Abstract

Mitochondria are essential double-membrane bound organelles and the major site of cellular energy production. Mitochondria form a highly dynamic network inside eukaryotic cells which is crucial for their activity. In order to tackle the physiological demands imposed on mitochondria, they actively monitor the integrity of their protein pool. In addition, due to the dynamic nature of mitochondria the maintenance of their membrane structure is of extreme importance. Mitochondrial membranes mainly consist of phospholipids. The mitochondrial phospholipids phosphatidylethanolamine (PE) and cardiolipin (CL) are particularly important for mitochondrial functionality, cell viability, and the pathogenesis of human diseases. While mitochondria actively contribute to the biogenesis and cellular supply of these lipids, regulatory mechanisms maintaining and adjusting PE and CL level still need to be resolved.

Recently, members of the conserved Ups1/PRELI-like family of proteins have been linked to the maintenance of mitochondrial phospholipids: yeast Ups1 and Ups2 reside in the intermembrane space (IMS) of mitochondria and regulate CL and PE level, respectively. Despite accumulating data molecular functions of Ups1 and Ups2 in regards to the regulation of mitochondrial CL and PE level remain elusive. A particular interest of the present study was to get further insights into the regulatory processes maintaining mitochondrial CL and PE level in the yeast *Saccharomyces cerevisiae*. The identification of factors which control the activity of Ups1 and Ups2 in mitochondria was in the focus of the present thesis. Futhermore, it was aimed to further uncover the physiological relevance and the functional conservation of Ups1 and Ups2.

Using a biochemical protein purification approach, Mdm35 was found to assemble with Ups1 and Ups2 in the IMS. Moreover, quantitative assembly with Mdm35 was found to be crucial for Ups1 and Ups2 stability, solubility, and activity. In addition, the IMS resident *i*-AAA protease Yme1 was identified to degrade unassembled Ups1 and Ups2 giving rise to the concept of a regulatory network controlling the accumulation of CL and PE in mitochondria. Loss of Mdm35 – Ups protein complexes results in a drastically decreased cellular tolerance to low temperatures, indicating that CL and PE regulation is particularly important under those conditions. Finally, all components of the described regulatory network are conserved from yeast to man, suggesting that there is a similar system assuring mitochondrial integrity in humans.