Mitochondria are complex and dynamic organelles responsible for many critical cellular processes. Organisms have developed specific mechanisms known as the mitochondrial quality control (mtQC) system, which ensures proper organelle function and, thus, cellular homeostasis. One of the quality control mechanisms responsible for removing damaged or aged mitochondria is the process of mitophagy, a form of selective autophagy. In this process, double-membraned autophagosomes enclose damaged mitochondrial areas or whole organelles and deliver the cargo to lysosomes (in animals) or vacuoles (in yeast and plants) for degradation. Despite the evolutionary conservation of mitophagy components between kingdoms, the molecular mechanisms of mitophagy in plants are still largely unknown.

Within organelles, mitochondrial proteases are one of the central components of mtQC. Our studies found that in the plant *Arabidopsis thaliana*, the loss of the inner mitochondrial membrane protease FTSH4 leads to morphologically altered, swollen mitochondria, the so-called giant mitochondria. Similar observations have been made in yeast *Saccharomyces cerevisiae* lacking the Yme1 protease, a homolog of the plant FTSH4. The presence of enlarged mitochondria in the *yme1* mutant and the discovery of the involvement of Yme1 in mitophagy in yeast prompted us to postulate that in the *A. thaliana* plants deficient in FTSH4, the elimination of enlarged mitochondria is defective or inhibited. The recognition of mitochondria by autophagy machinery may be achieved by the interaction of one of the outer or inner mitochondrial membrane proteins with ATG8, a central protein of the autophagy machinery. In *S. cerevisiae*, the outer membrane ATG32 protein acts as a mitochondrial receptor, which is cleaved by the Yme1 protease upon mitophagy induction. Very little is known about proteins having a similar function on the plant mitochondrial surface. Our preliminary proteomic data suggest that one of the receptor candidates is the outer membrane OMP85 protein, which is possibly cleaved by FTSH4. The *in silico* analysis also indicates that OMP85 possesses the conserved ATG8-interacting motif, suggesting its interaction with ATG8 and involvement in the formation of mitophagosome. Based on the current knowledge about mitophagy in other kingdoms and our preliminary data, we propose that in *A. thaliana*, the FTSH4 protease is involved in mitophagy by proteolytic cleavage of a specific mitochondrial protein functioning as a mitophagy receptor, and OMP85 is one of the candidates. Recently published data indicate that FTSH4 possesses an ATG8-interacting motif as well. Thus, FTSH4 may be a marker for mitochondrial degradation interacting with ATG8 upon severe mitochondrial damage.

In this proposal, we aim to uncover whether and how the FTSH4 protease participates in the process of mitophagy and, by that, to understand the functional relationship between the mitochondrial proteolytic system and selective autophagy in plants. We favor the scenario that under severe stress conditions, FTSH4 protease functions in “limited” proteolysis and cleaves off a fragment of a mitophagy receptor, mediating mitophagy in plants. However, we cannot exclude the second scenario, and both possibilities will be tested. We propose to apply complementary approaches in cellular imaging, biochemistry, and molecular biology to uncover the potential involvement of the FTSH4 protease in the process of mitophagy in plants. Using confocal and transmission electron microscopy, we plan to visualize mitophagosome formation in wild-type and *ftsh4* mutant plants. Applying the *in vivo* protein-protein interaction method, we want to determine the interaction between FTSH4 and putative mitophagy receptors and the ATG8-interacting mitochondrial receptors. Using a mass spectrometry-based proteomic approach, we plan to describe the protein composition of the isolated mitophagosomal fractions in wild-type and *ftsh4* mutants. At last, we want to estimate the employment of another mitochondrial quality control mechanism, the ubiquitin-proteasome pathway, in maintaining cellular homeostasis under potentially defective mitophagy in *ftsh4* compared to wild-type.

Considering that little is known about mitophagy in plants, and there is also no information on the involvement of the plant mitochondrial proteolytic system in the participation in mitophagy, the proposed approaches will help understand the underlying mechanisms of selective autophagy in plants. Our studies will also provide new insights into the two crucial components of the mitochondrial quality control mechanisms that ensure the cell’s normal functioning.