S12.45 Evidence that the mitochondrial phosphate carrier is a key component of the mitochondrial permeability transition pore

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The adenine nucleotide translocase (ANT) has been proposed to form the inner membrane channel of the mitochondrial permeability transition pore (MPTP). Opening of MPTP is enhanced by phenylarsine oxide (PAO) which is known to bind to ANT. However, here we show that this binding is prevented by pre-treatment of mitochondria with carboxyatractyloside (CAT) or bongkrekic acid (BKA) that activate and inhibit pore opening by inducing the "c" and "m" conformation of the ANT respectively. Yet, MPTP opening in mitochondria pre-treated with CAT or BKA is still activated by PAO treatment, implying that the ANT is not the major target of PAO action. We have identified 4 membrane proteins that bind to a PAO column following CAT-treatment, one of which is the mitochondrial phosphate carrier (PiC). Using both GST-CyP-D pull-down and co-immunoprecipitation we demonstrate that the PiC binds to cyclophilin-D (CyP-D), a key component of MPTP, in a cyclosporin A (CsA) sensitive manner. Our data suggest that the PiC may be the channel-forming component of the MPTP and we are investigating this further by using siRNA knockdown in Hep2G cells with subsequent determination of pore opening.

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S12.46 Hypoxic pulmonary vasoconstriction is modulated in UCP2 knock-out mice during acute and sustained hypoxia

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The aim of this study was to reveal the role of the mitochondrial membrane protein UCP2 in hypoxic pulmonary vasoconstriction (HPV). HPV is a physiological response of the lung vessels to alveolar hypoxia in order to match lung perfusion to alveolar ventilation. UCP2 is widely expressed in lung tissue with hitherto unknown function and might play a role in sensing hypoxia and modulating HPV. Therefore we compared pulmonary arterial pressure in isolated blood free perfused lungs of wild-type and UCP2 knock-out ($UCP2^{-/-}$) mice during hypoxic ventilation. In $UCP2^{-/-}$ mice acute hypoxic vasoconstriction (ventilation of 1% oxygen for 10 min) was significantly higher than in wild-type mice (increase in pulmonary arterial pressure 1.5+/−0.1 mmHg (n=5) vs. 1.0+/−0.1 mmHg (n=7), data as average+/−SEM, $p<0.01$). In contrast sustained hypoxic vasoconstriction (1% oxygen for 3 h) was significantly lower after 70–140 min of hypoxia (minute 140: wild-type 1.8+/−0.2 mmHg (n=9), $UCP2^{-/-}$ 1.2 mmHg +/− 0.2 mmHg (n=9) $p<0.05$). Normoxic pulmonary arterial pressure and pressure response to a thromboxane analogon was not different. Polymerase chain reaction showed presence of UCP2 mRNA in pulmonary arterial smooth muscle cells. We conclude that UCP2 plays an important role in regulation of pulmonary vasculare tone during hypoxia.

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S12.47 Glycine prevents mitochondrial impairment caused by left carotid occlusion

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Mitochondria play a sufficient role in neuronal function. Several cerebral disorders like a stroke results in neuronal degeneration and associated with substantial reduction in mitochondrial functional activity and apoptosis induction. Monitoring of mitochondrial capacity for oxidative phosphorylation could be used as an indicator of stroke development. There is evidence from clinical studies that glycine reduced brain damages caused by stroke. However, the mechanism of the protection afforded by glycine is not yet known. In the present study we attempted to elucidate the mechanisms of glycine anti-stroke activity. The left carotid artery occlusion was used as a model of brain ischemia. After occlusion respiratory control index of brain cortex mitochondria was measured. It was reduced from 6.7±0.1 to 4.2±0.1 after 24 h occlusion. The development of apoptosis process was also detected, DNA internucleosomal fragmentation and caspase-3 activation was observed. When animals were treated with glycine *per os* before occlusion the reduction of respiratory ratio and caspase-3 activation were prevented. Glycine allows mitochondria to maintain their respiratory activity in ischemic conditions. Our novel data indicate that anti-stroke glycine activity is associated with its ability to prevent mitochondrial disorder and apoptosis development in brain cortex tissue induced by ischemia.

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S12.48 The cannabinoid cb1 antagonist, rimonabant, improves hepatic mitochondrial function in rats fed a high fat diet

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The endocannabinoid system has recently emerged as an important regulator of energy homeostasis, involved not only in the control of food intake but also in the regulation of energy metabolism at peripheral level. The aim of this study was to investigate the effect of rimonabant on hepatic mitochondrial function in rats fed a high fat diet. Sprague Dawley rats fed a high fat diet during 13 weeks were treated with rimonabant (10 mg/kg/day) during the 3 last weeks and matched with pair-fed controls. Our data show an increase of oxygen consumption in isolated mitochondria from rimonabant treated rats. Especially an increase of mitochondrial oxygen consumption with palmitoyl CoA compared to those with palmitoyl-L-carnitine stating that the entry of fatty acids in mitochondria via the carnitine palmitoyltransferase system was increased in rimonabant treated rats. Moreover rimonabant has induced a reduction in the enzymatic activity of the ATP synthase whereas the quantity of mitochondrial DNA and the citrate synthase activity were remained unchanged. That seems to indicate that oxidation of respiratory substrates was increased in order to compensate the reduction of ATP synthesis but without variation of mitochondrial density. The results obtained suggest that rimonabant has a beneficial effect on mitochondrial function by facilitating fatty acids oxidation, thereby reducing fatty liver and so takes part in the increase of insulin sensitivity observed under rimonabant treatment.

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