

### **The aim of the project**

The aim of the project is the identification of abscisic acid non-activated SnRK2 kinases substrates in the roots of the model plant *Arabidopsis thaliana*, with particular emphasis on proteins involved in RNA metabolism. Also, the effect of phosphorylation on the function of these proteins will be investigated.

### **Research description**

Identification of potential substrates of indicated kinases will be performed using differential phosphoproteomic analysis, in which the experimental model will be *A. thaliana* roots subjected to the salt stress. Changes in phosphoproteomes of wild type (WT) and all four SnRK2 ABA non-activated kinases (*snrk2.1/4/5/10*) mutant roots will be compared after their exposure to the salt stress. Among all differentially phosphorylated proteins, a few the most interesting potential substrates involved in RNA metabolism will be subjected to further analysis. Since the phosphorylation of specific amino acids plays a unique role in a function and structure of a protein, I plan to localize accurately phosphorylation sites of SnRK2s' substrates and verify them by applying *in vitro* and *in planta* methods. Moreover, the effect of the phosphorylation on functions of identified substrates will be examined on both molecular and physiological level. Throughout the project, there will be plant lines generated with the use of a CRISPR/Cas9 genome-editing method. Those lines would enable to investigate a response to salinity in plants with mutations in substrates of group I SnRK2 kinases.

### **The reasons for undertaking research topics**

During their life cycle plants repeatedly face a vast variety of stressors both biotic and abiotic. To survive, they evolved a range of precise biochemical mechanisms that enable them to sense and respond to signals coming from the environment. Among all adverse environmental conditions, the water limitation due to drought or soil salinization is one of the biggest problems plants have to face. It has a great impact on the agricultural industry as a majority of plants, including crops, are susceptible to drought and excess of salts in the soil. Gradual progressive soil salinization compromises the productivity of crops and contributes to a decrease of area for crop farming. Phosphorylation is one of the most frequent posttranslational modification influencing on many properties of a protein. Changes in the phosphorylation of a protein may affect its stability, localization, affinity to substrates or other proteins. Involvement of SnRK kinases of subfamily two (SnRK2) mediated phosphorylation in RNA metabolism is an emerging new field of research. These protein kinases are activated in response to a variety of environmental stimuli triggering stress response by phosphorylation of their substrates. Proteins involved in RNA metabolism, like RNA binding proteins or splicing factors are identified among substrates of SnRK2s thereby indicating their role in the regulation of RNA metabolism. Unlike ABA-activated group III SnRK2s, the knowledge of substrates of kinases belonging to group I is still limited and thus worth investigation.

### **Expected results**

The research proposed in this project would allow to uncover more substrates of group I SnRK2 kinases and extend the knowledge of signalling mechanisms in plant cells in response to salt stress. As ABA-non-activated SnRK2s are rapidly activated upon salt stress administration, they may play a crucial role in shaping stress response on multiple levels of gene expression by influencing components of both posttranscriptional and co-transcriptional regulation pathways. Understanding of the mechanisms of plants response to salt stress may, in the long term, allow generating plants that will cope better with the restricted water supply what is important in the context of challenges for crop production in times of climate changes.