Objective of the project

Living organisms are affected by various environmental stresses that impact their growth, development and survival. Plants, as sessile organisms, cannot easily avoid these adverse conditions and therefore are particularly susceptible to them. To survive, plants developed sophisticated adaptive mechanisms and regulatory pathways that allow them to cope with external hazards such as drought, extreme temperatures or soil contaminations. Heat is one of the main stresses affecting plants, often leading to decreased crop yield and, consequently, to significant economic losses. For this reason, studies of molecular mechanisms of stress response are important topic of research.

Recently, we have found out that model plant *Arabidopsis thaliana* accumulates significant amounts of polyprenols is response to high temperature. Polyprenols are lipid compounds present in all living organisms. In Arabidopsis, they are synthesized by *cis*-prenyltransferase 7 (CPT7) enzyme from isopentenyl diphosphate (IPP) molecules that are produced by the MVA and MEP pathways. Our research has shown that absence of CPT7, and therefore polyprenols, negatively affects functions of chloroplasts - cellular organs responsible for assimilation of carbon dioxide. It is possible that polyprenols act as protective compounds, shielding chloroplasts against adverse effects of high temperature. Although we possess some knowledge on biological functions of *CPT7*, we still do not know how its expression, as well as expression of the MVA and MEP pathway genes, is regulated. Recently, we discovered that Heat Shock Transcription Factors of HSFA1 family, the master regulators of heat stress response, are necessary for proper functions of *CPT7* gene. Similarly, chromatin remodeling protein BRM, the key regulator of gene activity in Arabidopsis, seems to be involved in regulation of *CPT7* and selected genes of the MVA and MEP pathways. We believe, that HSFA1 factors and BRM cooperate in regulation of polyprenol biosynthesis. The goal of this project is to explore this hypothesis.

Research plan

We plan to test the possibility that HSFA1 factors and BRM act together in regulation of polyprenol biosynthesis using a wide set of molecular and genetic techniques:

1) We will measure transcript levels of the MVA and MEP pathway genes in mutant plants that do not produce HSFA1 proteins. This will tell us if HSFA1 factors are necessary for their regulation. If so, we will try to answer the question if this happens by direct binding of HSFA1 factors to their genes and to *CPT7* gene.
2) Similarly, we will check if BRM is necessary for proper regulation of *CPT7* and the MVA/MEP pathway genes by measuring their transcript levels in mutants lacking BRM protein. Again, we will test if this is a direct regulation.
3) We want to analyze cooperation of HSFA1 factors and BRM protein. First, we will try to find out if they form a single protein complex. Then, we will analyze what happens with polyprenol production in plants lacking both HSFA1 factors and BRM. In parallel, by using high-throughput techniques, we want to check how simultaneous absence of these proteins globally affect the ability of Arabidopsis to regulate expression of genes in response to heat stress. Finally, we plan to test if HSFA1 factors and BRM are reciprocally necessary for proper regulation of *CPT7* and the MVA/MEP pathway genes.

Impact of the project

Completion of this project will provide the first detailed data on how *CPT7* and genes of the MVA/MEP pathways are regulated. It will help us understand how heat-induced accumulation of polyprenols is achieved. Since the MVA and MEP pathways provide substrates for biosynthesis of not only polyprenols but also other important compounds, obtained results will fill the gaps in general understanding of plant metabolism. Investigation of BRM and HSFA1 cooperation will broaden our knowledge on how these protein factors operate. Finally, completion of this project will expand our knowledge on mechanisms of adaptation to heat stress conditions and on plant biology in general. We expect that obtained results will be presented on international conferences and published in high impact international scientific journals.