ABSTRACT

Mitochondria are semi-autonomous organelles that contain their own DNA. Human mitochondrial DNA (mtDNA) encodes a total of 37 genes: 13 subunits of oxidative phosphorylation complexes (OXPHOS), 22 transfer RNA (tRNA) molecules and 2 ribosomal RNA (rRNA) molecules. Pathogenic mutations in genes associated with mitochondrial translation are a common cause of mitochondrial disease. These mutations can be found in mtDNA or in nuclear genes encoding ribosomal proteins, initiation, elongation and termination factors, mitochondrial tRNA-modifying enzymes and aminoacyl-tRNA synthetases. Mitochondrial aminoacyl-tRNA synthetases (mt-aaRS) are enzymes that catalyse the addition of single amino acids to specific tRNAs.

The aim of the bachelor thesis was an introduction to the work in the tissue culture laboratory. To prepare samples for the following experiments, skin fibroblasts from five patients with mt-aaRS disorders (*AARS2*, *DARS2*, *NARS2*, *SARS2*) and control lines were cultured in glucose and galactose media. Subsequently, the procedure for determining the equilibrium amount of selected subunits of the OXPHOS complexes was optimized and applied to the analysis of fibroblasts from five patients with mt-aaRS disorder. When the cells were cultured in glucose medium, decreased levels of some subunits of complex I, complex III and complex IV of the OXPHOS system were observed in four patients. When galactose was used as a carbohydrate source in the medium, it was evident that there was no reduction of OXPHOS system complex subunits in the fibroblasts of the patients. Furthermore, the necessity of using an age-matched control group of patients was demonstrated. The thesis confirmed that fibroblasts from adult control is not suitable for comparison with fibroblasts from paediatric patients aged half to three years.