MitoFit Open Seminars

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Novel applications of high-resolution respirometry as useful tool to characterize rare metabolic disorders.

Congenital disorders of glycosylation (CDG) are rare inherited diseases caused by abnormal protein and lipid glycosylation. Recent published data and our preliminary results indicate a possible interconnection between glycosylation defects and mitochondrial function abnormalities.The study of mitochondrial metabolism in congenital disorders of glycosylation may contribute to the elucidation of pathomechanisms in unclear metabolic diseases.

The aim of study will be to analyze mitochondrial respiration and multisensor applications of fibroblasts from patients with CDG and their impact on cellular and energetic metabolism by high-resolution respirometry (Oroboros Instruments). Moreover, other bioenergetic applications (coenzyme Q redox state) will be investigated using the new applications of the NextGen-O2k. Mitochondrial respiration will be measured in fresh permeabilized fibroblast cell lines from patients with CDG. Our preliminary results indicate secondary functional abnormalities in mitochondria and glycolytic dysfunction due to a breakdown of the glycosylation pathway. We would like also to focus on measurements of membrane potential and ROS production in some of these CDG patients, because changes in ROS production and membrane potential were found previously in CDG by other methods in our laboratory. In parallel, we will analyze fibroblasts lines from patients with proved or suspect for other rare metabolic diseases. Reduced level of ubiquinone was revealed in some patient's fibroblasts by using HPLC method. Therefore, we would like to focus on measuring Q-redox changes in permeabilized fibroblasts to see how reduced Q level can involve in Q-redox changes.

This project should help us to find cellular pathways interconnections with help of complex approach by investigation of mitochondrial respiration, ROS production, membrane potential and Q-redox changes in O2k-FluoRespirometer and the NextGen-O2k. The same Substrate-Uncoupler-Inhibitor Titration (SUIT) protocol will be used for all methods, which should give us more valid correlation between all the measured parameters.

The results obtained from this STSM could reveal more information about pathomechanisms of CDG and other selected rare diseases, give us new knowledge about mitochondrial physiology and energetic metabolism in fibroblast cell lines and could show us cooperation between different metabolic pathways.