### 8P.4 The dopamine-$D_2$-receptor agonist ropinirole dose-dependently blocks the $Ca^{2+}$-triggered permeability transition of mitochondria

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The dopamine-$D_2$-receptor agonist ropinirole exerts neuroprotective activity. Assuming that this neuroprotection might be associated with inhibition of the apoptotic cascade underlying cell death, we examined a possible effect ropinirole on the permeability transition pore (mtPTP) in the mitochondrial inner membrane. Using isolated rat liver mitochondria, the effect of ropinirole was studied on $Ca^{2+}$-triggered large-amplitude swelling, membrane depolarization and cytochrome *c* release. In addition, the effect of ropinirole on oxidation of added, membrane-impermeable NADH was investigated. The results revealed doubtlessly, that ropinirole can inhibit permeability transition. In patch-clamp experiments on mitoplasts, we show directly that ropinirole interacts with the mtPTP. Thus, ropinirole reversibly inhibits the opening of mtPTP with an $IC_{50}$ of 3.4 μM and a Hill coefficient of 1.3. In both systems (i.e. energized mitochondria and mitoplasts) the inhibitory effect on permeability transition was attenuated by increasing concentrations of inorganic phosphate. In addition, we showed with antimycin A-treated mitochondria that ropinirole failed to suppress respiratory chain-linked ROS release. In conclusion, our data suggest that the neuroprotective activity of ropinirole is due to the inhibition of the release of apoptogenic factors from mitochondria.

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### 8P.5 Mitochondrial dysfunction in fibroblasts of patients with amyotrophic lateral sclerosis

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Amyotrophic lateral sclerosis (ALS) has been commonly regarded as a neurodegenerative disorder primarily involving the pyramidal motor system. There is however some evidence that disease-related degenerative changes also occur in extraneural tissues. Therefore, primary skin fibroblast cultures from ALS patients were analysed for mitochondrial anomalies. To verify the putative impairment of mitochondrial function in fibroblasts of patients with ALS, the oxygen consumption (respiration rate) of fibroblasts was measured using a high resolution oxygraph. To check possible metabolic consequences of altered mitochondrial function enzyme activity of aconitase (which harbors a highly ROS-sensitive iron–sulfur-cluster) was determined. Furthermore, mitochondrial DNA copy number analysis and deletion screening were performed. Age matched healthy subjects served as controls. We observed slightly lower maximal rates with NAD-dependent substrates glutamate + malate in the fibroblasts of ALS patients compared to controls. The succinate supported maximal respiration rates in the fibroblasts from these patients were also lower. Statistically significant deficiencies of respiratory chain complexes I (NADH:CoQ1 oxidoreductase) and IV (cytochrome *c* oxidase, COX) were identified by determination of elevated flux control coefficients of respiration in titrations with specific inhibitors (amytal for complex I and azide for complex IV). Furthermore, the aconitase activity levels were altered and diminished levels of mitochondrial DNA copies were also observed in ALS fibroblasts. Our results clearly show mitochondrial impairment in extracerebral tissue (fibroblasts) of patients with ALS. We propose that the mitochondrial changes described might potentially serve as biomarkers that allow objective ALS patient diagnosis and therapeutic monitoring in an accessible, peripheral tissue.

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