3,5-diiodo-L-thyronine (T2) administration to high fat fed rats, reduces adiposity and body weight gain by stimulating hepatic fatty acid oxidation with a concomitant less efficient utilization of lipid substrates. When injected into hypothyroid rats, T2 is able to increase resting metabolic rate, lipid metabolism and survival to cold. Skeletal muscle, endowed with significant mitochondrial capacity, is an important site for glucose disposal, lipid oxidation and thermogenesis, its mitochondrial proton-leak accounting for a significant fraction of resting metabolism, thus the effects of T2 on mitochondrial skeletal muscle would be of great physiological relevance. To test if T2 could affect mitochondrial skeletal muscle metabolism we injected a single dose of T2 (25 μg/100 g bw) in hypothyroid rats. Within 1 h following T2 injection we detected i) a significant increase in mitochondrial fatty acid oxidation rate (+80% vs hypothyroid rats) and total carnitine palmitoyl-transferase (CPT) activity (+35% vs hypothyroid rats), ii) the activation of AMPK-AcetylCoA carboxylase (ACC)-malonyl CoA signalling pathway, thus suggesting its involvement in the CPT activation by T2 iii) a significant increase (+55% vs hypothyroid rats) of the mitochondrial thioesterase-I activity and a less efficient utilization substrates through an induction of fatty acid-dependent mitochondrial uncoupling. These data give further insight on the effects of T2 highlighting mitochondrial skeletal muscle as one of its targets.

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S8.22 Regulation of mitochondrial complex I by nitric oxide during hypoxia

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Nitric oxide (NO) is known to cause persistent inhibition of mitochondrial respiration as a result of *S*-nitrosation of NADH: ubiquinone oxidoreductase (complex I). Mitochondrial complex I can exist in two interconvertible forms, active (A) and de-active, dormant (D). When the enzyme is idle, the catalytically active A-form is spontaneously converted into the D-form. We have now found in mitochondrial membranes that the conformational state (A or D) of complex I is an important factor for the interaction of the enzyme with nitrosating agents, since only the D-form was susceptible to inhibition by nitrosothiols and peroxynitrite. We also have observed the conversion of the enzyme into the D-form during hypoxic incubation of J774 macrophages, indicating that slowing down of the activity of the mitochondrial respiratory chain in hypoxic conditions leads to spontaneous de-activation of complex I. Treatment of the cells with NO following hypoxic incubation resulted in persistent inhibition of complex I. Thus de-activation of complex I and consequent modification by nitrosating agents prevents transition to the A-form so that the enzyme becomes locked in its D-form. Such locking of complex I in the D-form may have pathophysiological consequences, and may vary depending on the duration of hypoxia, type of tissue and/or the presence of natural effectors of A/D transition such as calcium and free fatty acids.

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S8.23 Oxygen dependency of mitochondrial respiratory chain function in hypoxic pulmonary vasoconstriction

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The aim of this study was to correlate the oxygen affinity and redox state of mitochondria of pulmonary arterial smooth muscle cells (PASMC) with hypoxic pulmonary vasoconstriction (HPV) to investigate the role of mitochondria in sensing of acute hypoxia. High resolution respirometry was used for determination of oxygen pressure at half maximal respiration (p50) in PASMCs and remission spectrophotometry for determination of redox state of mitochondrial cytochromes in the isolated lung. Respirometric analysis showed a p50 of 0.11±0.01 kPa (n=7) in PASMCs, which was not significantly different from aortic smooth muscle cells (0.10±0.01% kPa, n=8). Hypoxia induced calcium release in PASMCs was detected at 3% oxygen, which correlates with a 3.6±0.2% inhibition of maximal mitochondrial respiration calculated by the p50 according Michaels-Menten-Kinetics. By remission spectrophotometry we detected a reduction of mitochondrial cytochromes in the isolated blood free perfused and ventilated lung at an oxygen concentration of ≤1% oxygen, whereas HPV starts at an oxygen concentration of 10% oxygen. We therefore conclude that respiration in PASMCs is only slightly inhibited at oxygen concentrations at which acute hypoxic cell response is initiated and that this is not different from non oxygen sensing cells. All data were shown as average±standard error of the mean.

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S8.24 Cell-to-cell cross-talk between mesenchymal stem cells and cardiomyocytes: The role of mitochondria

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Many studies display efficiency of stem and progenitor cells in injured myocardium function refinement. Integration of transplanted cells into myocardial tissue is of a particular interest. We examined the possibility of cell-to-cell exchange of cytoplasm and organelles between mesenchymal stem cells (MSC) and rat cardiomyocytes. We revealed that after cocultivation cells formed intercellular contacts (through nanotubes or gap junctions) and transient exchange with cytosolic elements could be observed. Confocal and electron microscopy revealed mitochondrial-like structures in the intercellular cytoplasmic nanotubes, indicating a possibility of mitochondrial transport through the nanotubes. We revealed the result of such transport — mitochondria were transferred from MSC to cardiomyocytes. The cytoplasm transport between contacted cells was confirmed by exchange of the fluorescent probe Calcein between cells. Another result of coculturing was the appearance of human cardiac-specific myosin H-chain in MSC. We proposed that penetration of cardiomyocyte's cytoplasm to MSC caused such differentiation by donation of some signaling molecules to ignite MSC differentiation on the cardiac muscular pathway. We can speculate that cardiomyocytes-to-MSC cross-talk caused turning on differentiation of MSC towards contractile cells, and second MSC can donate mitochondria to