Report of the first meeting of the COST Action FA1006

PlantEngine I: Current State and vision for the future

The first annual meeting of the COST Action FA1006 was held 17.-18.11.2012 in Murcia, Spain. It included WG1-WG3 sessions as well as a MC meeting. As outlined in the MoU and in the welcome talk by the Chair of the Action, **Heribert Warzecha**, one of the major tasks of this COST Action is the assessment of the status quo of plant natural product research in Europe and the provision of a research network for strengthening collaborative research in Europe.

A total of 95 scientists attended the meeting, both senior as well as early stage researchers. They represented 20 countries (Argentina, Austria, Belgium, Czech Republic, Denmark, Finland, France, Germany, Greece, Israel, Italy, Norway, Poland, Portugal, Romania, Spain, Switzerland, The Netherlands, Turkey, United Kingdom), from academic institutions and also from five companies (Icon Genetics GmbH, Alkion Biopharma, ExPlant Technologies BV, Viveros Bermejo Biotech Division, Phytowelt GreenTechnologies GmbH).

To give an initial overview of the current research and funding schemes of Framework Program 7 (FP7) with focus on PNP research, the **WG1 session** featured presentations by coordinators of past or currently running FP7 collaborative research projects.

The opening talk was given by Dr. **Tomasz Calikowski**, Scientific Officer in the European Commission´s Biotechnologies unit. He pointed out that in the current KBBE funding scheme Theme 2 “Food, Agriculture and Fisheries, and Biotechnologies”, 27% of all projects in the portfolio were related to the field Plant (green) Biotechnology. While there is already a significant participation of industry and SMEs in the current projects, the 2013 call (currently in preparation) will offer an even higher support to SMEs and support the transition to the next framework program, termed “Common Strategic Framework Horizon 2020”.

The large integrated collaborative project “*SmartCell- rational design of plant systems for sustainable generation of value-added industrial products*” was introduced by the coordinator, **Kirsi-Marja Oksman-Caldentey** (VTT, Finland). *SmartCell* brings together 14 leading European academic laboratories and four industrial partners in order to create a comprehensive knowledge base of how secondary metabolite biosynthetic pathways operate in plants. The case study component i.e. manufacturing a valuable terpenoid in an optimized large-scale system gives SmartCell a unique opportunity to directly make transition from fundamental science to application. For long-term exploitation an integrated database, compound library, cell culture collection and a genebank are established.

METAPRO, presented by the coordinator **Paul Fraser** (UK), also has a focus on isoprenoids, with the lead compounds astaxanthin and crocin. The consortium is comprised of eight partners and activities in the METAPRO project are focusing on; (i) utilising systems biology to provide a better understanding of isoprenoid formation and its interaction with intermediary metabolism, (ii) elucidate and utilise mechanisms

of post-transcriptional regulation, (iii) alter organelle parameters to optimize sequestration and synthesis, (iv) improved the stability of the engineered metabolites in the plant cell and (v) implement novel engineering approaches.

TERPMED, another small-scale collaborative project, was presented by **Alain Tissier** (Germany). Again, with terpenoids as main focus, this project pointed sesquiterpene lactones (parthenolide) and phenolic diterpenes (carnosic acid and rosmanol) as their lead compounds for investigations. The project partners focus on pathway elucidation, metabolic profiling and metabolic engineering as well as on screening for alternative sources and pharmacological testings.

As a recently finished project, FLAVO was presented by **Stephan Martens** (Italy). With the health-promoting effects of several classes o flavonoids in their focus the project participants aimed to develop methods to define the optimal dose of flavonoids in foods for providing benefits to human health, to develop approaches for selection and transformation of plant raw materials to optimise the flavonoid composition and quality in foods, and to develop tools for growing, analysing, selecting, and engineering fruits and vegetables with optimised flavonoid content.

From the presentation it became clear that on an European level there is a strong effort to support research directed to green (bio)technology and that regarding PNP the currently most advances field is that of terpenoids. As a COST Action, benefits can be taken from the progress achieved so far on that field, e.g. in building on the so far achieved knowledge of pathway engineering, metabolomics approaches, data mining and storage. However, even with terpenoids as a putative proof of principle in metabolic engineering, it should also be focus to broaden the scope to other compound classes.

The WG2 session was devoted to presenting and discussing the most recent results achieved by the research groups joining COST Action FA1006 on developing new molecular tools and applying different metabolic engineering and biotechnological strategies to enhance and/or diversify bioactive and economically important PNPs.

The session included seven selected oral presentations, covering different aspects of plant molecular engineering, i.e. industrial applications of large scale plant tissue culture and use of somatic hybrids to produce large biomass for the extraction of bioactive plant metabolites (**Peter Welters**, Germany), development of combinatorial biochemistry in plants to create novel PNPs with new or superior biological activities (**Alain Goossens**, BE), identification of a glandular trichome-specific promoter, useful for engineering PNPs that are generally accumulating in such specialized organs (**Peter Brodelius**, Sweden), role of microRNAs in regulating expression of biosynthetic genes (**Turgay Unver**, Turkey), transient expression and metabolomics to evaluate metabolic engineering (**Harro Bouwmeester**, NL), potential bottlenecks of metabolic flux during lipid biosynthesis (**Brigitte Thomasset**, France) and the use of genome wide homology searches to characterize biosynthetic steps (**Antje Feller**, Italy). The session also comprised 25 poster presentations, reporting progress on metabolic engineering of crops as well as medicinal or industrial plants.

General discussion was focused on defying the main goals that WG2 would achieve in the first year:

A. Completion of collecting information on the resources and molecular tools available in the laboratories of the partner groups, by filling the form (annex 1), already sent to all the partners by the WG2 leaders. The form has to be returned to the WG2 leaders by February 29th, 2012.

B. Implementation of a database, available on the web site of PlantEngine (www.plantengine.eu), aimed at sharing and exchanging the molecular tools already available or new tools that will be developed in the partner laboratories and providing as well information on the targeted metabolic pathways, end-products and plant species. The website should provide restricted access to the partners of the COST Action FA1006, will be interactive allowing the extraction of specific information, with possible links to other COST Actions on the same topic (e.g. FA0804 Molecular Farming). The database will function also as a basis for new scientific collaborations and interdisciplinary approaches for problem solving.

C. Need to focus on research on the molecular mechanisms regulating transport of plant secondary metabolites and governing the spatial distribution of the enzymes, cofactors, and substrates that mediate biosynthesis of secondary metabolites within the plant cell. This knowledge will be critical to develop new tools to engineering efficiently the biosynthesis of PNPs, in parallel with the continuous discovering of potential biosynthetic genes coming out from the sequencing of plant genomes. It was discussed also the possibility to organize in 2013 a theoretical-practical course on "Compartmentalization and Molecular trafficking in Plant Secondary Metabolism", to give the opportunity to young scientists to be acquainted with the latest development in this forefront field for plant metabolic engineering.

The third session dedicated to **WG3 "System engineering approach"** comprises one key note lecture, two 30 min lectures and six shorter communications. In addition there was a presentation of 19 posters related to the WG3 topic. The key note lecture was delivered by **Gad Galili** who described his extensive study on plant regulatory aspects on the level of gene expression and metabolism related to environmental changes such as stress as well as some hormonal and nutritional cues. In this context he presented a bioinformatic approach developed by his group, which combines metabolites changes with gene expression and transcription factors in response to stress**. Kay Hamacher**, the second speaker, focused on the use of computational biology in the sophisticated modeling of intracellular networks based on stochastic approaches, which can have great impact on our COST Action particularly when it comes to engineering synthetic circuits and networks. The third invited speaker, Elefterios **Pilalis**, presented the construction of an *in silico* compartmentalized model of *Brassica napus* central metabolism able to perform an exploration of the whole steady state flux space.

The shorter presentations included a systemic study concerning the regulation of the production of trans-resveratrol by grapevine cell cultures based on molecular mechanisms such as early signaling pathways (**Belchi-Navarro**), as well as a proteomic analysis of cyclodextrin and methyl jasmonate mediate resveratrol accumulation in the same cell culture (R. **Bru-Martinez**). Cyclodextrins and methyl jasmonate were also used in the investigation of the early signaling network in tobacco cell (L. **Almagro**). A study on the genes involved in the carotenoid pathway based on a genome-wide QTL approach was presented (**M.Taylor**), while the relationship of taxane production and the genes involved in taxol biosynthesis was proposed by the extensive work of **M. Onrubia *et al*.** Two presentations were focusing in the development of tools important in systemic research as ^{1}H-NMR use for the studies for the variety selection of flaxseed based on metabolomics (F. **Mesnard**) and the temporary immersion system for contained plant propagation and biomass production (**C. Wawrosch**).

On the same line of the production of compounds with biological interest based on genomic and metabolomic approaches, were the posters presented focusing in taxol, resveratrol, psoralen, antioxidants and flavonoids, rosmaric acid, volatile phytochemicals, antocyanins, carotenoids, etc. as well as the development of methodological tools as ^{13}C labeling –GC/MS, or HPLC-ESI-MS-TOF. There was a general opinion that posters were of high quality and that the next time they must have more special attention.

The oral presentations were followed by a very interesting discussion between the speakers and the audience leading to a proposal for the future organization by our COST Action of “summer schools”. The main points of the discussion could be summarized as follows:

A. **Computational and Biological approach**. It is important to introduce computational tools in the systems under investigation in order to understand them better and to engineer them for the optimization of their productivity, based on their global study. More specifically, the goal of the systemic approach will be to correctly predict and engineering the behavior of biological systems, plants and their parts in our case. This requires that we will be able to model the responses of all the elements in that systems using genetic or environmental perturbations, accurate monitoring of the global responses to these changes at the level of genes, proteins, informational pathways, and phenotypes and the integration of these quantitative measurements into a dynamic data structure.

B. **A proposal for the organization of summer schools on “plant system biology”.** It is profound that there is a need for scientists coming from different disciplines as biologists, chemists, engineers, computer scientists, etc. to collaborate and establish a common language of communication. In order to facilitate this communication it will be important to organize in the frame of our COST action a series of “summer schools” for young researchers, for instance for experimental biologists who would like to learn how to use computational modeling to supplement their experimental studies and for computer scientists to have a better view of the problems and difficulties of the biological experiments. *These summer schools could foster the development of*

interdisciplinary research and develop a cohort of plant systems biologists in our action for future collaboration and increased uptake of quantitative methods in plant science.

Overall, the very intense discussions during the sessions and along with the posters showed that this COST Action has a very high potential to initiate new research collaborations on a European level.