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**FROM  
MOLECULES  
TO LIVING  
SYSTEMS**



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# 44th FEBS Congress

From Molecules to Living Systems

July 6–11, 2019

Krakow, Poland

Abstracts submitted to the 44th FEBS Congress, taking place in Krakow, Poland from 6th to 11th July 2019, and accepted by the Congress Organizing Committee are published in this Supplement of *FEBS Open Bio*. Late-breaking abstracts are not included in this supplement. The abstracts are available as two PDF files: Talks (Plenary Lectures, FEBS Special Sessions and Symposia) and Posters.

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\* Each poster has been given a unique number beginning with the letter P; the next part relates to the session in which the poster will be presented.

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myricetin, isorhamnetin, kaempferol, catechins and leukocyanins. This group of biologically active substances is also responsible for the choleretic, diuretic, anti-inflammatory actions. Moreover, studies have also proved their antimicrobial action against bacterial and fungal biofilms. Consequently, it is advisable to develop a method of quantitative determination for these biologically active substances. In this study, we analyzed the content of flavonoids in vitamin mixtures No. 1 – *Fructus Rosae: Sorbus aucuparia* (50:50) and No. 2 – *Fructus Rosae: Ribes nigrum* (50:50) and their components. We established the importance of pH value for the complexation reaction of flavonoids with aluminum chloride. A 1.56–2.25 pH value during the complexation reaction contributes to an increase in the selectivity of the reaction on the ketone group of ring C and the hydroxyl group of ring A of flavonoids. We also determined the possibility of identifying the *Fructus Rosae* and vitamin mixture No. 1 by spectral characteristics: 323, 404, 526 nm and 311, 414, 540 nm, respectively. The kinetics of the complexation reaction for the vitamin mixtures, individual fruits of *Rosae*, *Ribes nigrum*, *Sorbus aucuparia* and infusion from the vitamin mixtures was also analyzed. The optimal time of complexation was 40–50 min. For the infusion, this process is longer, since it is competing with hydrolysis, and, therefore, requires more time to establish equilibrium. We have developed a method for spectrophotometric determination of total flavonoid content. The method was tested and validated for *Fructus Rosae*, *Ribes nigrum*, *Sorbus aucuparia*, vitamin mixtures and their infusion. The publication has been prepared with the support of the “RUDN University Program 5-100”

#### P-28-023

##### ICML – algorithm for gene classification from different species by expression patterns

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Over the past decade, the development of high-throughput sequencing technologies has resulted in the accumulation of a large number of genome sequences for a large amount of species. As a result, there is a great need for functional annotation of these new sequences. Functional annotation of genes in most cases is carried out by searching orthologs in the nearest species and transfer to them functional data of orthologs. The search for orthologs is based on sequence similarity; there a variety of tools for orthology assessment such as OrthoFinder, OMA, OrthoMCL, and many others. However these methods have limitations especially in large and complex genomes shaped by whole genome and segmental duplications. Also, despite widely accepted assumption that the orthologs share the same function, this is not always the case. In order to improve the accuracy of the transfer of functional annotation from model species to non-model ones, it is necessary to use additional information, such as the gene expression profiles. We developed ICML (Interspecific Classifier based on Machine Learning) – the approach that allows classification of genes according to their expression profiles between two species. The main problem that arises when comparing expression profiles in species with differing morphologies and/or developmental rates is the inability of direct matching of samples. This makes it impossible to use standard metrics (for example, Euclidean distance) for the assessment of the similarity

of expression profiles. Thus we developed a new metrics based on machine learning methods. a proof of concept, we performed the comparison of the genes of two plant species belonging to distant taxonomic groups – *Arabidopsis thaliana* and *Fagopyrum esculentum*. We expect that the wider application of this algorithm will increase the accuracy of functional annotation in complex genomes. This research was funded by the Russian Science Foundation, grant number 17–14-01315.

#### P-28-024

##### C-terminal modification of TMV coat protein allows assembly of chimeric particles in plants suitable for “green” biotechnological applications

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Recombinant viruses based on cDNA copy of Tobacco mosaic virus (TMV) are useful for accumulation of chimeric particles carrying different immunogenic epitopes in plants. Earlier our group created vectors expressing conserved M2e and fp short antigens (23 and 14 aa) from Influenza A on the surface of TMV virions (Petukhova *et al.*, 2013, 2014; Gasanova *et al.*, 2016). Chemical conjugation of larger proteins to the particle could be efficient method to improve vaccination strategy. For this we constructed TMV genome containing additional reactive lysine at the C-terminus of coat protein (CP). Inserted sequence including lysine was optimized according to plant and viral codon usage. Agroinfiltration of *Nicotiana benthamiana* leaves led to delayed systemic spread of TMV-C-lys with symptoms different from the wild-type (wt) infection. We observed deformation (curling) of upper leaves (7–10 days post inoculation, d.p.i.) as well as necrotic lesions of stem and petioles of lower leaves (17–19 d.p.i.). Unlike TMV-wt and vector TMV-M2e-ser (Petukhova *et al.*, 2014) plant growth and development were not disturbed. Modified particles were successfully purified from non-inoculated leaves (21 d.p.i.). Electron microscopy of preparations demonstrated that shape of TMV-C-lys virions varied from rigid to slightly curved rods (300–700 nm in size). The resulting particles can be a perspective platform for chemical conjugation (1-Ethyl-3-(3-dimethylaminopropyl)carbodiimide/ N-Hydroxysuccinimide) with broad spectrum of Influenza (M1, NP) and other peptides and proteins. Stable complexes consisting of lengthy carrier and foreign protein(s) should be recognized by antigen-presenting cells (APC) such as dendritic cells and macrophages.

#### P-28-025

##### Effects of short-term drought on metabolism and nutritional properties of pea (*Pisum sativum*) seeds

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Drought is one of the major environmental stressors, affecting crop productivity. Plant response to dehydration is accompanied with metabolic changes, oxidative stress and enhanced generation of advanced glycation end products (AGEs), which are known to



be pro-inflammatory in mammals, impacting on the pathogenesis of diabetes mellitus and ageing. The protein-rich seeds of legumes might be a target of such deleterious changes. However, the effects of drought-related protein damage on the quality of crops, as well as its influence on human health are still unstudied. Therefore, here we address the changes in seed quality, metabolome, protein AGE contents and nutritional properties of pea accompanying short-term drought, applied at the step of seed maturation. For this, pea plants were grown on vermiculite for 6 weeks before transfer to aqueous medium, to which 5 days later polyethylene glycol 8000 (PEG-8000) was supplemented. Two days later, the plants were transferred to PEG-free substrate and grown till the end of seed maturation. After evaluation of seed quality, seed metabolites were analyzed by GC- and LC-MS. Modulation of inflammation was evaluated with corresponding exhaustive enzymatic hydrolyzates in the model of human neuroblastoma cell line SH-SY5Y. For this, dynamics of proteins involved in inflammation-related signaling was assessed by Lumines xMAP multiparametric immunofluorescence technology. In parallel, profiles of individual AGEs were addressed in the same hydrolyzates by LC-MS/MS. Although no effect on AGE profiles was observed, clear alterations in primary metabolome and regulatory response were observed. This work is supported by the Russian Science Foundation (Grant No. 17-16-01042).

### P-28-026

#### Characterization of the tomato P450-72 (SIP450-72) and analyze its function in tomato plant

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Plant P450s are necessary for the catalysis of many major reactions, typically in production of primary and secondary metabolites. The functions of these P450s involve the biochemical synthesis pathways include of terpenoids, fatty acids, lipids, as well biosynthesis of plant hormones. Also, they involve in many reactions that synthesis the metabolites which are related to defense mechanism. Plant P450s play such important roles in plants, and there are hundreds of genes in one plant. Tomato plants also contain 300 – 400 CYP genes in their genome, however many of them are unknown their function. The *SIP450-72* gene in CYP736A subfamily, which is not yet revealed its function in tomato, was isolated from tomato. To determine enzymatic character of the *SIP450-72*, it was heterologously expressed in *E. coli* and purified from them. The *SIP450-72* enzyme was analyzed its enzymatic activity and used for the substrate screening. In this study, we found out that the *SIP450-72* enzyme catalyze hydroxylation of resveratrol. To investigate its function in tomato plant, we developed the *SIP450-72* over-expressed transgenic tomato plants, and analyzed them.

### P-28-027

#### Comparison of the CaCPR1 and CaCPR2 expression pattern in hot pepper

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The cytochrome P450 is the largest enzymatic protein family in plants and they plays important roles in development and defense system in plant. For P450 reaction, NADPH-cytochrome P450

reductase (CPR) required for electron transfer from NADPH to cytochrome P450. There are two CPR genes in the hot pepper genome which are *CaCPR1* and *CaCPR2*. The CaCPRs expression levels were quantitative by real-time PCR in various hot pepper tissues and under stress conditions such as JA treatment, SA treatment and drought condition. The *CaCPR1* expression level was gradually increased during fruit ripening. However, the *CaCPR2* gene was constitutively expressed in all tissues but the expression level was lower than the *CaCPR1*. Under the stress conditions, both of the *CaCPR1* and *CaCPR2* expression levels were increased, the *CaCPR1* expression was maintained high level compared to the *CaCPR2*. These results suggest that the CaCPR1 is major enzyme during plant development and under stress condition. In addition, under stress condition, both enzymes are increased because more enzymes are needed against stress. To investigate the enzymatic properties, two *CaCPRs* were isolated from hot pepper (*Capsicum annuum* L. cv. *Bukang*) and heterologously expressed in *Escherichia coli*. The enzymatic activities were assessed using protein and chemical substrates such as MTT and ferricyanide. \*The authors marked with an asterisk equally contributed to the work.

### P-28-028

#### D/H modification of plant peptides and microelements metabolome

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The aim was to investigate changes in the metabolism of peptides and essential microelements in plants at various D/H ratios in water. Before D/H control of chemical kinetics in water solutions under low deuterium concentrations were investigated and summarized in details for molecular systems and living cells. Peptide analysis was performed on a time-of-flight mass spectrometer, equipped with a UV laser. Zinc was analysed in plants by atomic absorption spectrometry with electro-thermal atomization and Zeeman effect and X-ray fluorescence energy dispersive. Water samples: deionized high ohmic water, (BD, D/H=140 ppm), Deuterium depleted water (DDW, D/H=12 ppm). The germination of moss in waters with different isotope ratios led to the change in the qualitative composition of proteins. A total of 300 proteins were determined. 58% of them were observed both in BD and DDW. 13% were observed only in DDW, and 29% were only in the water with a natural D/H isotope ratio. This change leads to differences in metabolism in the plant as a whole. For example, in growing plants (*Capsicum annuum* L.) in DDW, the zinc content increases by 1.5 times compared with BD. In addition, the technology of essential microelements enrichment in the leaves *Callisia fragrans* L. at different ratios of hydrogen isotopologues in water has been developed. In 12 days the zinc content in the leaves reached µg/g: 0.01–0.02 (BD without chelate); 0.43±0.05 (BD with chelate); 1.75±0.25 (DDW with chelate). As can be seen, the coefficient of zinc accumulation in leaves during incubation of shoots in DDW with zinc glycinate differs many times. Biogeochemical limitations of Zn accumulation in a medicinal plant can be overcome by a kinetic approach — modified water isotopic composition. Such a result will allow offering special metal-modified medicinal and food plants as supplements in zinc-deficient states in the future.