tumours with ER stress inducing chemotherapy may be enhanced by modulation of glutamine metabolism. Indeed, the combination of ER stress inducing agents with glutaminase (GLS) inhibitors demonstrated strong antitumor potential *in vitro* and *in vivo* [2,3]. CB 839 – glutaminase 1 (GLS1) inhibitor –, is currently being tested in phase I clinical trials in myeloma patients and other haematological and solid tumours. Within our project, we investigate the role of glutamine metabolism in response to ER stress in PCM cell lines. Moreover, we aim to evaluate the potential of glutamine metabolism inhibition to sensitize chemotherapy-resistant PCM cells to ER stress-inducing agents. Our results show that PCM cell lines, both sensitive and resistant to anti-myeloma agents (such as dexamethasone, bortezomib or lenalidomide), strongly rely on glutamine metabolism for their growth and survival. These cell lines are characterized by upregulated basal levels of ER stress markers such as BIP, p-eIF2a or XBP1s. Moreover, in these cells, the ER stress-related mechanism of ASCT2 degradation is also observed. Currently, we are investigating the influence of glutamine deprivation combined with ER stress induction on proliferation and survival of a panel of PCM cell lines, both sensitive and resistant to anti-myeloma chemotherapeutics.

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P48 Alterations in sphingomyelin:phosphatidylcholine ratio in chromatin preparations from rat liver and thymus cells after the cisplatin *in vivo* action

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It is well known, that chromatin lipids are implicated in various processes such as DNA replication, transcription, chromatin assembly, acetylation and methylation of histones. The regulatory effects of chromatin lipids exhibit concentration depending disposition.

The results of our earlier investigations were revealed the dramatically changes in absolute quantities of all phospholipid fractions of chromatin preparations from rat liver and thymus cells after the antitumor drug cisplatin *in vivo* action. Along with decrease in quantities of other phospholipid fractions, a special interest was rendered to interdependent alterations of two choline inclusive lipids, namely sphingomyelin and phosphatidylcholine. Comparative analysis of alterations in absolute quantities and ratio of sphingomyelin and phosphatidylcholine in chromatin preparations from rat liver and thymus cells after the 24h. *in vivo* action of cisplatin was carried out.

We showed that the sphingomyelin:phosphatidylcholine ratio in rat liver and thymus chromatin preparations was equal to 0.47 and 0.36 respectively. The cisplatin action leads to multidirectional alterations in choline inclusive lipids absolute content and in sphingomyelin: phosphatidylcholine ratio in the investigated chromatin preparations. Thus, in liver chromatin the value of this ratio decreased up to 0.30, which is equal to 36% of diminition. On the contrary, in case of rat thymus chromatin the cisplatin caused increase in sphingomyelin:phosphatidylcholine ratio up to 0.53 (increase by about 49%).

These multidirectional alterations of absolute content and the ratio of

chromatin choline inclusive lipids in rat liver and thymus may be explained by difference of metabolic status of these tissues as well as by differences in sensitivity to cisplatin treatment of enzymes, that catalyze the degradation of lipids in rat liver and thymus nuclei.

Taking into consideration the regulatory role of chromatin phospholipids as well as the crucial importance of sphingomyelin:phosphatidylcholine crosstalk in cell fate, one can assume that these cisplatin caused alterations may be connected with the antitumor effects of the drug.

P49 Screening for ATPase inhibitory factor 1 (IF1) regulatory drugs

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Targeting mitochondria represents a promising strategy for the development of new anti-cancer drugs and other mitochondrial pathologies. The ATPase Inhibitory Factor 1 (IF1) is a major player in the regulation of the activity of the H^+-ATP synthase and hence of OXPHOS. We have recently described, that the phosphorylation of S39 in IF1 abolishes its capacity to bind to the H^+-ATP synthase (1). Only dephosphorylated IF1 binds and inhibits both the hydrolase and synthase activities of the enzyme. Hence, the phosphorylation status of IF1 regulates the flux of aerobic glycolysis and ATP production by OXPHOS during cellular differentiation (2), cell cycle, hypoxia and upon an enhanced metabolic demand (1). These findings stress the emerging roles played by IF1 in metabolic reprogramming in different physiological contexts and in cancer and encourage us to identify potential drugs that could regulate the phosphorylation status of the protein. Herein, we have tested the effect of 1018 FDA-Approved drugs on the Oligomycin Sensitive Respiration (OSR) of HCT116 colon cancer cells using the XF^{e}96 Seahorse Analyser. This primary high-throughput screening only contemplates the analysis of OSR, which indirectly assesses the mitochondrial capacity to synthesize ATP by the H^+-ATP synthase. Since our primary focus was to identify compounds that could regulate the phosphorylation status of IF1 we have treated HCT116 cells during 3h with 1 µM of the drugs to assess a short-term response to the different compounds. In this primary screen, we have identified 162 hits that modulate the OSR by 40% when compared to control cells treated with the vehicle. We have further studied the effect of the 162 hits on Basal Respiration, OSR and Maximum Respiration of HCT116 cells treated as above indicated using the XF24 Seahorse Analyser. After this secondary screen, we have obtained 15 hits which clearly have a short term impact on mitochondrial respiration. At present, we are initiating the study of these compounds on the activity of the H^+-ATP synthase in permeabilized cells (3) and on the phosphorylation status of IF1 using 2D-gels and Phos-tag analysis. We plan to test some of these drugs in tumor growth using HCT116-luc cells subcutaneously injected in immunocompromised mice.

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