

Negative impact of non-living factors exerted on living organisms in a specific environment is called abiotic stress and as a natural part of every ecosystem, affects organisms in a variety of ways. Biological organisms constantly exposed to environmental stimuli developed different strategies to cope with abiotic stress conditions. Because of their sedentary lifestyle, plants are restricted only to tolerance, resistance, and avoidance mechanisms and thus require efficient short-term strategies based on the manipulation of the existing genetic information. Therefore plants acquire resistance to the stress environment by reprogramming metabolism and gene expression, gaining a new equilibrium between growth, development and survival

Cold stress represents one of the most harmful abiotic stresses and significantly constrains the spatial distribution and agricultural productivity of soybean, affecting its growth and development. On the other hand, drought is a significant climatic risk that calls for effective mitigation strategies to sustain the supply of soybeans worldwide. Some soybean cultivars are more susceptible to drought than others, nevertheless, under dry conditions, a reduction in soybean yield by more than 50% has been reported.

Small noncoding RNA molecules exert gene expression regulatory roles, modulating, among others, growth and development of organisms, organ development, hormone signaling, and defense against pathogens. Numerous reports published in recent years confirmed the universality of small molecules derived from tRNAs (tRFs, tRNA-derived RNA fragments) and their impact on many processes occurring in organisms, including plant stress responses. Importantly, tRFs have been identified in all domains of life. Despite the fact that the effects of cold and drought stresses on soybean have been well studied, to date no report described changes in tRF abundances (or their possible roles) under neither cold nor drought conditions in soybean. Therefore the aim of the project is to identify and characterize tRFs that respond to cold and drought stress conditions in soybean.

For stress response studies, the choice of soybean varieties is of crucial importance. An important aspect that we took into account in our considerations was the possibility of cultivating selected varieties in the temperate climate. Moreover, we carefully analysed published data, considered our own experiences with soybean, and, for cold stress experiments we chose two previously used in our studies varieties - Augusta as cold resistant, and Toyomusume, as cold sensitive; for drought stress experiments we selected Acardia as drought-resistant, and Maja as drought-sensitive.

In the first part of the project we will employ high-throughput sequencing methods in order to identify tRFs and their targets changed under cold and drought stresses. More detailed analyses will cover functional characterization of tRF targets in abiotic stress responses by tRF/mRNA coexpression experiments. We will also analyse if tRFs function via RNAi pathway. The last step in the project will be understanding the roles of tRFs in soybean cold and drought stress responses via creation of transgenic soybean lines where selected tRFs will be silenced. The analyses will be widened by the phenotypic analyses of the created mutants and coexpression analyses of selected tRFs and their targets. The utilization of modern molecular biology tools ensures timely and qualitative realization of the proposed tasks.

Exploiting tRFs as stress markers gives opportunities for improving plant traits responsible for stress resistance and provides a potential tool for designing various crop improvement strategies. A combinatorial approach of multiple methodologies can help to decode the tRF-target interactions in plants. Systematic explorations on the molecular function of tRNAs and their derivatives in plants can certainly revolutionize the arena of plant biotechnology research.