subsarcolemmal mitochondria and rarefaction of intermyofibrillar mitochondria. Although the maximal activities of respiration of saponin-permeabilized muscle fibers and digitonin-permeabilized fibroblasts were only slightly affected by the MFN2 mutations, their sensitivity to the cytochrome c oxidase (COX) inhibitor azide was increased, which indicates a decrease of in vivo activity of COX.

In comparison to controls, the MFN2 fibroblast samples showed a decrease in the mitochondrial DNA copy number, which explains the observed mitochondrial respiratory chain dysfunction. Additionally, an increased amount of deletions was observed. However, the deletions are unlikely to contribute significantly to the detected respiratory impairment, because of their minor overall amounts in these patients.

Our findings support the viewpoint that impairment of mitochondrial fusion causes mild respiratory chain dysfunction through defective mitochondrial DNA replication.

References

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A lung cancer model linking apoptotic resistance and metastatic potential via defects in mitochondrial fission protein

Dynamin-related protein 1

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Resistance to apoptosis is a hallmark of cancer. Evasion of apoptosis is implicated in almost all aspects of cancer progression, as well as treatment resistance. Apoptosis is regulated in part by mitochondria, which control tissue homeostasis by eliminating damaged cells. In this study, resistance to apoptosis was identified in lung epithelial (A549) cells as a consequence of defects in mitochondrial and autophagic function.

Mitochondrial function is determined in part by mitochondrial morphology, a process regulated by mitochondrial dynamics whereby the joining of two mitochondria, fusion, inhibits apoptosis while fission, the division of a mitochondrion, initiates apoptosis. Mitochondrial length correlated with metastatic potential; lung epithelial cells with increased metastatic potential had mitochondria with an elongated phenotype—mimicking cells deficient in mitochondrial fission protein, Dynamin-related protein 1 (Drp1). A549 cells had impaired Drp1 mitochondrial recruitment and decreased Drp1-dependent fission. Cytochrome c release, caspase-3 and PARP cleavage were impaired both basally and with apoptotic stimuli in A549 cells.

Metastatic potential positively correlated with mitochondrial mass, suggesting defects in mitophagy (mitochondrial selective autophagy). A549 cells had decreased LC3-II lipilation and lysosomal inhibition suggesting that defects in autophagy occur upstream of lysosomal degradation. Immunostaining also indicated that mitochondrial localized LC3 punctae in A549 cells increased after mitochondrial uncoupling or with a combination of mitochondrial depolarization and ectopic Drp1 expression. Increased inhibition of apoptosis in A549 cells is correlated with impeded mitochondrial fission and mitophagy. We suggest that mitochondrial fission defects contribute to apoptotic resistance in lung cancer cells with a high propensity for metastasis.

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Mitochondrial fusion/fission proteins in NARP and Rho0 human osteosarcoma cells

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Dysfunctions of mitochondria are usually associated with numerous diseases like metabolic disorders, cancer and neurodegenerative diseases. Changes caused by the chronic mitochondrial stress include defects in respiratory chain complexes, morphology and organization of the mitochondria, mitochondrial membrane potential (Δψm), cytosolic Ca2⁺ concentration, ATP and ROS levels. These parameters are involved in the retrograde signaling from mitochondria to nucleus that triggers mitochondrial stress response (MSR) of the cell and its subsequent adaptation to altered mitochondrial functions [1,2]. Although knowledge about components involved in the mitochondrial retrograde signal transduction is still incomplete, it is likely that mitochondrial morphology and positioning within the cell can play an important role in mitochondrial–nuclear communication. Proteins implicated in dynamics of mitochondria were investigated in cells with chronic mitochondrial stress:

1) Rho0 human osteosarcoma cells, lacking mitochondrial DNA.
2) Cybrid NARP human osteosarcoma cells with point mutation T8993G in subunit 6 of ATP synthase (98% of heteroplasmy).

We have previously shown that many aspects of physiology (calcium homeostasis, ROS metabolism) as well as mitochondrial network and cytoskeleton organization in cells with chronic mitochondrial stress (NARP and Rho0) differs from that in WT cells [3]. Our new results indicate that the profile of proteins responsible for the dynamics of mitochondria (Drp1, Opal, Mfn1 and Fis1) is different in investigated cell lines. The observations carried out in the confocal microscope show changes in the organization of mitochondria within these cells.

References

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The MINOS complex: Keeper of mitochondrial membrane architecture

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Mitochondria are surrounded by two distinct membranes. While the outer membrane confines the organelle, the inner membrane has a key role in cellular energy metabolism. The inner mitochondrial membrane is subdivided into the inner boundary membrane, which is closely opposed to the outer membrane, and large irregular invaginations termed cristae. Narrow tubular openings – the crista junctions – connect crista membrane and inner boundary membrane domains. Additionally, sites of contact between inner boundary and outer mitochondrial membranes are frequently observed. We have recently discovered a mitochondrial inner membrane organizing system (MINOS complex) that is crucial for both, the formation of cristae junctions and membrane contact sites. MINOS is composed of six inner membrane proteins: the two highly conserved core subunits FCJ1/mitofilin and MIO10/MITOS1 together with Aim5, Aim13, Aim37 and Mio27. Deletion of FCJ1 or MIO10 in yeast abolishes MINOS formation and leads to a grossly altered mitochondrial ultrastructure with extended stacks of sheet-like cristae membranes and the loss of cristae junctions. Moreover, MINOS interacts with several protein complexes of the outer mitochondrial membrane, like the SAM/TOM complex or the TOM complex at membrane contact sites. Formation of MINOS contact sites supports the import of precursor proteins from the cytosol into the intermembrane space and outer membrane. A detailed structure/function analysis of FCJ1/mitofilin revealed that membrane tethering, MINOS integrity and formation of cristae junctions depend on different FCJ1/mitofilin protein domains.

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Diverse reactions of pea seedlings mitochondria depending on low temperature load
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Mitochondria play a central role in allowing plants to adapt to extreme temperatures. To operate in stressful conditions, plant mitochondria might have evolved distinctive features designed to increase their metabolic flexibility and stress tolerance. In our study we used various low temperature loads to reveal unique features or/and commonalities in cold responses of plant mitochondria. Here we are investigating the impact of hardening (+7 °C), mild stress (+2 °C) and severe stress (−7 °C) on respiration rates, malondialdehyde (MDA) accumulation and linking this to changes in alternative enzymes abundance in pea seedlings mitochondria.

TBARS assay revealed that mild and severe treatments increased MDA levels nearly 30 %, but no increase in whole seedling MDA level was recorded during hardening. Oxigraphic measurements showed that the control pea mitochondria well oxidized Malate/Glutamate with high respiratory control ratio (RCR) about 3.2. Oxidative rates for Succinate and NADH increased progressively whereas RCRs were 2.2 and 2.6 respectively. Hardening led to reduced respiratory rates and RCR with Malate/Glutamate as well as to the significant rise of AOX activity and content. Input of rotenone-resistant respiration under 1 complex work was slightly increased. Respiratory rates and RCRs with Succinate and NADH also were reduced, but no substantial changes in AOX activity occurred. Under the mild stress conditions Malate/Glutamate oxidative rate increased together with the rise of internal rotenone-insensitive NAD(P)/H dehydrogenases activity. However, no changes in AOX activity and alternative enzymes content took place. At the same time a significant rise of AOX activity during Succinate oxidation was observed. Short severe stress didn’t lead to the significant change in Malate/Glutamate oxidative rate and RCR, whereas external NAD(P)/H DHs activity grew noticeably but AOX activity and content decreased. It seems that under different cold conditions and depending on oxidative stress level, behavior strategies of pea seedlings mitochondria have different missions. During the hardening the mitochondria operate to increase a cold tolerance of whole cell and finally all plant. Under the oxidative stress conditions pea seedlings mitochondria function to survive and try to maintain ATP levels, thereby allowing primary and critical metabolism to proceed.

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Synaptic and nonsynaptic mitochondria in hypothyroid conditions
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The thyroid hormones (TH) are important regulators of growth, development, and metabolism, therefore, hypothyroidism is a disorder associated to a derangement of basic physiological functions. Thyroid hormones exert nongenomic effects on the mitochondrial energy metabolism and several clinical complications of hypothyroidism, like fatigue, cold intolerance, weight gain, bradychardia, etc. are associated with the decrease in basic metabolic rate and oxygen consumption [1]. It was be revealed that synaptic (SM) and non-synaptic mitochondria (CM) could differently respond to some pathological factors [2]. Therefore, our purpose was study the changes of some parameters of mitochondrial bioenergetics in SM and CM fractions of hippocampus of adult rats in following groups: euthyroid (control), hypothyroid (methimazol-treated), and T4-treated hypothyroid states.

The nNOS translocation to CM was observed with concomitant increase of mNOS activity in hypothyroid rats. In parallel, oxidation of cytochrome c oxidase and production of peroxides with substrates of complex I (glutamate + malate) were enhanced, whereas the activity of aconitate and mitochondrial membrane potential (ΔΨm) were decreased. Furthermore, the elevation of hexokinase activity in

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