ABSTRACT

Mitochondria's primary function is to produce energy through the process of oxidative phosphorylation. ATP synthase is a macromolecular rotary machine located in the inner mitochondrial membrane that catalyzes the synthesis of adenosine triphosphate (ATP) from adenosine diphosphate (ADP) and inorganic phosphate (Pi).

The mitochondrial disorders due to ATP synthase deficiency represent a heterogeneous group of diseases characterized by variable severity of the phenotype with onset at birth or later in life till adulthood. Mutations in both, mitochondrial or nucelar DNA encoded genes, may result in ATP synthase impairment, either isolated or combined with deficits of other complexes of oxidative phosphorylation.

The aims of the thesis were to characterize TMEM70 protein, an ATP synthase assembly factor, and to analyze the impact of novel disease variants leading to ATP synthase deficiency in patients´ derived samples.

TMEM70 is a 21 kDa hairpin structure protein localized in the inner mitochondrial membrane, with both termini oriented into the matrix, which forms higher oligomer structures. Our results demonstrated that the absence of TMEM70 protein leads to an isolated deficiency of complex V followed in some stage by adaptive/compensatory effect of respiratory chain complexes. Different severities of ATP synthase deficiency were observed in muscle mitochondria due to extremely rare heteroplasmic variants of *MT-ATP6*. While m.8851T>C (p.Trp109Arg) variant lead only to mild reduction of ATP synthase, m.8719G>A (p.Gly65*) variant resulted in diminished levels of ATP synthase holoenzyme and the presence of assembly intermediates. Similarly, a profound impact on ATP synthase was observed in the case of novel variants in *MTTK* and *PUS1* genes, which was accompanied by deficiency of other oxidative phosphorylation system complexes.

Keywords: mitochondria, oxidative phosphorylation system, ATP synthase, mitochondrial diseases, mitochondrial DNA, *MT-ATP6*, *MTTK*, *TMEM70*