

Iron is essential for the proper metabolism and physiology of cells and organisms. Dietary deficiencies of iron lead to serious illnesses such as anemia in humans and animals, or severe chlorosis and a decrease in photosynthetic activity in plants. The World Health Organization (WHO) estimates that about 30% of the world's population suffers from anemia caused by iron deficiency. Iron deficiency is common in the countries with low economic income, where the diet of people is based mainly on plants grown in the soils deficient in bioavailable iron. Due to the poor economic conditions and lack of access to diverse food resources, it is difficult to resolve this problem by a simple supplementation of the iron-enriched food. In contrast, genetic engineering of crops to fortify their Fe content appear to be a rapid and low-cost approach that could significantly improve the nutritional status of people suffering from Fe deficiency. The biofortification of crops with Fe can be achieved by the improvement of plants ability to mobilize iron from the soils and to accumulate iron in bioavailable forms. In order to realize it, recent research focus on the identification and molecular characterization of genes involved in iron uptake and accumulation to understand how they can affect the bioavailability of Fe from plant foods. Recent research indicate that the distribution of Fe within cells may be regulated by very complex pathways linking the activity of plasma membrane, mitochondrial, plastid and vacuolar transporters as well as the iron-storing and iron-chelating proteins. However, the detailed information on the localization, mechanisms of regulation and molecular properties of the proteins involved in the intracellular Fe trafficking in plant cells is still lacking. Therefore this projects aims to perform a detailed molecular and functional characterization of the genes encoding the two mitoferrins (MIT1 and MIT2) and mitoferrin-like proteins (MFL) from a model dicot plant *Arabidopsis thaliana*, that are homologous to the yeast and animal mitochondrial iron transporting permeases. Based on the available data and initial sequence analyses, we believe that *Arabidopsis* proteins localize to mitochondria (AtMIT1 and AtMIT2) or chloroplasts (AtMFL1) and transport iron into these organelles for the crucial metabolic processes, such as photosynthesis and respiration, and for the synthesis of heme and Fe-clusters – the prosthetic groups, that are essential for the activity of many cellular proteins. Since the level of free iron in mitochondria must be tightly controlled to avoid oxidative stress, we suggest that the Fe<sup>2+</sup> ions imported into the mitochondrial matrix through MITs are immediately transferred to the iron-chelating chaperone frataxin which delivers Fe to the target proteins or Fe-S cluster biosynthesis machinery. Since frataxin has been shown to directly interact with the proteins involved in Fe-S cluster biosynthesis, we think that the transfer of Fe<sup>2+</sup> between MITs and frataxin can also occur through a direct physical interaction of Fe permeases and Fe chaperone in mitochondria. As frataxin is also present in chloroplasts, we assume that similar mechanism of iron import and transfer to target proteins can occur in these organelles (MFL-frataxin interaction). Since the mitoferrins and mitoferrin-like proteins are small proteins with a molecular mass ranging from 28 to 35 kDa, we also assume that they may act as the homo-oligomeric or hetero-oligomeric complexes. In order to verify our hypotheses, we are going to determine the subcellular localization of *Arabidopsis* proteins, their target metal substrates, the molecular mechanisms of their regulation including the identification of the amino acid residues crucial for their localization and activity, and their ability to form high molecular mass complexes. We believe that the results of our studies will greatly improve the understanding of the molecular mechanisms involved in the maintaining of cellular iron homeostasis in plant cells, and thus will be useful during the development of the future strategies for efficient biofortification of staple crops in bioavailable iron.