

## ABSTRACT

Disorders of ATP synthase, the key enzyme of mitochondrial energy provision belong to the most severe metabolic diseases presenting mostly as early-onset mitochondrial encephalo-cardio-myopathies. Mutations in four nuclear genes can result in isolated deficiency of ATP synthase, all sharing a similar biochemical phenotype – pronounced decrease in the content of fully assembled and functional ATP synthase complex. The thesis summarises studies on two distinct causes of ATP synthase deficiency. First is TMEM70 protein, a novel ancillary factor of ATP synthase, which represents most frequent determinant of severe inborn deficiency of ATP synthase. TMEM70 is a 21 kDa protein of the inner mitochondrial membrane, facilitating the biogenesis of mitochondrial ATP synthase, possibly through TMEM70 protein region exposed to the mitochondrial matrix, but the proper regulatory mechanism remains to be elucidated. In TMEM70-lacking patient fibroblasts the low content of ATP synthase induces compensatory adaptive upregulation of mitochondrial respiratory chain complexes III and IV, interestingly by a posttranscriptional mechanisms.

The second type of ATP synthase deficiency studied was mtDNA *m.9205delTA* mutation affecting maturation of MT-ATP8/MT-ATP6/MT-CO3 mRNA and thus biosynthesis of Atp6 (subunit *a*) and Cox3 structural subunits. With the help of transmitochondrial cybrids with varying mutation load it was possible to elucidate gene–protein relationship of the pathogenic mechanism with mutation threshold close to homoplasmy. Characterisation of resulting enzyme deficiencies revealed pronounced decrease of cytochrome *c* oxidase biosynthesis contrasting with increased amount of structurally and functionally altered ATP synthase, unable to produce ATP.

**Key words:** Mitochondrial diseases, OXPHOS system, ATP synthase deficiency, ATP synthase biogenesis, TMEM70, mtDNA mutation, *MT-ATP6*, heteroplasmy, threshold effect.