examined a total of 92 patients, of which, 30 (chronic periodontitis [CP] group) were selected for the above analysis. The institutional ethics committee approved this study and informed written consent was obtained from each participant. All clinical parameter scores of the tissue sampling areas in the CP group were significantly higher (<0.001) than in the control group. Complete mtDNA sequencing revealed a total of 264 variations in the patients, including 16 novel mutations, of which 3 were missense mutations (A4234T, A7796G, and G8115R). We also observed significant change in the membrane potential and protein levels of NADH dehydrogenase, Cytochrome *c* oxidase and HSP 60 in the patients.

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S12.11 Differential inhibition of energy-producing pathways of hepg2 cells by 3-bromopyruvate

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It was studied the effects of the alkylating agent 3-bromopyruvate (3-BrPA) on ATP producing pathways of HepG2 cells. 3-BrPA decreases HepG2 viability (10% with 10–100 μM; 25% with 300–1000 μM at 60 min; 60% with 10–100 μM; 75% with 300–1000 μM at 180 min). 150 μM 3-BrPA did not affect hexokinase-II activity, but incubation with 150 μM for 30 min decreased in 60% lactate production. Cells were incubated with 3-BrPA as above in glucose medium (GM-cells) or glucose-free medium (GFM-cells) and the basal respiration was 10,5 ± 0,62 (GM-cells) or 15.3 ± 0.96 (GFM-cells) nmol $O_2/5 \times 10^6$ cell × min^{-1}. A decrease of 22% or 50% of basal respiration by 3-BrPA was detected in GM or GFM cells, respectively. Proton leak was increased only in GM-cells (3.3 ± 0.45 to 5.1 ± 0.5). Maximum respiration was decreased only in GFM-cells (17.3 ± 1.1 to 9.5 ± 1.3). 3-BrPA decreased respiratory control ratio either in GM or GFM cells. In digitonin-permeabilized cells, complex I supported respiration was decreased by 50% in GFM-cells and complex-II supported respiration was inhibited by 50% in both media. Our results suggest that glycolysis and specific sites of mitochondria play a role in 3-BrPA-induced HepG2 death. The toxic effects of 3BrPA depend on the oxidizable substrates supplied to cells.

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S12.12 Restriction of glucose metabolism induces a metabolic switch to oxidative metabolism and drastic alteration of gene expression in glioblastoma cell line LN18

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The aim of this study was to determine the cellular effect of restricting glucose metabolism using 2-deoxyglucose in LN18 cells on oxygen consumption rate (OCR) and glycolysis rate (ECAR) as well as alterations in gene expression. The cellular OCR and ECAR were determined using XF24 Analyzer which measures the two parameters simultaneously in microplates. Gene expression was analyzed using Affimatrix human genome arrays. The normalized OCR of 2-deoxyglucose treated LN18 cells was increased while ECAR was decreased compared to control after 48 h treatment. This metabolic shift towards OXPHOS was confirmed by an observed increase in the sensitivity of cellular ATP levels to oligomycin as well as increased mitochondrial respiration capacity. Apoptosis induced by staurosporine was increased in 2-deoxyglucose treated cells. Gene expression analysis revealed a striking alteration in global gene expression as the result of restricted glucose supply. OXPHOS, pentose phosphate pathway, IGF-1 signaling, PI3K/AKT signaling, cell cycle check point, apoptosis signaling and oxidative stress pathways were among 2710 genes showed significantly altered expression. Our data demonstrated that restricting glucose drove LN-18 cells to a more oxidative state accompanied by growth suppression and drastically altered gene expression in pathways that converge at cellular energy metabolism and cell proliferation as well as death.

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S12.13 Hepatitis C virus proteins cause calcium-mediated mitochondrial dysfunction and hif-linked bioenergetic compensatory adaptation

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Hepatitis C virus (HCV) infection induces a state of oxidative stress that is more pronounced than that in many other inflammatory diseases. In this study we used well-characterized cell lines inducibly expressing the entire HCV open-reading frame to investigate the impact of viral protein on cell bioenergetics. It was shown that HCV protein expression has a profound effect on mitochondrial oxidative metabolism, with specific inhibition of complex I and $F_1F_oF_1F_0$-ATPase activities, depression of mtΔΔ and oxidative phosphorylation coupling efficiency, increased production of reactive oxygen and nitrogen species. Importantly, all these effects were causally related to mitochondrial calcium overload, as inhibition of mitochondrial calcium uptake completely reversed the observed bioenergetic alterations. Noteworthy, in spite of the oxidative phosphorylation impairment, survival of HCV proteins-expressing cells was assured by an adaptive up-regulation of glycolytic enzymes. This was linked to normoxic stabilization of the hypoxia-inducible factor (HIF 1α). Overall, the results presented show that expression of HCV proteins causes deregulation of ER-mitochondrial calcium homeostasis occurring upstream of further mitochondrial dysfunction. The expected bioenergetic unbalance is however compensated by a HIF-dependent transcriptional mechanism. These observations provide new insights into the pathogenesis of hepatitis C and may offer new opportunities for therapeutic intervention.

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S12.14 Novel Mt-DNA missense mutation in ND1 (A3418G →N38D) associated with mitochondrial dysfunction in megakaryoblastic leukaemic cells

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