Abstract

Mitochondrion is believed to be an ubiquitous organelle which occurred about 1,5 billion years ago by a single endosymbiotic event. Mitochondria is mostly dependent on the protein import from cytosol thus the establishment of protein import machinery was essential for seizing the new endosymbiont. Possibilities of studying the evolution of protein import machineries are quite limited given that no „free living“ mitochondria or amitochondriate organisms are known nowadays. One alternative is to study mitochondrial secondary reductive evolution of anaerobic parasitic protists.

Giardia intestinalis is flagellated protozoan living in microaerofilic environment of the small intestine. It containes one of the most reduced mitochondrion (mitosome) described so far. Hence it serves as a great model for studying mitochondrial evolution. Although it is well understood that all mitosomal proteins are transported from cytosol, many aspects of protein import pathway remain elusive.

While the main channel Tom40 is present in the outer membrane, two other main translocases (Sam50 which is required for betta-barrel assembly in the outer membrane and Tim17/22/23 which is essential for protein translocation through the inner membrane) have not been identified so far. Protein translocation through Tim17/22/23 channel is in classical mitochondria driven by PAM complex together with membrane potential. Although some components of PAM complex are present in the inner mitosomal membrane, no membrane potential was detected. Hence, *G. intestinalis* might employ a unique translocase which cooperates with the residual PAM complex without contribution of membrane potential and which is adapted to import an extremely small set of proteins.

This work is focused on characterisation of this unknown mitosomal inner membrane translocase from import apparatus in *G. intestinalis*. Main objectives of this project were to (i) introduce a “molecular plug” into the import channel to isolate and characterize the translocase and (ii) to test our hypothesis that the endoplasmic reticulum Sec61α translocase is also present in mitosomes and plays potential role in protein import pathway.

Key words: mitochondria, molecular evolution, protein transport, organelle biogenesis