sun light; Rather than succumbing to photodamage C. ohadii undergoes major structural and compositional changes emphasizing the unique PSII functioning as well as highly efficient reductant metabolic utilization downstream of the photosynthetic reaction centers. When grown under optimal laboratory or controlled outdoor conditions, this alga, recently isolated from one of the harshest environments (a biological desert sand crust), exhibits the fastest growth rates ever reported for an alga, division times shorter than 2 h were recorded. Growth of batch cultures under continuous high light (3000 µmol photons m-2 s-1) combined with metabolome analyses revealed a highly coordinated metabolic switch, supporting growth to higher densities than those achieved if abolished, and regulated by specific signaling molecules of the Polyamines group (Treves et al., 2017). We applied an array of systems biology tools to reveal regulation of gene networks at the metabolic, redox and expression levels, for the first time under extreme illumination. Combined multi-omics analyses identified key response regulators and provided novel insights into the mechanism underlying its exceptional photodamage resistance, including growth vs. stress signaling, morphological response and starch excess effects. These, together with a transformation system developed these days for C. ohadii will allow to dissect what distinguishes this alga from its more sensitive counterparts. Treves et al., 2016 New phytologist Treves et al., 2017 Current Biology.

ISPS2019-220

Genome-wide analyses of miniature transposable elements revealed new insights into the evolution of Triticum-Aegilops group

Danielle Keidar-Friedman[1], Inbar Bariah[1], Chen Doron[1] and Khalil Kashkush*[1]

[1] Department of Life Sciences, Ben-Gurion University of the Negev, Beer-Sheva 8410501, Israel

*Corresponding Author Email: kashkush@bgu.ac.il

Wheat has been the subject of intensive research into polyploidization and genomic evolution due to multiple allopolyploidization events. The newly formed allopolyploid genome reacts in reorganizations that include activation or deactivation of transposable elements (TEs). Plant genomes harbor an enormous amount of TEs, which might account for up to 90% of some genomes such as in wheat. Despite extensive studies, the role of TEs in reshaping nascent polyploid genomes remains to be fully understood. In this study, we retrieved and analyzed miniature TEs from the recently published genome drafts of T. aestivum, T. dicoccoides, T. durum, Ae. tauschii and T. urartu. We have found high copy numbers of miniature TEs and characterized a new family that has not been identified before (named “Inbar”). About 50% of the insertions were found within or near (less than 100 bp) coding genes and over 400 insertions were found in T. aestivum (bread wheat) transcriptome, indicating their impact on wheat genome expression. We have analyzed the association of Au SINE family with wheat protein-coding genes and found that that both regular transcripts and alternative Au SINE-containing transcripts were simultaneously amplified in the same tissue, indicating exonization of Au SINE-containing introns. Many wheat genes possess different splice variants, in which some transcripts contain a TE insertion and others do not. Gene expression analysis of MITE (miniature inverted-repeat TEs) containing transcripts showed that in some cases, these insertions lead to a higher expression in some wheat species, while others show similar expression levels of the transcript without an insertion. Our data sheds new light on the role of miniature TEs in the diversification of allopolyploid wheat species.

ISPS2019-221

Heterosis in pine hybrids (Pinus brutia X Pinus halepensis)

Naomi Houminer*[1], Yagil Osem[1], Joseph Riov[1] and Rakefet David-Schwartz[1]

[1] Department of Agronomy and Natural Resources, The Volcani Center, Bet Dagan 50250, Israel

*Corresponding Author Email: naomi.houminer@mail.huji.ac.il

The planted forest in Israel is dominated by Pinus halepensis (Aleppo pine) which is considered native in this region. Since the 1970'sthe local forest service gradually turned to the exotic Pinus brutia as the leading conifer used for afforestation due to its tolerance to the Israeli pine bast scale Matsucoccus josephi. When P. brutia (as female) and P. halepensis (as male) overlap geographically, natural hybridization occurs. Recently, mature hybrids of the two species were identified in several forests in Israel. These hybrids exhibit morphological traits and a vigorous growth that distinguish them from adjacent P. brutia and P. halepensis of similar age. However, it is not known what the frequency of hybridization is, what percentage of hybrids exhibits hybrid vigor and to what extent it is affected by abiotic or biotic conditions. Hybrids are not easy to identify by morphology alone and can be easily mistaken as variants of P. brutia or P. halepensis. Therefore, they are identified by molecular markers targeting mitochondrial and chloroplast DNA. These markers however, are only good for F1 hybrid due to the paternal inheritance of chloroplasts and maternal inheritance of mitochondria in pines and cannot identify advanced generation hybrids. To better identify hybrids and to answer the above questions, we generated and analyzed the transcriptomes of P. brutia and P. halepensis needles and identified 12 nuclear Single Nucleotide Polymorphisms (SNP) based markers. The SNPs can be identified by Cleaved Amplified Polymorphic Sequences (CAPS) and High-Resolution Melt (HRM) analyses. Preliminary survey was done, we looked for brutia stands that had some vigorous trees (that were suspected as hybrids). In each plot, 80 random trees were screened for height, diameter at breast high (DBH) and genotype. We found that hybrid trees frequencies are between 2.5-6%. Also, the overwhelming majority of hybrids exhibit higher vigor. All trees

ISPS2019-222

The ABI4 transcription factor is post-transcriptionally regulated by abiotic stress and hormones

Tzofia Maymon[1], Nadav Eisner[1], Ruth Finkelstein[2] and Dudy Bar-Zvi*[1]

[1] Department of Life Sciences, Ben-Gurion University of the Negev, P.O. Box 653, Beer-Sheva 84105, Israel

[2] Department of Molecular, Cellular and Developmental Biology, University of California at Santa Barbara, Santa Barbara, California 93106, USA

*Corresponding Author Email: barzvi@bgu.ac.il

The Abscisic Acid Insensitive 4 (ABI4) gene encodes an AP2-family transcription factor that plays a role in ABA signaling. ABI4 is also involved in seed development and germination, response to abiotic stresses such as drought and salinity, control of the lipid mobilization in the embryo, lateral root formation and is also required for redox control. ABI4 is relatively highly expressed during seed maturation and in early stages of seed germination. Its transcription levels dramatically reduced at later developmental stages. It is activated by glucose, ABA, salt and repressed by the growth hormone auxin. It was also shown that the protein levels of ABI4 are very low, suggesting that it protein levels may be also regulated posttranscriptional. We used transgenic Arabidopsis plants expressing eGFP-tagged ABI4 driven by the strong viral CaMV 35S promoter, to study posttranscriptional regulation of this highly modulated transcription factor. We found that steady state levels of ABI4 were extremely low in the roots of seedling grown in optimal conditions. These levels were markedly enhanced upon exposure of the seedlings to abiotic stress and ABA. In addition, ABI4 is rapidly degraded by the 26 S proteasome. Interestingly, although the expression of the eGFP-tagged ABI4 was driven by the constitutive 35S CaMV promoter, that is highly active in most plant cells, the fluorescent ABI4 –tagged protein was observed primarily in the stele. This study suggests that both the level and the cell type accumulation of ABI4 protein is tightly regulated. Interestingly, abiotic factors and plant hormones have similar effects on its transcript and protein levels. These double-check control of ABI4 reflects on its central role in plant development and cellular roles.

ISPS2019-223

Correlations between circadian rhythms and growth in challenging environment in wild barley

Yuri Dakhiya*[1], Duaa Hussien[1], Eyal Fridman[2] , Moshe Kiflawi[3] and Rachel Green[1]

[1] Department of Plant and Environmental Sciences, Silberman Institute for Life Sciences, Hebrew University, Givat Ram, Jerusalem 91904, Israel

[2] Plant Sciences, Vegetable and Field Crops, Agriculture Research Organization (ARO), The Volcani Center, Rishon LeZiyon, Israel

[3] Department of Life Sciences, Ben Gurion University, Beer Sheva, Israel

*Corresponding Author Email: yuri.dakhiya@mail.huji.ac.il

Circadian clocks are biological time-keeping mechanisms that allow organisms to execute important physiological functions at the appropriate time of day as well as predicting daily environmental changes. Circadian rhythms that can match changes in the environment were shown to be adaptive in several model plants. Manipulation of the circadian system is a promising path for increasing the vitality and yield of crop plants but unfortunately, not many studies of circadian clocks have been done outside a small group of model plants. Our goal is to examine how different challenging environmental conditions affect circadian phenotypes while focusing on agriculturally relevant plants like wild barley (Hordeum vulgare ssp. spontaneum), the progenitor of cultivated barley (Hordeum vulgare ssp. vulgare). Early results suggest that certain circadian phenotypes are correlated with specific environmental conditions, which might reveal interesting, and potentially valuable, adaptations to these environments.

ISPS2019-224

A mechanism controlling polymerization of silica in grass leaves

Rivka Elbaum*[1] and Santosh Kumar[2]

[1] Institute of Plant Sciences and Genetics in Agriculture, The Robert H. Smith Faculty of Agriculture, Food, and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

[2] Weizmann Institute of Science, Rehovot, Israel

*Corresponding Author Email: rivka.elbaum@mail.huji.ac.il

Grasses uptake silicic acid from soils and deposit it in their leaves as solid silica. This mineral comprises 3-10% of the grass dry weight. Nevertheless, silica deposition process is not understood. In this work, we compared a sorghum mutant defective in silicic acid uptake to wild-type plants. We show that specialized epidermal leaf cells condense silicic acid into silica while viable. The silica starts polymerizing at the cell periphery, and condenses into the cell volume. A live non-silicified cytoplasm is restricted to a limited volume by the growing silicified cell wall. We show that a protein that may be secreted from the silica cells induces the silicic acid polymerization. This biomineralization course is opposed to the common understanding that silica forms because of passive water transpiration.

ISPS2019-225

Green Algal Hydrogenase Activity Is Outcompeted by Carbon Fixation before Inactivation by Oxygen Takes Place

Yuval Milrad*[1], Shira Schweizer[1], Yael Feldman[1] and Iftach Yacobi[1]

[1] School of Plant Sciences and Food Security, The George S. Wise Faculty of Life Sciences, Tel Aviv University, Ramat Aviv, Tel Aviv 69978, Israel

*Corresponding Author Email: yuvalmilrad@gmail.com

Photoproduction of hydrogen by green algae is considered a transitory release valve of excess reducing power and a potential carbon-free source of sustainable energy. It is generally accepted that the transitory production of hydrogen is governed by fast inactivation of hydrogenase by oxygen. However, our data suggest that photosynthetic electron loss to competing processes, mainly carbon fixation, stops hydrogen production, supports hydrogen uptake, and precedes the inevitable inactivation by oxy- gen. Here, we show that when transitioning from dark anaerobiosis to light, hydrogen production ceases within 2 min, regard- less of the presence of oxygen. Simultaneous monitoring of the active hydrogenase pool size shows that it remains entirely intact up to 4 min after illumination and is inactivated only later. Thus, our data reveal a window of 4 min in which the hydrogenase pool is not being degraded by oxygen. Furthermore, we show that electron loss, prominently to carbon fixation, outcompetes hydrogen production and leads to hydrogen uptake. Indeed, supplying additional reducing power to hydrogenase at the cessa- tion point regenerates the accumulation of hydrogen. Our results imply the fast cessation of hydrogen production is governed by electron loss rather than oxygen inactivation, which takes place minutes later. Photosynthetic.

ISPS2019-226

Candidate genes that could mediate cucumber fruit set and its inhabitation by older fruit: expression study and CRISPR inactivation

Deblina Mitra*[1], Yula Shnaider[1], Amalia Bar-Ziv[1] and Rafael Perl Treves[1]

[1] Department of life Science, Bar-Ilan university

*Corresponding Author Email: perl.lab1@gmail.com

Cucurbits represent an attractive model to explore the dynamics of fruit set. A fertilized ovary integrates signals from distant plant parts and "decides" whether to set fruit, or remain inhibited and later abort. We set to characterize first fruit inhibition (FFI), i.e. the inhibitory effect of the first fruit on subsequent development of younger ovaries. The effect was physiologically studied and quantified, and we compared its strength and persistence in pollinated versus parthenocarpic ovaries. We compared gene expression profiles of pollinated ovaries (committed to set fruit) with respect to those affected by FFI, and to non-pollinated ovaries (undergoing senescence). The three fates were characterized by wide changes in gene expression, and potentially interesting genes - transcription factors, hormone-metabolism, cell wall remodeling, and defense genes were identified, being up- or down-regulated as early as one day post-anthesis. Transgenic cucumbers that over-express invertase genes were generated to try and alter assimilate flux and sink strength in the plant. Trehalose-6-phosphate is an emerging sugar signal that could be associated with fruit set and assimilate partitioning; we found that trehalose-6-phosphate phosphatase (TPP) genes seem to respond to FFI. We successfully mutated two TPP family members (as well as two invertase genes) by CRISPR-Cas9 technology, to see whether such mutations affect fruit or seed set phenotypes in cucumber.

ISPS2019-227

The wild emmer wheat stripe rust resistance gene Yr15 was 'left behind' during domestication due to its narrow geographical distribution

Valentina Klymiuk*[1], Andrii Fatiukha[1] and Tzion Fahima[1]

[1] Institute of Evolution and Department of Evolutionary and Environmental Biology, University of Haifa

*Corresponding Author Email: valentinaklymiuk@gmail.com

Stripe (yellow) rust, caused by the fungus Puccinia striiformis f. s. tritici (Pst), is a destructive disease of wheat spread globally. The wild emmer wheat (Triticum dicoccoides; WEW) gene, Yr15, which confers a broad-spectrum resistance to Pst, encodes a tandem kinase protein designated WTK1 that has multiple homologous copies in the wheat genome. However, only the WTK1-1B from WEW accession G25 is conferring resistance to Pst. Here, we describe the development of functional molecular markers (FMMs) that differentiate between functional (Wtk1) and non-functional (wtk1) Yr15 alleles. Screening of a worldwide collection of 545 wheat cultivars and wheat related species with Yr15 FMMs showed that only 32 of them contained introgression of Yr15 from G25, while all the rest contained the wtk1 allele. Furthermore, only 16% of 382 WEW accessions collected across the Fertile Crescent harbored the functional Wtk1 allele. Wtk1 was found only among WEW natural populations that reside along a narrow axis of 140 km from Mt. Carmel to Anti-Lebanon mountain ridge, at elevation of above ~500 meters, where the climatic conditions are favorable for pathogen development and, thus, may exert positive selection in favor of the Wtk1 allele. Since this narrow region is not overlapping with southeast Turkey where wheat is believed to be domesticated, we propose that Yr15 gene was 'left behind' and therefore it is not included in modern domesticated genepool. Our results highlight the importance of conservation of WEW populations in their natural habitats for the discovery of novel R-genes and the study of host-parasite co-evolution.

ISPS2019-228

A plant DNMT3 ortholog establishes de novo DNA methylation in Physcomitrella patens independently of RdDM

Rafael Yaari[1], Aviva Katz[1], Katherine Domb[1], Keith D. Harris[1], Assaf Zemach[1] and Nir Ohad[1]

[1] School of Plant Sciences and Food Security, The George S. Wise Faculty of Life Sciences, Tel Aviv University, Ramat Aviv, Tel Aviv 69978, Israel

*Corresponding Author Email: Niro@tauex.tau.ac.il

To properly regulate the genome, cytosine methylation needs to be established and maintained by DNA methyltransferases (DNMTs). DNMT3 is a conserved eukaryotic DNMT known to establish methylation in mammals. While the plant altered DNMT3 homologs, DOMAINS REARRANGED METHYLTRANSFERASEs (DRMs), was shown to establish methylation via the RNA directed DNA methylation (RdDM) pathway, the role of true plant DNMT3 orthologs remained elusive. To elucidate the role of plant DNMTs, we profiled genomic methylation and de novo methylation in the basal plant, Physcomitrella patens, mutated in each of its PpDNMTs. To evaluate the activity of P. patens DNMTs in de novo methylation, we introduced the repetitive DNA sequence (RPS) from Petunia hybrida uncommon to moss P. patens. DNA methylation analysis of RPS was conducted in the first transgenic generation (T1) and within the same transformed plant tissue, using bisulfite sequencing. Our results show that RPS is methylated in WT cells in all three methylation contexts, CG, CHG, and CHH, implying on its ability to be de novo methylated in P. patens. To our knowledge, RPS methylation by PpCMT serves as novel evidence for a CMT DNMT performing de novo methylation in vivo. The differential effect observed in dnmt3b on RPS de novo CG methylation but not on preexisting CG methylation, while CG methylation was nearly eliminated in met for both RPS and genome wide as expected for a maintenance DNMT, indicates PpDNMT3b is a de novo CG DNMT. Altogether, our results reveal that de novo methylation in P. patens is mediated by DNMT3b and by CMT can be established without the involvement of DRMs or the canonical RdDM pathway.

ISPS2019-229

Genetic biofortification of common wheat by exploitation of alleles from wild emmer wheat

Andrii Fatiukha*[1], Valentina Klymiuk[1], Tzion Fahima[1] and Tamar Krugman[1]

[1] Institute of Evolution and the Department of Evolutionary and Environmental Biology, Faculty of Natural Sciences, University of Haifa, Haifa, Israel

*Corresponding Author Email: avfatyha@gmail.com

Common wheat (Triticum aestivum, genome BBAADD) is one of the most important food crop, which is providing ~20% of all calories consumed worldwide, but lacking necessary micronutrients for human diet. Wild emmer wheat, WEW (Triticum turgidum ssp. dicoccoides, genome BBAA) was shown to confer high grain protein (GPC) and mineral content, therefore harbors a great potential for improvement of cultivated wheat nutrition value. Biofortification is a cost-effective and sustainable process of genetic enhancement of micronutrient in crops through plant breeding, which can be facilitated by application of marker-assisted selection (MAS). Here, we report a successful case of genetic biofortification of common wheat using MAS, which started with identification of nutrient QTLs and resulted in the introgression of wild emmer wheat alleles into common wheat. A recombinant inbred line (RIL) population derived from a cross between T. durum var. Langdon and WEW (acc. G18-16) was genotyped using high-throughput SNP array. Based on the constructed ultra-dense genetic map, QTL mapping of nutrient content was performed resulting in 12 GPC QTLs with a LOD score range of 2.5–11.1 and PEV range of 1.1–17.2%. Three major GPC QTLs (located on 4BS, 5BS and 7AL) with pleotropic effects on sulfur, zinc, iron, copper and calcium content were selected and introgressed into four Israeli common wheat varieties (Ruta, Gedera, Yuval and Zahir). Field validation experiment of BC3F3 near isogenic lines (NIL) showed an increase in GPC for all introgressions with an average increase in GPC for the tested backgrounds ranging from 8.7% to 15.1%. Effects of the introgressions on GPC were independent from grain yield. The obtained pre-breeding materials can serve as valuable source for development of wheat cultivars with improved quality and nutrition.

ISPS2019-230

Singlet Oxygen Plays an Essential Role in the Root's Response to Osmotic Stress

Tomer Chen[1] and Robert Fluhr*[1]

[1] Department of Plant and Environmental Sciences, Weizmann Institute of Science, Rehovot, Israel

*Corresponding Author Email: tomerlivechen@gmail.com

The high osmotic potentials in plants subjected to drought stress can be mimicked by the application of high molecular weight polyethylene glycol. Here, we quantified the effects of exposure to polyethylene glycol on the growth of the main and lateral roots of Arabidopsis (Arabidopsis thaliana) seedlings. The effects on root growth were highly correlated with the appearance of singlet oxygen, as visualized using the singlet oxygen-specific probe singlet oxygen sensor green. The production of singlet oxygen was followed by cell death, as indicated by the intracellular accumulation of propidium iodide due to the loss of membrane integrity. Cell death began in the epidermal region of the root tip and spread in a dynamic manner to meristematic sections. In parallel, gene expression changes specific to the presence of singlet oxygen were observed. The accumulation of other reactive oxygen species, namely; hydrogen peroxide, nitric oxide, and superoxide, did not correlate with cell death. In addition, both the singlet oxygen scavenger His and the lipoxygenase inhibitor salicylhydroxamic acid specifically inhibited singlet oxygen accumulation and cell death. These results suggest a light-independent, type-I source of singlet oxygen production. Serpin-protease interactions were used as a model to assess the possibility of vacuolar-type cell death. Osmotic stress induced the accumulation of complexes between the cytoplasmic serpin AtSERPIN1 and its cognate vacuolar proteases, indicating that vacuolar integrity was compromised. These findings imply that singlet oxygen plays an essential role in conveying the root response to osmotic stress.

ISPS2019-231

The complete plastid genome sequence and the photosynthetic activity of the putative mycoheterotrophic orchid *Limodorum abortivum*

Sofia Shevtsov*[1], Omer Murik*[1], Hagit Zer[1], Ofir Weinstein[1], Nir Keren[1], Ori Fragman-Sapir[2] and Oren Ostersetzer-Biran[1]

1 Dept. of Plant and Environmental Sciences, Institute of Life Sciences, The Hebrew University of Jerusalem, Givat Ram, Jerusalem 9190401, Israel.

2 Jerusalem Botanical Gardens, Herbarium, The Hebrew University of Jerusalem, Givat Ram, Jerusalem, 9021904 Israel

*Corresponding Author Email: sofia.shevtsov@gmail.com

The sparsely distributed *Limodorum abortivum* is a European-Mediterranean orchid species, which grows on decomposing plant material. Although some chlorophyll are observed in the degenerated scales-shaped leaf and stems regions, its photosynthetic capacity is assumed to be insufficient to support the full energy requirements of an adult plant. In Israel, *L. abortivum* shows a patchy distribution patterns in the Galilee, Golan, Carmel and Judean regions. To gain more insights into the physiology and photosynthetic activity of *L. abortivum*, we analyzed the organellar morphologies and photosynthetic activities and determined the chloroplast-DNA sequence by Illumina-HTS. Microscopic analyses indicated to the presence of mature chloroplasts with well-organized grana-thylakoids in the leaves and stems of *L. abortivum*. However, the numbers of chloroplasts per cell and the grana ultrastructure density within the organelles were notably lower than those of model plant species and fully photosynthetically active orchids. The cpDNA of *L. abortivum* (154,954 bp) encodes 60 proteins, 34 tRNAs and 4 rRNAs. The coding-regions of 24 genes are interrupted by 26 group-II intron-sequences. While many genes related to photosynthesis (RuBisCo, PSI, PSII and cytochrome *b6/f* subunits) have remained intact in the cpDNA, the majority of the NADH-dehydrogenase (*ndh*) subunits were either lost or became nonfunctional (i.e., pseudogenized). In agreement with previous reports, the photosynthetic-rates of adult *Limodorum* plants were found to be very low, further indicating that carbon-assimilation activity is insufficient to support the energy requirements of an adult plant, and may suggest that *L. abortivum* have adopted nutritional strategies similar to that of mycoheterotrophic orchid species.

ISPS2019-232

Mechanical Considerations Underlying Context Specific Impact of Brassinosteroids in the Arabidopsis Root

Yulia Fridman*[1], Eduard Federov[1], Neta Holland[1], Tom Shemesh[1] and Sigal Savaldi-Goldstein[1]

[1 1] Technion, Israel institute of Technology, Haifa, Israel.

*Corresponding Author Email: zarka@bgu.ac.il

Intracellular factors controlling cell proliferation, cell expansion and directionality are the underpinnings of plant morphogenesis. These factors respond to mechanical signals, interconnected hormonal pathways and corresponding transcriptional networks. Despite dramatic advances in identifying hormonal signaling components, how their spatiotemporal activities integrate to coordinate these processes are not fully understood. Using the steroid hormone brassinosteroid (BR) signaling pathway, we uncovered opposing tissue-specific effects of the BR signal that dictate the longitudinal (apical-basal) and the radial dimensions of the Arabidopsis root meristem. In agreement, our translatome mapping in response to the hormone suggested its context-specific effect on gene expression. This provides an entry point to explore the mechanisms underlying tissue-specific interpretation of the hormone, as a means of achieving balanced growth. Here we evaluate whether mechanical considerations could explain this differential growth output. Towards this end, we use imaging and dedicated software to quantify the volume of virtually all cells, in combination with readout of BR signaling strength, along the root meristem in wild type and BR mutants with tissue-specific perturbation of BR signaling. These data is applied in theoretical models to assist us in evaluating the relevance of mechanical input on growth decisions and hence, further bridge the gap between the established core signaling cascade and its less understood interpretation at the cell, tissue and organ levels.

ISPS2019-233

Plant mitochondria group II introns splicing: A window into the evolution of the nuclear spliceosomal machineries

Corinne Best[1], Laure Sultan[1], Sofia Shevtsov[1], Oren Ostersetzer-Biran*[1]

[1]Department of Plant and Environmental Sciences, The Alexander Silberman Institute of Life Sciences, The Hebrew University of Jerusalem, Givat-Ram, Jerusalem 91904, Israel.
*Corresponding Author Email: oren.ostersetzer@mail.huji.ac.il

Mitochondria are key sites for cellular energy metabolism and play pivotal roles in the biosynthesis of numerous essential metabolites. As descendants of a free-living bacterium, mitochondria contain their own genomes. In plants, the mtDNAs are notably larger and more complex in structure than their relatives in Animalia. These are also remarkable with respect to the complex RNA-metabolism. The maturation of the plant mitochondrial transcripts include extensive RNA-editing and the splicing of numerous introns, which are closely-related to group-II-type sequences found in many prokaryotic genomes. In bacteria, the removal of group-II-related introns from their coding-sequences is facilitated by proteins encoded within the introns themselves (Maturases), which bind specifically to their host-introns, and act as retrohoming elements, able to mobile their cognate-introns into ectopic genome-loci. Remarkably, under non-physiological (high salt and temperature) conditions, in the absence of any protein-cofactors, the group-II introns are able to excise themselves from the pre-RNAs, by a mechanism identical to that utilized by the spliceosomes. Structural and phylogenetic data suggest that the spliceosomal-RNAs may have evolved from group-II ancestors. Yet, it remains unclear how such general spliceosomal players have evolve from a monospecific Maturase/group-II system. Analysis of the splicing activities in plant mitochondria provide us with important clues into the evolution of the spliceosomal system. Genetic and biochemical studies led to the identification of numerous enzymes, which belong to a diverse set of protein-families and facilitate the splicing of plant mitochondrial pre-RNAs. We found that the plant Maturases function on multiple intron-targets, thus seem to be acting as organellar proto-spliceosome cofactors. We therefore hypothesize that the early mitochondrial self-splicing and mobile group-II introns spread into the host genomes and later 'degenerated' into a universal splicing machinery, known as the spliceosome. The similarities between Maturases and the core spliceosomal factor, Prp8, may support this intriguing theory.

ISPS2019-234

Adjustment of Auxin Output in Tomato Leaf Development by Three ARF Transcription Factors

Alon Israeli[1], Yossi Capua[2], Idan Efroni[1] and Ori Naomi[1]

[1] Institute of Plant Sciences and Genetics in Agriculture, The Robert H. Smith Faculty of Agriculture, Food, and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

[2] Department of Plant and Environmental Sciences, Weizmann Institute of Science, Rehovot, 7610001, Israel

*Corresponding Author Email: AlonIsraeli2@gmail.com

Plant organs display a continuum of shapes, obtained by modulating the specific timing and location of local growth. The plant hormone Auxin regulates many of these patterning processes during the plant life. But how auxin is perceived to form a phenotypic continuum is not completely understood. In developing tomato compound leaves, auxin promotes leaflet formation and blade outgrowth, and in the intercalary regions the auxin response is inhibited by the Aux/IAA protein ENTIRE (E). The e mutant forms simple leaves due to ectopic blade outgrowth in the intercalary domain. Here, we utilized this unique Aux/IAA loss-of-function phenotype to investigate how specific interactions between E and activating Auxin Response Factors (ARFs) can lead to a range of phenotypes. We show that combined activity of auxin transport and response components leads to the quantitative adjustment of blade outgrowth. Genetic analysis shows that the three activator ARFs, SlARF5/SlMP, SlARF19A and SlARF19B act partially redundantly to promote blade outgrowth downstream of auxin and E in a dosage-dependent manner. Our findings demonstrate how quantitative adjustment of an auxin output by a specific module of auxin-response components enables flexible patterning.

ISPS2019-235

Identifying defense metabolites that Setaria plants synthesis in response to aphid attack

Anuma Dangol[1], Chandrashekhar Kottakota[1] and Vered Tzin[1]

[1] French Associates Institute for Agriculture and Biotechnology of Drylands, Jacob Blaustein Institute for Desert Research, Ben Gurion University, Sede-Boqer Campus 84990, Israel

*Corresponding Author Email: anuma@post.bgu.ac.il

Setaria viridis is a C4, short life cycle emerging model plant, which belongs to Panicoideae. It also serves as a resilient crop that gives good yields even in dry and marginal land. The defense mechanism of Setaria against insect herbivory is unstudied. Therefore, we are interested in studying the major defense mechanism of Setaria against the cereal aphid Rhapalosiphum padi. To identify the aphid defensive metabolites, we performed the metabolomic analysis of aphid-infested and uninfested tissues using Gas Chromatography/Mass Spectrometry (GC/MS). This analysis revealed that the Trp-derived metabolite serotonin is highly produced in response to insect attack. The accumulation of serotonin was preceded by tryptamine increment by over-expression of one isozyme of Tryptophan Decarboxylase (TDC) genes. A transcriptomic analysis identifies this TDC gene was significantly induced after 6 and 96 hours of aphid infestation. An artificial diet supplemented with serotonin demonstrated that this molecule suppressed the survivability and fecundity of R padi. These findings are the first indication that S. viridis plants synthesize serotonin as a defense metabolite and also expose the biosynthetic genes involve in this pathway. Exploiting the major defense metabolite of this model-crop plant is fundamental for understanding its adaptation for biotic stresses and it is critical for crop improvement of more complex plant systems such as maize, rice wheat, and sorghum.

ISPS2019-236

Volatile organic compounds emitted by the desert truffle *Terfezia boudieri* induce changes in root architecture and plant growth of its host plant *Helianthemum sessiliflorum*

Lakkakula Satish[1,2], Ariel Kushmaro[2], Varda Kagan-Zur[3], Nurit Rot-Bejerano[3], Einat Bar[4], Efraim Lewinsohn[4] and Yaron Sitrit*[1]

[1] The Jacob Blaustein Institute for Desert Research, Ben-Gurion University of the Negev, Beer Sheva, 84105, Israel.

[2] Department of Biotechnology Engineering, Ben-Gurion University of the Negev, Beer Sheva, 84105, Israel.

[3] Department of Life Sciences, Ben-Gurion University of the Negev, Beer Sheva, 84105, Israel.

[4] Department of Vegetable Crops, Newe Ya'ar Research Center, Ramat Yishay, 30095, Israel.

*Corresponding Author Email: sitrit@bgu.ac.il

Volatile organic compounds (VOCs) are involved in many ecological interactions between organisms. We have previously analyzed the VOCs composition of various desert truffles and found that 1-octen-3-ol (mushroom alcohol) is the major volatile present. Fungal VOCs play an important signaling role in the symbiosis interactions with their host plants in their natural environments. Experiments were conducted to investigate the effect of *T. boudieri* mycelia VOCs and 1-octen-3-ol on the development of its host plant *Helianthemum sessiliflorum*. Co-cultivation of *T. boudieri* mycelia and *H. sessiliflorum* plantlets in a bipartite petri dish (without direct contact) caused a significant suppression in primary root growth and stimulated lateral roots formation. In control experiments in the absence of *T. boudieri* mycelia the primary roots were longer and no lateral roots produced. These experiments demonstrated that fungal VOCs have profound effects on the host root morphology. Moreover, *T. boudieri* VOCs treated plantlets showed a marked inhibition of shoot growth and caused significant decreases in biomass compared to the control plantlets after a week of co-cultivation. The effect of 1-octen-3-ol administration on seed germination was also evaluated and a marked dose-dependent inhibition of germination was noted. After exposure of seeds for 3 days to 1 mg/L 1-octen-3-ol, germination was markedly inhibited and no seeds germinated at 10 and 100 mg/L 1-octen-3-ol. The effects of VOCs and 1-octen-3ol mimic auxin effects, but auxin has a more localized effect. It seems that *T. boudieri* mycelia may evoke beneficial effects on host development through a variety of mechanisms including auxin secretion, which affects root architecture. The auxin-like effect caused by fungal VOCs and 1-ocen-3-ol is not due to secretion of auxin by fungus and clearly under natural conditions the volatiles may affect roots but not shoots. Our results suggest that VOCs may have auxin-like activity or evoke auxin synthesis *in-planta*.

ISPS2019-237

Chemodiversity in Mandragora spp. is associated to loss of functionality of MoH6H, a hyoscyamine 6β-hydroxylase gene

Daniel Schlesinger[1], Rachel Davidovich Rikanati[2], Sergei Volis[3], Adi Faigenboim[4], Federica Cattonaro[5], Matthew Hooper[6], Matthew Taylor[6], Yaron Sitrit[7], Moshe Inbar[1] and Efraim Lewinsohn*[2]

[1] Department of Evolutionary & Environmental Biology, University of Haifa, Haifa 31905, Israel

[2] Department of Vegetable Crops, Newe Ya'ar Research Center, Agricultural Research Organization, PO Box 1021, Ramat Yishay, 30095 Israel.

[3] Kunming Institute of Botany

[4] Institute of Plant Sciences, The Volcani Center, Agricultural Research Organization, Bet Dagan, 50250, Israel

[5] Applied Genomics Institute (IGA), Udine, Italy

[6] The James Hutton Institute

[7] French Associates Institute for Agriculture and Biotechnology of Drylands, Jacob Blaustein Institute for Desert Research, Ben Gurion University, Sede-Boqer Campus 84990, Israel

*Corresponding Author Email: twefraim@volcani.agri.gov.il

Tropane alkaloids derivatives scopolamine and hyoscyamine (often as its racemic form atropine) are used in modern medicine as pain and motion sickness relievers, eye pupil dilators and antidotes against organo-phosphate poisoning. An intermediate in the conversion of hyoscyamine to scopolamine is 6β - hydroxyhyoscyamine (also referred to as anisodamine). Hyoscyamine, anisodamine and scopolamine are commonly present in members of the Solanaceae such as Datura, Hyoscyamus, and Mandragora. Israeli populations of Mandragora officinarum contain 5 to 10-fold higher levels of hyoscyamine but lack of anisodamine and scopolamine, unlike the European Mandragora spp., in which scopolamine and its precursors are most prominent. Hyoscyamine is converted to 6β - hydroxyhyoscyamine and then to scopolamine by the action of hyoscyamine 6β-hydroxylase (H6H), a 2-oxoglutarate dependent dioxygenase enzyme. Transcriptomic and CAPS analyses allowed the identification of polymorphisms in the gene encoding for hyoscyamine-6β hydroxylase in Mandragora spp. H6H is highly conserved among scopolamine-producing Solanaceae, including M. autumnalis but is mutated in M. officinarum in eight amino acid residues. One of the mutations is the change of glycine amino acid by cysteine in residue 220 which is localized in the 2-oxoglutarate co-substrate binding region.

Functional expression analyses revealed that MaH6H, a gene isolated from a M. autumnalis accession of Moroccan origin and DiH6H, a gene isolated from Datura inoxia encode active H6H enzymes while the mutated H6H sequences isolated from several accessions of Israeli M. officinalis roots were functionally inactive. In addition, a deliberate Glycine220Cysteine mutation in the D. inoxia DiH6H gene caused the loss of functionality of the protein. Our results suggest that a mutation in the MoH6H sequence results in chemo-variation in the tropane alkaloid content of Mandragora spp.

ISPS2019-238

Topo climate effects on phenology and berry metabolism of red and white wine grapevine cultivars under extreme desert conditions

kelem Gashu Alamrie[1], Eran Harcavi[2], Elyashiv Drori[3], Amnon Bustan[4], Nurit Agam*[1] and Aaron Fait*[1]

[1] French Associates Institute for Agriculture and Biotechnology of Drylands, Jacob Blaustein Institute for Desert Research, Ben Gurion University, Sede-Boqer Campus 84990, Israel

[2] Ministry of Agriculture and Rural Development, Agricultural Extension Service - Shaham, POB 28, Bet-Dagan 50250, Israel

[3] Agriculture Research Department, Eastern Region Research and Development Center, Ariel 40700, Israel

[4] Desert Agriculture Research Center, Ramat Negev R&D, Halutza D.N. 85515, Israel

* Corresponding Author Email: agam@bgu.ac.il, aaron.fait@gmail.com

Desert conditions are considered beyond the climate frame of traditional wine producing belts due to water scarcity, high temperatures and excess light/UV intensity, all negatively affecting fruit metabolism and wine quality. However the semiarid to arid regions are inevitably becoming wine grapevine growing area in Israel, the Negev desert offers diverse topo climate conditions, with altitudes ranging from 250 to 900 m asl. To identify cultivars with crop quality potentials under desert environments two experimental vineyards were setup in Ramat Negev R&D Center, and Ramon, at 300 and 850m asl, respectively comprising 10 white, and 20 red cultivars. In the first two harvest years (2015 and 2016), Chennin Blanc and French Colombard among the white cultivars, and Petit Verdot and Malbec among the red ones, exhibited promising wine qualities, with some advantages to the relatively cooler region, Ramon. A consistent two-week difference in plant and berry phenology between the two vineyards was preserved from bud break to véraison, while the differences among cultivars at each site were small in 2017 and 2018 growing seasons. The ripening period from véraison to harvest was on average 50% longer in the significantly warmer Ramat Negev vineyard, where a considerable number of fruit clusters shriveled before reaching the BRIX harvest threshold. Metabolic data of berry skin and pulp are being processed to assess environmental and varietal interaction on primary and secondary metabolism.

ISPS2019-239

Characterizing gibberellins flow in planta using photocaged gibberellins

Shira Wexler[1], Hilla Schayek[1], Kondhikonda Rajendar[1], Iris Tal[1], Eilon Shani[1], Yasmine Meroz[1], Roman Dobrovetsky[1] and Roy Weinstain*[1]

[1] Tel Aviv University, Israel

* Corresponding Author Email: shira.her@gmail.com

Gibberellins (GA) are ubiquitous plant hormones that coordinate central developmental and adaptive growth processes in plants. Accurate movement of GA throughout the plant from its sources to its destination sites is emerging to be a highly regulated and directed process. We report on the development of novel photocaged gibberellins that, in combination with a genetically encoded GA-response marker, provide a unique platform to study GA movement at high-resolution, in real-time and in living, intact plants. Applying this platform to the Arabidopsis thaliana endogenous bioactive gibberellin GA4, we measure kinetic parameters of its flow, such as decay length and velocity, in vivo for the first time.

ISPS2019-240

Identifying QTL for the control of flowering time under multiple environments in lettuce

Leah Rosental*[1,2], Youngsook You[3], David Still[3], Ivan Simko[2] and Ryan Hayes[2]

[1] Department of Life Sciences, Ben Gurion University of the Negev, Beer-sheva, Israel

[2] U.S. Department of Agriculture-Agricultural Research Service, Crop Improvement and Protection Research Unit, 1636 East Alisal Street, Salinas, 93905, CA

[3] California State Polytechnic University, Pomona

* Corresponding Author Email: leah.zucker@mail.huji.ac.il

Premature bolting and early flowering is an economic problem, exacerbated by heat conditions, where many varieties will bolt before harvest, producing bitter latex and rendering the product unsalable. In order to better understand the genetic factors controlling the environmental response to this trait a RIL population was studied in six independent field studies in three different locations. The parents for the population are Salinas, a slow bolting iceberg type lettuce, and PI251246, a fast bolting lettuce grown for oil in its seeds. Environmental conditions during the field studies ranged from maximum temperate of 32.2°C to 42°C. Based on GBS genotyping of the 161 RILs we constructed a linkage map of 753 markers, with good coverage of the nine lettuce chromosomes. QTL mapping revealed four significant QTLs for bolting and flowering, with different effects for the two traits. All four QTL have strong environmental interactions and have a significant effect on the traits only under some of the conditions tested. A QTL on chromosome 2 (QTL2.1) had a higher effect on bolting and flowering in warmer experiments, while the large QTL on chromosome 7 (QTL 7.2) had a stronger effect on bolting during cooler long day conditions. Another QTL on chromosome 7 had a smaller though significant effect on both bolting and flowering. The QTL on chromosome 6 had a high effect on bolting time under five conditions, but a lower effect on flowering. The three highest LOD QTLs were investigated further by fine mapping and gene expression studies. We found lettuce FT gene to be a likely candidate for QTL 2.1 and two known flowering time genes, COL-9 and PhyC to be likely candidates in QTL 7.2. The information revealed in this work is being used with marker assisted selection to improve lettuce breeding lines for resistance to premature bolting.

ISPS2019-241

The low monoterpene content of tomato fruit is apparently due to low expression levels of LeGPPS.SSU, a gene encoding for the small subunit of geranyl diphosphate synthase

Gal Hivert*[1], Rachel Davidovich-Rikanati[2], Einat Bar[2], Arthur Schaffer[2], Joseph Hirschberg[1], Natalia Dudareva[3] and Efraim Lewinsohn*[2]

[1] Department of Genetics, Hebrew University of Jerusalem, Jerusalem 91904, Israel

[2] Department of Vegetable Crops, Newe Ya'ar Research Center, Agricultural Research Organization, PO Box 1021, Ramat Yishay, 30095 Israel.

[3] Department of Horticulture and Landscape Architecture, Purdue University, 625 Agriculture Mall Drive, West Lafayette, Indiana 47907, USA.

*Corresponding Author Email: Gal.hivert@gmail.com, twefraim@volcani.agri.gov.il

The tomato (Solanum lycopersicum L.) is the highest-value vegetable crop worldwide. Nonetheless, deterioration in flavor quality of modern tomato cultivars is a major cause of consumer complaints. Volatile compounds such as mono- and sesquiterpenes may contribute to the acceptable flavor of tomatoes but commercial tomatoes contain low amounts of such. In contrast, wild relatives of tomato display variation in the mono- and sesquiterpene composition and content of their fruit. During domestication the cultivated tomato fruit apparently lost its ability to produce volatile terpenes while acquiring the ability to produce large amounts of carotenoid pigments such lycopene. Preliminary evidence indicates that the formation of monoterpenes in cultivated tomato fruit is limited by the availability of the precursor geranyl diphosphate (GPP). The structural genes involved in GPP synthesis have not been identified in tomato fruit. In other plants GPP is produced by the action of geranyl diphosphate synthase (GPPS). GPPS is often a heteromeric enzyme consisting of a small (GPPS.SSU) and a large subunit (GPPS.LSU). GPPS.LSU is similar to geranylgeranyl diphosphate (GGPP) synthases involved in diterpene and carotenoid biosynthesis. GPPS.SSU is usually inactive by itself but upon interaction with the GPPS.LSU directs the synthesis to GPP. We have isolated a gene (LeGPPS.SSU) that is homologous to other plant GPPS.SSU and is present in the tomato genome and its wild relatives, but only negligibly expressed in cultivated tomato fruits. We also demonstrated the ability of LeGPPS.SSU to support GPP formation with the interaction of the tomato LeGGPPS2 in enzymatic assays. In addition, the levels of the volatile terpenes of wild tomatoes corresponded with the levels of expression of LeGPPS.SSU in the fruit. Our results support the hypothesis that LeGPPS.SSU was down regulated during the domestication of tomato which resulted in the loss of the volatile terpenes in favor of carotenoids.

ISPS2019-242

Metabolic Interaction between Tomato Scion and Rootstock Under Salt Treatment

Chao Song[1], Tania Acuna[1], Michal Meir[1], Simon Barak[1], Shimon Rachmilevitch[1] and Aaron Fait*[1]

[1] French Associates Institute for Agriculture and Biotechnology of Drylands, Jacob Blaustein Institute for Desert Research, Ben Gurion University, Sede-Boqer Campus 84990, Israel

* Corresponding Author Email: fait@bgu.ac.il

Salinity is an increasingly expansive problem limiting plant growth and affecting crop production. Grafting is regarded as a promising tool to improve the resistance to soil salinity. Understanding the mechanisms mediating the rootstock affecting the scion is key to broaden the salinity optimum for crops cultivars. In our experiment, we used tomato (Solanum lycopersicum) cv M82 grafted onto 254 different tomato rootstocks, including wild and commercial races and cherry tomatoes. Salt treatment was applied at a concentration of 200 mM NaCl and plant tissue was collected in consecutive weeks for metabolite profiling, ROS estimation, transcript analysis. Plant height, branches per plant, fresh weight of shoot and fruit were measured at harvest. Results showed that grafting same scion onto different rootstocks resulted in phenotypic diversificaiton. Compared with M82 self-grafted (control), the MDA content was regarded as the trait with highest CV and standard deviation cross the populations. In contrast, the ratio of fruit weight to total weight was the trait with the lowest CV. 8 best and 4 worst tomato lines, compared with control, were screened from 254 lines according to the fruit fresh weight, harvest index and MDA content. Metabolic data showed that both of proline and serine decreased compared with control cross the populations. Among the screened lines, these best lines accumulated more amino acids, such as alanine, valine and glycine, than worst lines. Further metabolic and transcriptive analysis are being processed to understand the mechanisms that scion was mediated by the rootstock.

ISPS2019-243

Development of Vegetative Propagation Methods for Mature Eucalyptus Trees Valuable for the Apiary Industry and Research of the Mechanisms underlying Adventitious Roots Formation

Avi Eliyahu*[1], Roy Weinstein[2], Mohamad Abu-Abied[1], Mira Carmeli-Weissberg[1], Felix Shaya[1], Arnon Dag[1] and Einat Sadot[1]

[1] The Plant Science Institute, Agriculture Research Organization (ARO), The Volcani Center, Rishon LeZiyon, Israel

[2] School of Plant Sciences and Food Security, The George S. Wise Faculty of Life Sciences, Tel Aviv University, Ramat Aviv, Tel Aviv 69978, Israel

*Corresponding Author Email: avi.eliyahu9@gmail.com

The Israeli summer and fall are highly arid and only few local species bloom consistently during these seasons. Without supplementary food supplies, beehives suffer during this time from malnutrition, which affects apiaries on an annual basis – the honey is produced in lower quantities, colonies become weaker and more sensitive to pathogens and in severe cases face collapse.

Some *Eucalyptus* species which naturally bloom during the dry seasons, serve as a good food source for honeybees (*Apis melifera*), when most local species do not bloom. Nevertheless, the flowering phenotypes of some of these species are highly variable. Studies have shown that this variability is based on genetic background, at least partially, thus, uniformity can be improved through clonal propagation. Elite trees were chosen according to the advice of experienced beekeepers. Data regarding the flowering intensity, nectar secretion and concentration, and attractiveness to honeybees of some of the excellent trees were collected through field observations. Using novel rooting enhancers developed by our group, we have managed to propagate clones of the following recommended individuals: *E. camaldulensis*, *E. leucoxylon macrocarpa*, *E* x *trabuti*, and an individual suspected as a *E. brachyphylla* hybrid. Moreover, we have also successfully grafted scions from an exceptional blooming *C. ficifolia* on *C. maculata* rootstocks.

To reveal mechanisms underlying AR formation in *Eucalyptus* in response to the new rooting compounds, we characterize hormones and gene profiles in the treated cuttings. In parallel, using light and confocal microscopy, we follow the morphological changes occur in the cutting bases.

ISPS2019-244

Characterizing gibberellin transport activity by the NPF family

Jenia Binenbaum[1], Nikolai Wulff[2], Hussam Hassan Nour-Eldin[2], Eilon Shani*[1]

[1] French Associates Institute for Agriculture and Biotechnology of Drylands, Jacob Blaustein Institute for Desert Research, Ben Gurion University, Sede-Boqer Campus 84990, Israel

[2] Department of Plant and Environmental, University of Copenhagen

* Corresponding Author Email: eilonsh@post.tau.ac.il

The plant hormone gibberellin (GA) is fundamental for many aspects of plant growth and development. The best-known contribution of GA manipulations to agriculture is the introduction of dwarfing alleles into staple crops, which are one of the foundations of the so-called "Green Revolution" that resulted in increased global wheat and rice yields. Movement of GA within plants has been documented and proven to be essential for proper plant growth, however little is known about the molecular mechanisms executing GA transport. Our group recently identified NPF3 as the first bona fide GA transporter in plants. Other groups showed that additional Arabidopsis NITRATE TRANSPORTER 1/PEPTIDE TRANSPORTER family (NPF) family members promote GA uptake in yeast. This line of evidence makes the NPF proteins excellent candidates for further GA transport research. There are 53 NPFs in Arabidopsis, divided into 8 functionally redundant sub families. This generates high complexity of specialization within the family. By employing a multi-targeted amiRNA approach we identified an amiRNA targeting two Arabidopsis genes on the NPF2 sub-clade: NPF2.12 and NPF2.14. We show that NPF2.12 is capable of facilitating GA and nitrate import while NPF2.14 promotes GA export in a heterologous system transport assays conducted in Xenopus oocytes, providing first evidence of GA export activity in plants. Altogether indicating that there are additional unstudied GA transporters within the NPF family. By characterizing the biochemical activity and localization of the novel GA transporters discovered in this work we try to shed light on the NPF2 sub-family activity in GA transport and the GA spatial distribution on the whole organism level of the plant.

ISPS2019-245

Primary metabolic profiling of Broomrape (Phelipanche aegyptiaca)

Yael Hacham*[1,2], Noam Nativ [1], Joseph Hershenhorn[3], Evgenia Dor[3] and Rachel Amir[1,2]

[1] Migal Galilee Technology Center, Kiryat Shmona, Israel;

[2] Tel-Hai College, Upper Galilee, Israel;

[3] Weed Research Dept. Newe Ya'ar Research Center, ARO, Ramat-Yishay, Israel

* Corresponding Author Email: yaelh@migal.org.il

Broomrape (Phelipanche aegyptiaca), a root holoparasitic plant, is among the most destructive agricultural weeds worldwide. Metabolic profiling done for Broomrape at the early developmental stage (tubercle) and for infected and non-infected tomato roots revealed that Broomrape differs significantly from the host roots. Most of the primary metabolites belonging to several biochemical groups of sugars, amino acids, organic acids, TCA metabolites and polyols were higher in Broomrape than at the host's roots. This suggests that Broomrape has the ability to alter and accumulate metabolites derived from the host. In addition, the levels of most of the metabolites were similar at infected and non-infected roots, suggesting that the parasite did not significantly affect the host metabolic pathways at an early stage.

Profile analysis of five Broomrape organs (adventitious roots, lower and upper shoots, floral buds and flowers) in mature flowering plants showed that in accordance with non-parasitic plants, significant changes were found between Broomrape reproductive and vegetative organs. Higher levels of free amino acids and total nitrogen were found in Broomrape flower buds. Using tissue culture and labeled nitrate and ammonium, we showed that Broomrape callus can survive on inorganic media and that the genes necessary for nitrogen assimilation are expressed and active.

ISPS2019-246

New insights into the role of LC-PUFA in Lobosphaera incisa revealed by the comparative mutant analysis

Amit Kugler*[1], Boris Zorin[1], Olga Gorelova[2], Sammy Boussiba[1], Alexei Solovchenko[2] and Inna Khozin-Goldberg[1]

[1] French Associates Institute for Agriculture and Biotechnology of Drylands, Jacob Blaustein Institute for Desert Research, Ben Gurion University, Sede-Boqer Campus 84990, Israel

[2] Department of Bioengineering, Faculty of Biology, Moscow State University

*Corresponding Author Email: amitkug@post.bgu.ac.il

Polyunsaturated fatty acids (PUFA) are important components of membrane lipids in living cells, contributing to membrane homeostasis and regulation of their biophysical characteristics. Unicellular eukaryotic microalgae, mainly marine species, are the primary natural producers of long-chain PUFA (LC-PUFA) with C20 and C22 acyl chains. Less often LC-PUFA occur in freshwater microalgae, whose membrane lipids are generally rich in C18 PUFA. A rare exception is the freshwater chlorophyte Lobosphaera incisa, which accumulates high proportions of the n-6 LC-PUFA, arachidonic acid (ARA), in membrane lipids and in storage lipid triacylglycerols (TAG). This organism represents a potent photosynthetic and non-polluted resource for biotechnological applications, as well as an interesting model organism to study the role of LC-PUFA in microalgae. In this work, we set to provide further insights into the role of ARA in L. incisa. We attempted to use an RNAi approach to engineer strains with reduced expression of the Δ5-desaturase gene, to lessen ARA levels. Screening for putative transformants, revealed lines, named M2-35 and M5-78, which have altered their LC-PUFA biosynthesis pathway, and accumulate oleic acid (OA) and gamma-linolenic acid (GLA), respectively. Remarkable modifications in the acyl group composition of the altered strains in the thylakoid and extraplastidial membrane lipids classes, as well in TAG, were determined. We also show that alterations in the biosynthesis pathway affected the physiological performance and photosynthetic activity of the algal lines when grown under favorable and stress conditions such as chilling temperatures and nitrogen (N)-starvation, as well as the ability to recover from N-starvation. Remodeling of glycerolipids, ultrastructural modifications, and contrasting photosynthetic responses collectively showed that LC-PUFA in L. incisa are essential for proper photosynthetic apparatus functioning and acclimation to abiotic stresses.

ISPS2019-247

Studying the interrelation between the flavonoid and stilbene pathways in Vitis vinifera cv. Gamay Red cell suspension

Ru Wang*[1], Sangram Keshari Lenka[1], Moran Oliva[1,2], Noga Sikron-Persi[3], Kelem Gashu[3] Aaron Fait[3] and Michal Oren-Shamir[1]

[1] Department of Agronomy and Natural Resources, The Volcani Center, Bet Dagan 50250, Israel

[2] Department of Plant and Environmental Sciences, Weizmann Institute of Science, Rehovot, 7610001, Israel

[3] French Associates Institute for Agriculture and Biotechnology of Drylands, Jacob Blaustein Institute for Desert Research, Ben Gurion University, Sede-Boqer Campus 84990, Israel

*Corresponding Author Email: ru-wang@outlook.com

Grapes (Vitis vinifera L.) are important sources of health-promoting bioactive compounds including phenolic compounds such as stilbenes and flavonoids. Despite of extensive studies that have been conducted to modify flavonoids and stilbenes biosynthesis by metabolic engineering strategies, there is still no answer to the interrelation of these two pathways. The main goal of our project is to elucidate the molecular and metabolic interrelations between the flavonoid and stilbene pathways in Vitis vinifera cv. Gamay Red cell suspension. We hypothesize that by genetically targeting this metabolic interception in a grape cell suspension originating from red grapes, we will have an effective tool to challenge the phenylpropanoid metabolism and tackle the interrelation between flavonoids and stilbenes pathways. We established double transformed grape cell cultures by over-expressing 3-deoxy-D-arabino-heptulosonate 7-phosphate synthase (AroG*) with flavonoid synthase (FLS) or stilbene synthase (STS) in grapevine cells and began a metabolic characterization of these transformed lines to the interrelation between the phenolic branches leading either to flavonoid or stilbenes biosynthesis. Our preliminary metabolic analysis of transgenic cell suspension lines, AroG* FLS and AroG* STS suggest that the interrelation between the two pathways may be complex, since AroG* FLS, directing the carbon flux towards flavonoid production, also caused a significant accumulation of stilbenes, vice versa. Subsequently, transgenic cells will be profiled by RNA-seq analysis and metabolic fluxes will be quantified via stable isotope enrichment analysis. Our integrative analysis can provide comprehensive knowledge of interrelation between two branching pathways in Vitis vinifera cv. Gamay Red cell suspension. This knowledge will help us to improve fruit nutritional quality.

ISPS2019-248

The Effects of Combined Salinity and Low Temperature Stress on Grafted Tomato Plants

Naomi Waldman*[1], Shimon Rachmilevitch[1] and Naftali Lazarovitch[1]

[1] French Associates Institute for Agriculture and Biotechnology of Drylands, Jacob Blaustein Institute for Desert Research, Ben Gurion University, Sede-Boqer Campus 84990, Israel

*Corresponding Author Email: naomiw@post.bgu.ac.il

Stressful environmental conditions are causing major losses to agricultural production worldwide, and therefore are being highly investigated from a number of different aspects. Among these stresses, salinity is a significant environmental abiotic stress factor and approximately 20% of agricultural lands worldwide are affected by salinity, mostly arid and desert lands. Furthermore, agricultural crops may be influenced by a combination of abiotic stresses simultaneously. One kind of combination could be salinity and chilling stress: a winter crop abiotic stress factor which can occur in open fields or non-heated greenhouses. Earlier studies have discovered that the combination of different stresses causes a unique response that cannot be determined directly as a response to each of the occurring stresses. Nevertheless, little information is available in response to combined stresses in grafted plants. The aim of this research is to study and compare the effect of combined salinity and chilling stresses on different tomato rootstocks. Furthermore, it aims to explore root structure and function as a mechanism to sustain combined salinity and chilling stresses. Eight tomato (Solanum lycopersicum L.) rootstocks were exposed to two levels of temperature and two levels of salinity, creating single and combined salinity and chilling stresses. The grafted plants were grown on rhizoslides, a two-dimensional paper-based growth system, in controlled growth chambers. The rhizoslides enable the study of root growth rate and architecture under different conditions in non-invasive methods. Transpiration rates, elemental composition, osmolality and root growth and structure were examined. Tolerant rootstocks are expected to exclude salt ions or restrict their transport to the upper parts of the plant as well as to alter the plant's metabolism in order to protect it under stressful conditions, thereby improving the plant's physiological state. Finally, this research will hopefully provide knowledge for good management of tomato cultivation in semi-arid environments.

ISPS2019-249

The role of HEAT SHOCK PROTEIN 70-1 in the regulation of root elongation

Anat Idan-Molakandov*[1], Daniela Arazi-Tenenbaum[1], Ofir Kuperman[1] and Smadar Harpaz-Saad[1]

[1] Institute of Plant Sciences and Genetics in Agriculture, The Robert H. Smith Faculty of Agriculture, Food, and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

*Corresponding Author Email: anat.idanmolaka@mail.huji.ac.il

Plants continuously modify their morphogenetic program in response to environmental signals. Modification of plant growth is obtained in part by modulating the turgor pressure, as well as by remodeling plant cell wall composition and structure. The receptor-like kinases FEI1 and FEI2 are essential to maintain root growth under restrictive conditions of high-salt or high-sucrose concentrations. Genetic screen identified three independent HEAT SHOCK PROTIEN 70-1 (HSP70-1) alleles as suppressors of the fei1fei2 short and swollen root phenotype. These alleles, named hsp70-1shou5, suppress also the swollen root phenotype of salt overly sensitive 5 (sos5), previously shown to function in the same pathway. HSP70s are highly conserved ATP-driven chaperones encoded by enlarged gene families in plants. Interestingly, hsp70-1 loss-of-function allele (SLAK_135531) was unable to suppress the fei1fei2 phenotypes due to functional redundancy with other cytosolic-HSP70s. Transgenic plants expressing FLAG-tagged form of either HSP70-1shou5 or the HSP70-1WT were generated. Using blue native gel electrophoresis, HSP70-1 was identified as part of about 600KDa protein complex. Immuno-precipitation followed by mass spectrometry reveal that the four proteins involved in thalianol synthesis were preferentially bound to HSP70-1WT, but not to HSP70-1shou5. Further study will be required to determine whether HSP70-1 and the thalianol pathway function as part of a cell wall integrity sensing mechanism or downstream of the FEI-SOS5 signaling pathway.

ISPS2019-250

FASCICLIN-LIKE18 is a Novel Element Required for Root Elongation Under Abiotic Stress Conditions in Arabidopsis thaliana

Hewot Allelign-Ashagre*[1], Smadar Harpaz-Saad[1], David Zaltsman[1] and Oren Tzfadia[1]

[1] Institute of Plant Sciences and Genetics in Agriculture, The Robert H. Smith Faculty of Agriculture, Food, and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

*Corresponding Author Email: hewotashagre@gmail.com

The plasticity of root development represents a key trait enabling plants to adapt to diverse abiotic stress conditions. Lateral roots (LRs) are formed repetitively from the primary-embryonic root (PR) determining root system architecture. Both PR and LR growth extent and direction are largely determined by the pattern of plant cell wall deposition. Nonetheless, PR and LRs have been shown to display differential growth dynamics in responses to different environmental cues. A tissue-specific co-expression analysis was conducted in order to identify additional elements involved in cell-wall deposition during root elongation. This led to the identification of FASCICLIN-LIKE 18 (FLA18), an extra-cellular arabinogalactan protein, as a new player required for LR elongation under abiotic stress conditions. Two independent fla18 insertion mutant lines display short and swollen lateral roots, as compared to wild type. Interestingly, double mutant with salt overly-sensitive 5/fla4 displays a synergistic effect on both LR and PR elongation under stress conditions. Like fla4, fla18 exhibits hyper-sensitivity to the cellulose-synthase inhibitor, Isoxaben supporting a role for FLA18 in cell wall deposition. Moreover, fla18 was also hyper-sensitive to abscisic acid (ABA) synthesis inhibitors, as previously described for fla4. Gene expression analysis suggests that both mutants display basal activation of the stress-induced ABA pathway also under ambient conditions. In-depth study of the role of FLA18, as well as other FLA proteins, is anticipated to shed new light on the mechanism involved in the regulation of PR vs. LR elongation determining root system architecture in changing environments.

ISPS2019-251

Eexogenous ABA treatment facilitates date palm fruit ripening

Sa'ar Elbar*[1], Amnon Bustan[2] and Smadar Harpaz-Sa'ad[1]

[1] Institute of Plant Sciences and Genetics in Agriculture, The Robert H. Smith Faculty of Agriculture, Food, and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

[2] Agro-Desert Research Centre, Ramat-Negev Research and Development

*Corresponding Author Email: saar.elbar@mail.huji.ac.il

The date palm (Phoenix dactylifera L.) is a subtropical monocot tree. Date fruit development and ripening requires extremely high temperature and low relative humidity. In habitats where these terms are inadequately supplied, delay in fruit ripening results in deterioration of large yield portions. The main objective of the current study is to uncover mechanisms regulating date palm fruit ripening. Based on the role of the plant hormone abscisic acid (ABA) in the regulation of non-climacteric fruit ripening, we hypothesized that ABA might play a role in the regulation of date palm fruit ripening. Monitoring ABA levels along the course of fruit development reveals a constant increase in endogenous ABA levels at the fruit-pericarp, starting from 16 weeks post-pollination (WPP), as fruit colour changes from green-to-yellow. Exogenous ABA application to the developing fruit 16 WPP enhanced chlorophyll degradation, facilitating the transition from green-to-yellow, compared to control fruits. In addition, the ABA treatment facilitated sugar accumulation, as indicated by an earlier increase in the pericarp Brix value, compared to control fruit. Interestingly, post-harvest ABA treatment facilitated the ripening of mature date palm fruit, as indicated by a significantly faster occurrence of peel browning and fruit flesh softening, compared to control fruits. These results suggest that ABA plays a pivotal role in the regulation of date palm fruit ripening. Fine-tuning of ABA applications may provide practical tools to control date fruit maturation and ripening under sub-optimal conditions, thus reducing yield losses and harvesting costs, and improving fruit quality.

ISPS2019-252

The molecular dissection of the crop maturity trait in peanut

Srinivas Kunta*[1], Sarah Agmon[2], Yael Levy[3], Ilan hedvat[3] and Ran Hovav[3]

[1] Institute of Plant Sciences and Genetics in Agriculture, The Robert H. Smith Faculty of Agriculture, Food, and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

[2] Tel Aviv University

[3] Institute of Plant Sciences, The Volcani Center, Agricultural Research Organization, Bet Dagan, 50250, Israel

*Corresponding Author Email: kunta.srinivas@mail.huji.ac.il

Time to maturation is an important agronomic trait in peanut. Ranging from 90-180 days post planting (DPP) among different cultivars, this trait is important for adaptability and yield. However, not much is known regarding the genetic and molecular control of this trait. Here in our presented study, we tried to look intensely into the genetic control of this trait in the Virginia-type peanut background. Our study involved 243 recombinant inbred lines (F6:F8 RILS) derived from two closely related cultivars that differ in their maturity time. Lines were inspected in the field in a randomized block design. Pod maturity was evaluated at ~140 days post planting by pod blasting and visually categorizing pods based on their mesocarp color into three groups and scoring. Other important traits were recorded after the harvest such as total pod yield and harvest index. Trait mapping was performed by applying the new peanut Affimetrix SNP-array, containing ~2900 polymorphic SNPs among the RILS. Three QTLs were found for maturity explaining 0.08-0.17 of the total variation. Maturity was found to be significantly correlated with harvest index and iron deficiency based yellowing, demonstrating their involvement in the process. Also, maturity was associated with the branching habit (BH) trait, but with opposite to the phenotype of the parental lines. The genomic region for the BH trait was investigated further. We succeeded to narrow down the trait to 337.03kb on the chromosome 15 and found more recombinants with KASP SNP assay for further screening and identifying the gene. This study offers the first insight into the genetic control of maturity trait in Virginia-type peanut and serves as a preview for MAS.

ISPS2019-253

A Microtubule Associated ROP effector Modulates Abiotic Stress Response

Gil Feiguelman[1] and Shaul Yalovsky*[1]

[1] School of Plant Sciences and Food Security, The George S. Wise Faculty of Life Sciences, Tel Aviv University, Ramat Aviv, Tel Aviv 69978, Israel

*Corresponding Author Email: shauly@tauex.tau.ac.il

Microtubules (MTs) form a dynamic three-dimensional filamentous protein network with the ability to polymerize, depolymerize and reorganize in response to developmental and environmental signals. MTs dynamics regulate a range of cellular functions that are related to cell division, cell growth, cell shape formation, pathogen invasion, and abiotic stresses. MT dynamics is governed by Microtubule-Associated Proteins (MAPs), which regulate their nucleation, stability, cross-linking, severing, membrane interaction and orientation. My research focuses on a MAP named ICR2, a member of the plant specific family of coiled coil domain Rho of Plants (ROP) effectors designated ICR (Interactor of Constitutively active ROP). Genetic mutant analysis has implicated ICR2 in regulation of abiotic stress responses. During germination, *icr2* null mutants display decreased response to ABA, the GA biosynthesis inhibitor paclobutrazol, salt and osmotic stress. Mature *icr2* plants display increased sensitivity to drought stress. Interestingly, the *icr2* plants display decreased sensitivity to transpiration dependent salt stress. ICR2 association with MTs has been confirmed both *in vivo* and *in vitro*. *icr2* seedlings exhibit faster MTs re-organization in response to salt stress. In addition, *icr2* seedlings exhibit higher tolerance to a combined treatment of NaCl and the MT stabilizing drug Paclitaxel, implicating ICR2 in MTs organization and stability. To explore the possible functional redundancy between ICR2 and its closest homologs, I have created double and triple mutants in *ICR2*, *ICR3* and *ICR5* using CRISPR mediated genome editing. To study the function of ICR2 as a MAP, I have generated transgenic *icr2* mutant plants that express a fluorescent-tagged version of ICR2 under regulation of its endogenous promoter. In a yeast two-hybrid screen, we have identified novel interactors of ICR2 that are involved in cell wall composition and gene expression. Together, my studies implicate ICR2 in diverse cellular responses through its interaction with ROPs, MTs and additional effector proteins.

ISPS2019-254

Novel Probes for Elucidating Cell and Tissue Specific Activation Status of ROP GTPases

Noga Gershon[1] and Shaul Yalovsky*[1]

[1] School of Plant Sciences and Food Security, The George S. Wise Faculty of Life Sciences, Tel Aviv University, Ramat Aviv, Tel Aviv 69978, Israel

*Corresponding Author Email: shauly@tauex.tau.ac.il

ROPs (Rho of Plants) are the only signaling Ras superfamily small GTPases in plants. ROPs function as major regulators of cell polarity, cytoskeleton organization and dynamics, vesicle trafficking, hormonal signaling, biotic and a-biotic stress responses. To date, relatively little is known on the molecular pathways that regulate ROP activation and inactivation, primarily due to absence of appropriate markers. To address this deficiency, I have generated ratiometric fluorescent probes that would enable assessment of the activation status of ROPs in cell and tissue specific fashion. The probes are based on two ROPs effectors: ICR1 and RIC4, which following their interaction with activated ROPs are recruited to the plasma membrane. To alleviate potential toxic effects that could result from constitutive expression of ROP effectors, I am using an estradiol inducible system to express the ROP binding domains (RBD) of ICR1 and RIC4 rather than the full-length proteins. The system allows expression under the same promoter of either ICR1 or RIC4 RBDs fused to eGFP and their cognate ROP non-interacting mutants fused to mCherry. The expression of both ROP interacting and non-interacting probes under the same promoter, would be used for ratiometric imaging that would allow determining the relative levels of plasma membrane associated ICR1 or RIC4 RBDs. Determining the relative rather than absolute levels of plasma membrane associated probes should overcome potential artefacts that could result from differences in the expression levels. The probes were constructed in a modular system, which allows expression under regulation of a promoter of choice. Until now, I have generated transgenic plants that express the probes under the p*ROP2* and p*ROP11* promoters as well as epidermis and pollen specific promoters. The probes I have developed would enable a comprehensive analysis of ROP-mediated signaling as well as future screens for mutant and small regulating molecules analysis.

ISPS2019-255

Successful recovery of CRISPR-CAS9 mutations in melon disease resistance genes

Shahar Nizan*[1], Amalia Bar-Ziv[1], Katya Pashkovski[1], Tal Dahan-Meir[1] and Rafael Perl-Treves[1]

[1] Department of life Science, Bar-Ilan university

*Corresponding Author Email: perl.lab1@gmail.com

We have targeted three disease resistance genes in Cucumis melo L. for functional validation by CRISP-Cas9 mutagenesis. All three genes encode nucleotide binding-leucine rich repeat (NBL) proteins. Fom-2, cloned by Joobeur et al. (Plant Journal 2004) based on its map position, confers resistance to Fusarium oxysporum f.sp. melonis races 0 and 1. Fom-1 and Prv control resistance to Fusarium races 0 and 2, and to Papaya ring spot virus, respectively. They reside in a single locus in head-to head orientation, and were cloned in our previous study (Brotman et al. 2013). We used the Golden Braid cloning system to clone two guide-RNAs, the Cas-9 nuclease, and the NptII marker on a single binary plasmid. Transgenic melons from the appropriate resistant genotypes were regenerated, with high frequency of bi-allelic mutations in the target gene. We observed entire deletions of the region between the two targets, and even beyond that area. To our best knowledge, this is a first report of CRISPR mutants in melons. Plants were fertile, and their progeny will be tested for breaking the resistance phenotype, and also to test for a possible functional interaction between the two paired R-genes in the Prv-Fom-1 locus.

ISPS2019-256

Fine mapping of *PmG3M*, a powdery mildew resistance gene derived from wild emmer wheat

Zhen-zhen Wei[1], Ying-hui Li[1] and Tzion Fahima*[1]

[1] Institute of Evolution and the Department of Evolutionary and Environmental Biology, University of Haifa, Mount Carmel, Haifa, 3498838, Israel

*Corresponding Author Email: tfahima@evo.haifa.ac.il

Powdery mildew, caused by the parasitic fungus *Blumeria graminis f.sp. tritici (Bgt)* is one of the most destructive diseases of wheat. Wild emmer wheat (*Triticum turgidum ssp. dicoccoides*), the tetraploid progenitor of cultivated wheat, is a valuable source for novel disease resistance genes. A novel dominant gene, *PmG3M*, derived from wild emmer wheat, confers a broad-spectrum resistance to 55 powdery mildew isolates from Israel, Switzerland, USA, Chile, Paraguay, the Netherlands, and China. *PmG3M* was genetically mapped on the distal side of chromosome arm 6BL of wheat using segregating mapping populations produced by crossing the resistant *T. turgidum ssp. dicoccoides* (accession G305-3M) donor line with the susceptible *T. turgidum* ssp. durum (cv. Langdon). In the current study we saturated the distal region of chromosome 6BL harboring *PmG3M* with 33 new DNA markers (STSs, EST-SSRs, SSRs, CAPSs and KASPs) developed based on *Triticum aestivum* cv. Chinese Spring IWGSC RefSeq assembly v1.0 and wild emmer wheat Zavitan WEWSeq v.1.0. The *PmG3M* locus was mapped within a region of 0.41 cM, flanked by the two closest markers (0.18 cM proximal and 0.23 cM distal to *PmG3M*). Several candidate genes were predicted to reside within the target gene region, based on the annotated reference genome sequences. We transferred *PmG3M* into four Israeli bread wheat cultivars by marker-assisted selection (MAS) and obtained highly resistant BC_3 lines that can be used for wheat resistance breeding. Microscopic observations revealed a post-haustorial resistance mechanism, since the development of fungal haustoria is blocked at the haustorial bulb stage in the resistant G305-3M, 24 hours post inoculation, while in the susceptible Langdon the haustoria are developing normally. These studies emphasize the importance of wild emmer gene pool for improvement of cultivated wheat.

ISPS2019-257

Changes in Hydraulic Properties of Seminal Roots During Wheat Evolution

Elisha Hendel*[1], Guy Golan[1], Zvi Peleg[1] and Nimrod Schwartz[2]

[1] Institute of Plant Sciences and Genetics in Agriculture, The Robert H. Smith Faculty of Agriculture, Food, and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

[2] Department of Soil and Water Sciences, The Robert H. Smith Faculty of Agriculture, Food, and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

*Corresponding Author Email: elisha.hendel@mail.huji.ac.il

Root architecture influences the ability of plants to extract water from the soil, and it reflects plants strategies to survive under limited resources. During wheat domestication, the number of seminal roots increased (from 3 to 5), and consequently, seedlings lost their ability to recover from water stress. Moreover, this increase in the number of seminal roots was associated with a decrease in water movement. Here we analyzed the changes in the root hydraulic conductivity in domesticated and wild wheat. Using anatomical, physiological and computational modeling, we examine the changes in root hydraulic properties associated with wheat evolution under domestication. Anatomical characterization of the seminal roots in wild emmer wheat (Triticum turgidum ssp. dicoccoides) and domesticated durum wheat (Triticum turgidum ssp. durum) revealed that while the total root surface area was similar between both lines, the wild accession had significantly larger metaxylem elements. Thus, the calculated axial conductance of the wild plants was significantly higher (24%) as compared with the domesticated seedlings. To test the hydraulic properties of the roots, we analyzed 11-days-old seedling of wild and domesticated wheat using pressure chamber. In agreement with the anatomical results, the wild plants had higher hydraulic conductivity. We used the anatomical and physiological information as inputs to a numerical model for water flow in soil and root system (RSWMS). This model enables to study how the different root architectures of wild and domesticated wheat affect water state in the soil and in the plant. A construct realistic scenarios, representing different environmental conditions relevant to wheat were tested. Our results shed new light on the modification in seminal root architecture and hydraulic properties involved in wheat domestication.

ISPS2019-258

Analysis of novel transcribed regions responding to heat stress during Arabidopsis pollen development and maturation.

Nicholas Rutley[1], Gilad Luria[1], Laetitia Poidevin[2], Alejandro Ferrando[2], Jeffery Harper[3] and Gad Miller[1]

[1] The Mina and Everard Goodman Faculty of Life Sciences Bar Ilan University, 52900 Ramat-Gan, Israel

[2] Instituto de Biología Molecular y Celular de Plantas CSIC-Universidad Politécnica de Valencia, Valencia, Spain

[3] Department of Biochemistry and Molecular Biology, University of Nevada, Reno, MS330, Howard Building, Reno, NV 89557, USA

*Corresponding Author Email: gad.miller@biu.ac.il

Exposure to heat stress has a severe impact on plant fertility, with pollen (the male gametophyte) being acutely sensitive to elevated temperatures often leading to pollen abortion and collapse. Interrogating RNA-Seq data from A. thaliana pollen that matured during a 3-day heat stress regime (day maximum 37°C, night minimum 16°C) as well as pollen from control conditions, we identified 269 previously unknown genes originating from unannotated regions of the genome. 217 of these novel genes were recently annotated yet still remain completely uncharacterized. Cross-referencing our 269 novel genes with an independent pollen RNA-Seq dataset generated from chronic temperature stress (day maximum 40°C, night minimum -1°C), we found that the majority (~77%) were expressed. 34 of our 269 novel genes were significantly up- or downregulated in response to heat stress during pollen maturation. Transcripts from many of our novel genes contain short open reading frames and can be categorized as long non-coding RNAs (lncRNAs). A subset of transcripts is predicted to be targeted by miRNAs, and those transcripts targeted by miRNAs are more likely to be differentially expressed between control and heat-stressed pollen. 28 of our 269 novel genes are conserved in A. lyrata, and 3 are conserved within the Brassicaea family. Most recently, we have found that several of our novel genes are engaged with ribosomes in pollen germinated during heat stress. Our work is uncovering novel transcription in the male gametophyte during development and maturation, and generating targets for reverse genetics to dissect what role if any these novel transcribed regions play in pollen response to heat stress.

ISPS2019-259

PIFs as Mediators of Metabolic Control of the Circadian Clock.

Ayelet Kurtz[1], Raisa Poltavskaya[1], Ekaterina Shor[1] and Rahcel Green[1]

[1] Institute of Plant Sciences and Genetics in Agriculture, The Robert H. Smith Faculty of Agriculture, Food, and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

*Corresponding Author Email: ayelet.kurtz@mail.huji.ac.il

The role of the circadian system in regulating metabolism is fundamentally important. In plants, one of the most essential metabolic processes controlled by the circadian system is the timing and coordination of photosynthesis. Recently photosynthetic products, in particular sucrose, have also been shown to feedback and entrain circadian oscillators. Work in our laboratory has shown that in the model plant, Arabidopsis thaliana, the PIF (PHYTOCHROME INTERACTING FACTOR) transcription factor family is involved in numerous signaling pathways including sucrose signaling to the oscillator. Other laboratories have demonstrated a sucrose signaling pathway involving PRR7, a PSEUDO RESPONSE REGULATOR and key component of the circadian oscillator. pif mutants (pifQ) and prr7 mutants have both been found to be insensitive to the effects of sucrose on circadian rhythms. However, until now whether PIFs and PRR7 pathways interact and, more importantly how they mediate photosynthetic entrainment of the oscillator, is poorly understood. We suggest here mechanisms by which these pathways may regulated circadian rhythmicity. We also show that PIF activity is dependent on light quality; red and blue light have opposite effects indicating that PIF-regulated metabolite signaling is specific to certain times of day. Finally, perhaps surprisingly given the importance of metabolism and the circadian system, there is little known about how sucrose levels correlate with changes in the circadian system. We are collaborating to use GC-MS to measure precisely how different environmental conditions affect internal sucrose levels and observing how internal sucrose affects the clock. Our results give us valuable insight into how metabolism and the circadian system interact.

ISPS2019-260

The regulatory network connecting the COP9 Signalosome and glucosinolates biosynthesis and turnover

Mark Berliner*[1] and Daniel Chamovitz[1]

[1] School of Plant Sciences and Food Security, The George S. Wise Faculty of Life Sciences, Tel Aviv University, Ramat Aviv, Tel Aviv 69978, Israel

*Corresponding Author Email: markberliner@mail.tau.ac.il

Glucosinolates are sulfur- and nitrogen-containing secondary metabolites found in the Brassicaceae plant family. They are stored in an inactive form in the vacuole until herbivory, or any tissue damage initiates hydrolysis of glucosinolates yielding breakdown products that are toxic to herbivores and pathogens. Glucosinolate composition differs between species and plant developmental stage. Accordingly, plants must have discrete mechanisms to regulate the biosynthesis and activation of these chemicals. MYB and MYC transcription factors are considered master regulators of glucosinolate biosynthesis. We hypothesize that the COP9 signalosome, a highly-conserved protein complex that functions in the ubiquitin-proteasome pathway, regulates glucosinolate metabolism in Arabidopsis, contributing to the diversity and complexity of glucosinolate pathways. We suggest that COP9 signalosome regulates the stability of specific transcription factors (including MYB and MYC) that regulate glucosinolate biosynthesis. In the proposed project, I will determine the effect of perturbation of CSN function on glucosinolate accumulation and composition, and initiate studies to analyze the influence of the CSN on the stability of transcription factors.

ISPS2019-261

Heat stress partially rescue the phenotypes of csn5 hypomorphic mutant

Amit Kumar Singh[1], Shanmuhapreya Dhanapal[1], Alin finkelshtein[1] and Daniel A. Chamovitz*[1]

[1] School of Plant Sciences and Food Security, The George S. Wise Faculty of Life Sciences, Tel Aviv University, Ramat Aviv, Tel Aviv 69978, Israel

*Corresponding Author Email: dannyc@tauex.tau.ac.il

The COP9 (COSTITUTIVE PHOTOMORPHOGENIC 9) signalosome (CSN) signalosome is an eight-subunit protein complex which is conserved throughout evolution. Regulation of protein degradation by deneddylation of cullin-RING E3 ligase (CRL) is the most studied function of the CSN. Complete loss-of-function of any of the eight CSN subunit results in an essentially identical constitutive photomorphogenic phenotype, seedling lethality and anthocyanin accumulation. While most of the studies of CSN function have employed null mutants, here I used a genetic series of hypomorphic strains in order to understand the role of the CSN in regulating plant responses to the environment. Using phonemics techniques, we found that most mutants are hypersensitive to multiple stresses including UVC, salt, flooding and drought. However, *csn5a-1* thrives in terms of growth and yield under heat stress, growing better than under normal conditions. This increase in growth is not due to an increase in CSN dennedylation activity. Further molecular physiological analysis indicates that increase in growth is due to an effect of heat on auxin pathway such that heat rescues the auxin-deficiency in this mutant.

ISPS2019-262

Metabolic Regulation of Source-Sink Communication among Rice Species

Anuradha Singh[1,2] and Aashish Ranjan*[1]

[1] National Institute of Plant Genome Research (NIPGR), 110067, New Delhi

[2] French Associates Institute for Agriculture and Biotechnology of Drylands, Jacob Blaustein Institute for Desert Research, Ben Gurion University, Sede-Boqer Campus 84990, Israel

*Corresponding Author Email: aranjan@nipgr.ac.in

The yield potential of the crop which was raised during 'green revolution' is nowadays neither sufficient nor sustainable. To boost food production for a rapidly growing global population, optimized performance of source organs and carbon allocation patterning to improve sink strength (activity and size) is needed. In the present study, we systematically investigated the metabolic regulation of source-sink communication among *Oryza* species as it has a wide natural genetic diversity. A considerable variation was observed at individual genotypes for both source and sink characteristics. All the wild rice species had higher photosynthetic rate compared to most of cultivated *O. sativa* rice varieties. In contrast, cultivated rice species accumulated significantly the much higher amount of starch (~57.0 %) over wild species (~10.5%) in their sink organs. Wild rice species exhibited more biomass which was contributed by their increased plant height/growth, vigorous stem length, and longer, wider and thicker leaves including flag leaves. Similarly, cultivated rice variety possesses thick stems, short basal internodes, more productive tillers, panicle with a large number of fertile spikelet, to support the increased grain weight. The source organ of wild rice accumulated more photosynthate related metabolites including sucrose and sugars (glucose, fructose), suggesting their role to increase source strength. Conversely, the sink organs of cultivated rice have increased amount of hexoses, and fatty acids, suggesting their role in sink strength. The underlying genes of these metabolites showed increased expression pattern (by quantitative real-time PCR and RNA sequencing) in the source and sink tissues of wild and cultivated rice species, respectively. The knowledge gained from this study extends our understanding of metabolic regulation underlying optimized source and sink strength and have a great potential value for rice breeding with desirable source and sink features.

ISPS2019-263

Can singlet oxygen resilience be used as a marker for photosynthetic functioning of Nanochloropsiss oceanica strains in excess light environments?

Avraham Ben-Sheleg*[1], Nurit Novoplansky[1] and Avigad Vonshak[1]

[1] French Associates Institute for Agriculture and Biotechnology of Drylands, Jacob Blaustein Institute for Desert Research, Ben Gurion University, Sede-Boqer Campus 84990, Israel

*Corresponding Author Email: boomieschlagman@gmail.com

The desire to optimize *N. oceanica's* and other photoautotrophic organisms' growth both for the sake of production in industry and scientific understanding has put great importance on understanding the process leading to optimization of organisms to specific growth environments. Singlet oxygen ($_1O^2$) is an interesting candidate for study due to the strong consensus that singlet oxygen plays a role in photo-inhibition. Many studies have been conducted evaluating the exact role of singlet oxygen ($_1O^2$) on photo-damage and repair. In the present study, we focus on the question of whether singlet oxygen resilience causes a shift in optimal growth conditions. In order to address this question, two *N. oceanica* mutant strains, RB2 and RB113, both selected for high resilience to Rose Bengal (RB), a singlet oxygen ($_1O^2$) photosensitizer, were characterized and evaluated for robustness in high photon density environments.

Both of the RB resistant mutants showed higher photosynthetic capabilities when grown in high photon flux density environments relative to the wild type (WT) strain. This was evident by higher Pmax and photosynthetic efficiency of both the mutant strains relative to the WT strain when grown in a optimal temperature and an excess light environment (25^{o}c and 500 photons µmole sec^{-1} m^{-2}). This phenomenon was even more evident when excess light environments were accompanied by sub optimal temperatures (17.5^{o}c and 300 photons µmole sec^{-1} m^{-2}). These results led us propose that Rose Bengal resilience in photoautotrophic organisms could serve as a marker for selecting species or strains better suited for growth in high light environments.

ISPS2019-264

The carotenoid profile of fruits from the citrus germplasm collection in Volcani

Matat Zohar*[1], Lena Fidel[2], Livnat Goldenberg[2], Yosef Yaniv[2], Tal Arad[2], Ron Porat[2], Amir Sherman[2], Nir Carmi[2] and Tal Isaacson[1]

[1] Newe Ya'ar Research Center, Agriculture Research Organization (ARO), The Volcani Center, Ramat Yishay, 30095, Israel.

[2] Agriculture Research Organization (ARO), The Volcani Center, Rishon LeZiyon, Israel

*Corresponding Author Email: matat@agri.gov.il

Carotenoids are natural pigments found in green tissues and are essential for photosynthesis. In addition, different carotenoids accumulate in many plant tissues, providing them with red, orange and yellow colors, as well as distinctive aroma, since carotenoids serve as precursors of important volatiles. Moreover, carotenoids are indispensable in our diet and are known to protect against various chronic disorders. The color of the skin (flavedo) and flesh of citrus fruit depends mostly on the content and composition of the carotenoids in the tissues. The Israeli citrus germplasm collection, located at the Volcani center (Bait Dagan, Israel), includes about 200 different accessions from various species. In this work we have analyzed the carotenoid profile of both flavedo and juice of fruit from mandarins (*C. reticulate*), sweet oranges (*C. sinensis*), pomelo (*C. grandis*), grapefruit (*C. paradise*), lemon (*C. limon*), bitter oranges (*C. aurantium*), citron (*C. medica*), and their hybrids, all together about 140 accessions, using an HPLC system based on a C30 carotenoid column. The results show that different species accumulate different intermediates of the carotenoids biosynthesis pathway. For instance, the most dominant carotenoids in yellow grapefruits are the colorless phytoene and phytoflouene. In mandarins the most dominant carotenoids are beta-cryptoxanthin and violaxanthin and in oranges violaxanthin is the main pigment. The phenotypic data obtained in this project is used in a genomic analysis to detect genetic loci, in the various citrus species genomes, controlling carotenoid content and composition within the different accessions of a given species and among different citrus species. Isolation of genetic markers responsible for the differences in carotenoid profiles could be applied to assist in breeding of new citrus cultivars with desired skin and flesh colors and elevated nutritional value.

ISPS2019-265

The role of the autophagy mechanism in Arabidopsis plants under varying carbon levels

Sahar Magen*[1] and Tamar Avin-Wittenberg[1]

[1] Alexander Silberman Institute of Life Sciences, Edmond Safra Campus
The Hebrew University of Jerusalem

*Corresponding Author Email: sahar.magen@mail.huji.ac.il

Plants use light energy to fix carbon from the air via photosynthesis. The major form of transported carbon in the plant is sucrose, also used as a signaling molecule. During carbon starvation, plants suffer from growth arrest and utilize alternative energy sources, such as amino acids. In contrast, an excess of sucrose in plant growth medium affects photosynthetic efficiency by reducing key photosynthetic enzymes. Thus, carbon availability and homeostasis are important for plant growth and development.

Marcoautophagy (hereafter termed 'autophagy') is a conserved eukaryotic degradation mechanism. During autophagy, double-membrane vesicles, termed 'autophagosomes,' are formed, which target macromolecules or proteins for degradation and recycling in the cell's lytic organelle (vacuole in yeast and plants and lysosome in animals). The genes participating in autophagy are termed ATG genes, and many of their homologs were characterized in plants. Autophagy in plants is induced by carbon starvation, and knockout mutants of ATG genes (atg mutants) display increased sensitivity to starvation and decreased recovery capacity. However, working with constitutive mutants does not allow the separation between the role of autophagy during starvation and recovery. Additionally, the function of autophagy under sugar excess was never examined.

Our work aims to understand the function of autophagy during different carbon regimes. Firstly, we examined the role of autophagy during the progression of carbon starvation and recovery in Arabidopsis by evaluating autophagic activity and ATG gene expression. Moreover, we prepared a system of inducible knockdown lines, affecting ATG7 and ATG5. This allows us to downregulate autophagy specifically during starvation or recovery. We will examine the plants morphologically as well as analyze their metabolic content. Secondly, we are testing the effect of excess carbon on WT and atg mutant plants. We were able to see significant morphological changes in the root architecture of atg mutants compared to WT plants.

ISPS2019-266

Interdependent Nutrient Availability and Steroid Hormone Signals Facilitate Root Growth Plasticity

Eshel Elishav[1] and Sigal Savaldi-Goldstein[1]

[1] Faculty of Biology, Technion-Israel Institute of Technology, Haifa 3200003, Israel

*Corresponding Author Email: eshel.e@techniom.ac.il

plants acquire essential elements from an inherently heterogeneous soil in which phosphate and iron availability vary. Consequently, plants developed adaptive strategies to low iron and low phosphate levels including alternation between root growth enhancement and attenuation. How this adaptive response is achieved remains unclear. We have recently showed that phosphate and iron levels modulate the brassinosteroid signaling pathway. In turn, brassinosteroid activity controls iron accumulation in elongating cells. This interdependent interaction determines the extent of root elongation (Singh et al, Dev Cell, 2018). We currently expand our study and ask if this module also regulates root growth of arabidopsis and tomato roots at different developmental stages.

ISPS2019-267

Carotenoid accumulation in different stone fruits

Matat Zohar[1], Mark Jasinski[2], Keren Shemesh[1], Irit Bar-Ya'akov[1], Kamel Hatib[1], Doron Holland[1], Douglas Bielenberg[2], Ksenija Gasic[2] and Tal Isaacson*[1]

[1] Newe Ya'ar Research Center, Agriculture Research Organization (ARO), The Volcani Center, Ramat Yishay, 30095, Israel.

[2] Clemson University SC USA

*Corresponding Author Email: tali@volcani.agri.gov.il

Stone fruits (*Prunus* sp.), including peach, plum and apricot, exhibit wide range of fruit colors. Carotenoids are the main colorants contributing to their yellow and orange hues. Carotenoids are tetraterpene molecules produced by all photosynthetic organisms. They serve as pigments in many fruit and flower tissues, and some of their degradation products are volatiles, providing distinctive aromas. In addition, some carotenoids are essential components of our diet and carotenoids are known as agents helping in protection against various chronic diseases. We have characterized the carotenoid content and composition of stone fruits harvested from germplasm collections in Israel and the USA, representing wide genetic variability: ~120 apricot accessions and ~70 Japanese plum accessions from the Newe Ya'ar germplasm collection in Israel, and ~270 yellow peach accessions from collections in Clemson University and UC Davis in the USA. Our results demonstrate that although peach, apricot and Japanese plum are genetically closely related, their fruit accumulate very different carotenoids. While apricot accumulates large amounts of the first intermediates of the carotenoid biosynthesis pathway, such as phytoene, phytofluene and even cis-isomers of lycopene, as well as beta-carotene, Japanese plum contains mainly beta-carotene, and the main carotenoid found in peach fruit is Violaxanthin. an end product of the carotenoid biosynthesis pathway. The data obtained will be used in combination with genomic characterization of the different *Prunus* accessions, to better understand the genetic factors controlling carotenoid accumulation in stone fruit. Elucidating the causes of variability in carotenoid profiles within each species and the differences in the carotenoid composition among peach, plum and apricot, could help in breeding new cultivars with desired carotenoid profiles, having attractive colors and elevated nutritional value.

ISPS2019-268

The role of DNA-methyltransferases in regulating genes and transposons in the moss Physcomitrella patens

Katherine Domb[1], Aviva Katz[1], Rafael Yaari[1], Nir Ohad[1] and Assaf Zemach*[1]

[1] Department of Molecular biology and ecology of plants, Tel Aviv University, Ramat Aviv, Tel Aviv 69978, Israel

*Corresponding Author Email: katherined@mail.tau.ac.il

DNA methylation, the addition of a methyl group to cytosine base, is a prominent DNA modification in plants, found in all examined land plants. It can regulate genomes in different ways, including transcription, transposition, recombination, and chromosomal organization. Cytosine methylation is established and maintained by DNA methyltransferases (DNMTs). The moss P. patens encodes for two DNMT3 genes, which are closely related to mammalian DNMT3 and do not exist in A. thaliana, a single MET1 gene, a single CMT, and two DRM genes. The functions of these genes were revealed by analyzing genome-wide methylation landscapes and de-novo methylation profiles of an exogenous sequence introduced into P. patens genome in the respective mutants. De-novo methylation in CG and CHH contexts was found to be performed by PpDNMT3b, and the methylation of CHG sites by PpCMT. The maintenance of CG, CHG, and CHH methylation is almost entirely dependent on PpMET, PpCMT, and PpDnmt3b, respectively. DRM might target weakly euchromatic transposons. Here, we analyzed transcriptomic data of P. patens mutants in DNMT3, MET, CMT and DRM genes, as well as met/dnmt3b and cmt/dnmt3b double mutants. The loss of any DNMT other than DRM resulted in upregulation of transposable elements transcription, transposition, and up- and downregulation of gene expression, including genes involved in development, metabolic processes and environmental responses. The strongest effect was observed in cmt mutant, with over 800 differentially expressed genes, some of which might explain previously reported developmental defects. Interestingly, a total elimination of CG methylation in met mutant did not cause severe transcriptional changes. Our data further suggests that the level of total methylation rather than context eventually determines the overall methylation effect. We propose that CG methylation in angiosperm has greater effect than in moss due to differences in the level of crosstalk between CG methylation and non-CG methylation pathways.

ISPS2019-269

Improved berry quality of cv. Gewürztraminer by mediating radiation regime

Yaara Zohar[1], M. Westram[1], S. Rauchberger [2], Udi Gliksman[2], Nurit Agam[1] and Aaron Fait*[1]

[1] French Associates Institute for Agriculture and Biotechnology of Drylands, Jacob Blaustein Institute for Desert Research, Ben Gurion University, Sede-Boqer Campus 84990, Israel

[2] Teperberg winery

*Corresponding Author Email: yaara.zohar@mail.huji.ac.il

Recent climatic changes are affecting grape quality, making viticulture in arid and semiarid regions an increasingly relevant study model. While direct sunlight is a limiting factor in the production of high-quality grapes in temperate climates, winegrowers in arid regions face extreme radiation conditions, leading to berry surface temperatures of up to 50 °C in exposed berries [1]. Consequential oxidative stress, sunburn necrosis and browning, greatly reduce berry quality [2]. Adaptation of an adequate trellising system to the climate and cultivar is a simple and inexpensive way to control the radiation regime. This study aims to compare berry and wine quality of Gewürztraminer cv. grapes grown in the desert, that were trellised on the common VSP system or on SAYM, a trellising method with a higher canopy light interception [3]. In terms of quantity and quality, SAYM crop was characterized by a higher berry fresh weight and yield, higher titratable acidity and a lower pH. In 2018, a lower trend of sugar content was found as well. Also, SAYM trellising significantly reduced oxidative stress in 2017 berries, and a similar trend was repeated in 2018 berries. Sensorial analysis of the wines was conducted on the 2017 vintage, concluding that SAYM retains varietal characteristics (i.e. smell and taste) better, with a higher total score. A volatile compound analysis of the wines by GCMS has revealed a higher trend of monoterpenes in the VSP wines. Untargeted analysis of the wine data to identify possible off-flavors is in progress, as well as further analysis of the berries (UPLC, carotenoid content). This will result in a better understanding of the link between irradiance and metabolic processes and will stimulate the optimization of aromatic white grapes viticulture practices in arid/semi-arid regions.

ISPS2019-270

Overexpressed maturase proteins in *Chlamydomonas reinhardtii* leads to increased photosynthetic hydrogen production rate

Yael Feldman[1], Iddo Weiner[1] and Iftach Yacoby*[1]

[1] Department of Molecular biology and ecology of plants, Tel Aviv University, Ramat Aviv, Tel Aviv 69978, Israel

*Corresponding Author Email: iftachy@tau.ac.il

Hydrogen production by photosynthetic microalgae is a promising source of renewable fuel. During this process, water molecules split into protons and electrons by a gradual enzymatic process of oxidation. In turn, protons reduction into hydrogen is catalyzed by the enzyme hydrogenase (hyd). This photosynthetic process can be found in a variety of green algae but is missing in higher plants. A key step in this process is the reduction of hydrogenase by the electron carrier ferredoxin (fd) that solely links the electron transfer from photosynthesis to hydrogenase. However, since many other electron acceptors compete for ferredoxin's electrons – the natural hydrogen production rate is poor. To overcome this, a fusion between ferredoxin and hydrogenase (fd-hyd) was bioengineered and showed a significant ~4.5-fold increase in *in vivo* H_2 production rate compared to the native hydrogenase enzyme, likely as a result of better localization near the electron donor site, photosystem-I.

By using an optimization tool for heterologous expression in the nucleus, a *Chlamydomonas reinhardtii* fd-hyd nuclear overexpressing strain was built and named OP68. This mutant showed high levels of expression, but most of the protein pool was non-mature, and thus not active. In order to solve the fd-hyd maturation barrier, we aim to study the phenotypic differences when overexpressing maturases within the chloroplast of the prior engineered strain. Our study is focused in overexpressing three known maturases which are responsible for construction and insertion of the [Fe-Fe]-hydrogenase [2Fe] subcluster into the mature hydrogenase: hydE, hydF and hydG, each with a different role in the maturation process. Our preliminary results show the expression of each of the maturases results in an increase in hydrogen production rate.

ISPS2019-271

Image processing software for high throughput quantification of colony luminescence

Eyal Dafni[1], Iddo Weiner[1], Tamir Tuller[1] and Iftach Yacoby*[1]

[1] School of Plant Sciences and Food Security, The George S. Wise Faculty of Life Sciences, Tel Aviv University, Ramat Aviv, Tel Aviv 69978, Israel

*Corresponding Author Email: iftachy@tau.ac.il

Luminescent markers are widely used as reporters for various biologically interesting traits. In colony luminescence assays, the levels of luminescence around each colony can be used to compare the levels of a trait of interest between different strains, treatments, etc., using quantitative measurements of the luminescence. However, automatic methods of obtaining this data are lacking, making this a laborious manual process, especially when analyzing large numbers of colonies. In this work we develop an automatic, high-throughput tool for quantitative analysis of colony luminescence assays, which will allow fast collection of qualitative data from these assays and thus increase their overall usability. To examine the performance of this tool, we used an assay testing hydrogen production levels in unicellular algae. We achieved high correlation between the measurements from the identification tool and previous measurements of the algae, demonstrating this tool's potential for fast and reliable analysis of colony luminescence assays.

ISPS2019-272

The role of *Vernalization Insensitive Like* (*VIL*) genes in tomato development

Neta Kovetz*[1]

[1] Institute of Plant Sciences and Genetics in Agriculture, The Robert H. Smith Faculty of Agriculture, Food, and Environment, The Hebrew University of Jerusalem, Rehovot 76100, Israel

*Corresponding Author Email: Neta.kovetz@mail.huji.ac.il

The POLYCOMB REPRESSIVES COMPLEX 2 (PRC2) is a chromatin-remodeling complex that plays major roles in the transition between different plant developmental states. In Arabidopsis, PRC2 promotes flowering following cold exposure by repressing the flowering repressor gene *FLOWERING LOCUS C* (*FLC*) to ensure flowering in spring time, in a pathway known vernalization. VERNALIZATION INSENSITIVE 3 (VIN3) promotes flowering in Arabidopsis by recruiting PRC2 to the *FLC* locus. In tomato (*Solanum lycopersicum*), there is no *FLC* orthologue and the transition to the reproductive stage is autonomous. Nevertheless, the tomato genome contains three VIN3-LIKE homologs: SlVIL1, SlVIL2 and SlVIN3. The tomato *crawling elephant* (*crel*) mutant was isolated in a genetic screen for suppressors of the *entire* (*e*) mutant, which has simple leaves. *CREL* encodes a VRN5/VIL1 ortholog. *crel* mutants suppressed the *e* simple leaf phenotype, and single *crel* mutants show a dramatic increase in leaf complexity. In addition, *crel* mutants show a number of aberrant developmental phenotypes such as a "crawling" stem, sterility and delayed organ formation. To further understand the role of *VIN3-LIKE* family in tomato, we generated CRISPR/Cas9-derived mutants in two additional members of the *SlVIL* family. Like *crel, slvil2* mutants display multiple alterations in plant development, such as round cotyledons, distorted leaf pattern, short internode and retardation in anthesis time. Homozygous mutations in *slvin3*, the third member of VIL family, did not display a noticeable aberrant phenotype, though further work is required to characterize this mutant. These results indicate that VILs play broad roles in tomato development, and the research is expected shed light on these roles.

ISPS2019-273

Monitoring the Chloroplast Glutathione Redox State in High Temporal Resolution during a Diurnal Cycle and under High Light and Fluctuating Light Conditions

Zechariah Haber[1] and Shilo Rosenwasser*[1]

[1] The Robert H. Smith Institute of Plant Sciences and Genetics in Agriculture, The Hebrew University of Jerusalem, 7610001, Rehovot, Israel.

*Corresponding Author Email: shilo.rosenwaser@mail.huji.ac.il

Plant exposure to high light can result in over-reduction of the photosynthetic machinery and accumulation of harmful reactive oxygen species (ROS), due to the incompatibility between the strong irradiance and the relatively lower capacity of photosynthesis. To avoid photodamage, various mechanisms allowed plants to adapt to dynamic light environments. Fluctuating light further exacerbates the stress due to the need to constantly adjust metabolite levels, gene expression and protein activity. Electrons from the Glutathione (GSH) pool are used to detoxify ROS, leading to oxidized glutathione and a shift in the glutathione redox potential (EGSH .(A redox-sensitive GFP (roGFP) has recently been implemented as an *in vivo* ,non-destructive and compartment-specific biosensor of EGSH.

Our main objectives were to elucidate chloroplast-specific redox fluctuations under normal and stressful light conditions in high spatiotemporal resolution and to correlate the E_{GSH} to commonly used photosynthetic parameters.

We monitored the chloroplastic roGFP oxidation degree in Arabidopsis plants during a diurnal cycle with a six-hour high light or fluctuating light period and this was coupled with chlorophyll fluorescence measurements.

Shift of plants from normal growth light to high light (above 750µE) resulted in a decline in *Fv'/Fm'* and roGFP oxidation, followed by gradual reduction. In addition, peaks of high roGFP oxidation during day-night transitions and significant reduction at nightfall followed by gradual oxidation during the night were observed. Shift of plants from normal growth light to fluctuating light resulted in a similar reduction in *Fv'/Fm'* and an oscillating roGFP oxidation with different responses for the different frequencies.

Further analysis of the high light results suggests two different states of the chloroplast antioxidant machinery. Measuring the chloroplast EGSH under fluctuating light conditions revealed frequency-dependent EGSH alterations. We believe that high-resolution monitoring of organelle specific EGSH can provide new insights into light acclimation strategies.

ISPS2019-274

Overexpression of the ribosomal S30 subunit or NAD-dependent formate dehydrogenase leads to indole-3 –carbinol tolerance in Arabidopsis thaliana.

Alin Finkelshtein*[1], Luu Anh Tuan[1], Daniel A. Chamovitz[1]

[1] School of Plant Sciences and Food Security, Tel Aviv University, Tel Aviv, Israel.

*Corresponding Author Email: Alinfin@gmail.com

Indole-3-carbinol (I3C) is a hydrolysis product of indole glucosinolate and is formed in response to tissue rupture. I3C is toxic molecule to herbivorous insects and pathogens. In mammals, I3C is extensively studied for its properties in cancer prevention and treatment. Endogenous functions of I3C were recently discovered in plants where I3C acts as a development regulator of plant growth. In the Arabidopsis model system, I3C reversibly inhibits root elongation in a concentration-dependent manner by interfering with auxin signaling pathway. So far little is known about the mechanism of I3C as a signaling molecule in plants. The aim of this work is to discover and characterize the pathways that involve I3C as a signaling molecule in plant. Two putative I3C-tolerant strains, I3CT1 and I3CT3, were identified during screening a Full-length cDNA Over-eXpression library (FOX) for root elongation when exposed to high concentrations of I3C. I3CT1 carries the AT2G19750 gene which encodes a S30 ribosomal protein, and I3CT3 contains AT5G14780, which encodes NAD-dependent formate dehydrogenase (FDH). Overexpression of S30 gene, and not the knock out of the underlying gene, causes tolerance to I3C. I3CT1 is less affected and recovers faster from exogenous I3C than the wt. The tolerance is specific to I3C, since I3CT1 did not exhibit any pronounced tolerance to other indol or benzoxanoids molecules tested. I3C acts as an auxin antagonist both in Col-0 and in I3CT1 mutant. S30 loss of function mutants do not have any phenotype due to redundancy and cover up expression of two homologous genes. However, amiRNA (blocking expression of all three homologous genes) caused a severe developmental delay as well as defected flower structure and reduced productivity, implying S30 has a developmental function. Transcriptome analysis hints that the tolerance may be due to a priming effect of the over-expression of the gene.

ISPS2019-275

Elucidating the mechanism of Aphid resistance in Teff

Nathan Mante Gyan*[1] and Vered Tzin[1]

[1] French Associates Institute for Agriculture and Biotechnology of Drylands, Jacob Blaustein

Institute for Desert Research, Ben Gurion University, Sede-Boqer Campus 84990, Israel

*Corresponding Author Email: gyan@post.bgu.ac.il

Plant-insect interactions have led to many defense mechanisms from either the host or the insect to maximize their potential from the interactions. Some defenses are constitutive while others are induced although the insecticidal defense compound or protein classes are often similar. These specialized plant defense compounds may repel insects, while defense proteins often interfere with their digestion (Fürstenberg-hägg, et al., 2013). Aromatic amino acids are part of the precursors for secondary metabolites including alkaloids, phenylpropanoids, flavonoids, and lignin (Kessler & Baldwin, 2002); hence, aromatic amino acids also have played a role in plant defense. *Eragrostis tef* is an annual cereal grass, a C4 plant and is an intermediate between a tropical and temperate grass. Teff, as nutritious and gluten-free grain, is gaining economical interest worldwide. The major chemical defense metabolites in Teff is yet unknown. The main objective is to identify the metabolic pathway of Teff against different pest and to characterize the local pest in Israel. We hypothesize that Teff plants induce Trp-derived secondary metabolite in defense against the main cereal pest (*Rhopalosiphum padi*) and Locust.

ISPS2019-276

The biosynthesis of mango (*Mangifera indica*) fruit aroma volatiles

Matan Mire*[1], Einat Bar[1], Rachel Davidovich-Rikanati[1], Moshe Inbar[3], Amir Sherman[2], Yuval Cohen[2] and Efraim Lewinsohn[1]

*Corresponding Author Email: miurematan@gmail.com

[1]Department of Vegetable Crops, Newe Yaar Research Center, Agricultural Research Organization, The Volcani Center, Israel.

[2]Department of Fruit Trees Sciences, Institute of Plant Sciences, ARO, The Volcani Center, Israel.

[3]Department of Evolutionary and Environmental Biology, University of Haifa, Israel.

A wide variety of volatiles impart unique aroma and flavor of fruits. Volatiles can be grouped by biochemical origin. The largest and the most varied of these groups are the terpenes. Terpene synthases (TPS) are key enzymes in the terpene biosynthetic pathway and are responsible for converting phosphorylated prenyl-chain precursors into terpene skeletons. Another group of volatiles, which have a significant effect on the aromas of fruit, are the esters. The final stage in ester biosynthesis is catalyzed by alcohol acyltransferase (AAT) enzymes that transfer an acyl group to an alcohol substrate forming esters. Mango (*Mangifera indica*) fruit is extremely rich in volatiles. More than 300 volatiles have been identified in mango fruit and their interaction with sugar and acids creates the typical mango's flavor and aroma. The study of the formation of mango volatiles is limited and mainly restricted to descriptions of profile of volatiles in a number of mango varieties. In the research described here, we characterized the volatile profiles of different varieties of mango grown in the collection of the Volcani Center. Preliminary results in these examinations indicate a great difference between species originating in India in which a wide variety of volatiles was found in compared to cultivars grown in western countries that contain relatively fewer volatiles. A number of candidate genes similar to known TPS and AAT from other plants were detected in fruit's mesocarp transcriptome and were cloned into bacteria in an attempt to identify their functionality. The functional identification of these genes, will allow better understanding of the biochemical and genetics of aroma volatiles formation in mango fruit. These genes can be used as genetic markers in the future in order to optimize the segregation of desirable aroma traits for use in the selection processes to attain mango cultivars with desirable aroma.