Fatty liver disease is a progressive metabolic disorder characterized, among other features, by accumulation of excessive lipids, leading to hepatocellular dysfunction. Despite extensive research, the role of autophagy in the development of fatty liver disease remains controversial, as autophagy regulates lipid metabolism and lipid droplet degradation. Transcription factor Nrf2 plays a protective role in fatty liver disease by regulating antioxidant defenses and lipogenic genes, which promote hepatosteatosis. However, the role of Nrf2, autophagy and their molecular crosstalk under lipotoxic conditions is incompletely defined.
The purpose of this study is to investigate the cross regulation of autophagy and Nrf2-Keap1 pathway under lipotoxic conditions and to estimate their involvement in lipid droplet formation and cellular dysfunction. Therefore, we induced lipid accumulation in HepG2 cells by exposing them to palmitic acid (PA). According to our study, autophagy plays a key role in the lipid droplet content. Western blot analysis of autophagy-related proteins revealed an impairment of the autophagic flux. Additionally, immunocytochemistry and western blot analysis indicated increased p62 levels and activation of Nrf2/Keap1 pathway. Under these conditions, Nrf2 knockdown experiments are being performed to elucidate the impact of Nrf2 on lipid droplet formation and autophagy.

102. Automated immuno-histo-enzymatic investigation of metabolic enzyme activity in skin and organotypic skin models shows epidermal metabolism regulation by metformin

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Metabolic activity of cells in tissues depends on type, nutrient availability, position, differentiation state, and other parameters. We developed an automated microscopy method that relates the cell individual enzymatic activity to marker expression and position within the epidermis. We adapted StrataQuest software (Tissuegnostics) to detect the epidermis and its strata based on nuclear density mapping and distance-based algorithms. The sections were IF-stained for differentiation and proliferation markers. The activity assay for the enzymes G6PD and GAPDH was performed with enzyme specific substrates and a tetrazolium based chromogenic dye. We could predict with high precision the basal, first suprabasal and stratum corneum layers of human epidermis and in organotypic skin equivalents and verify the differentiation state with markers. We could assign enzymatic activity of G6PD to single cells within the predicted strata. Addition of Metformin two days before harvest led to a significant increase in G6PD activity, whereas UVB pre-exposure of embedded Kerantinocytes led to reduced G6PD activity. In conclusion, we were able to present an automated assay for epidermal image analysis and could prove responsiveness of metabolic enzyme activity to Metformin which is not only used for treatment of diabetes but also the most investigated anti-aging drug.

103. Green tea epigallocatechin-3-gallate (EGCG) oxidative stress and DNA damage

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Epigallocatechin-3-gallate (EGCG) antioxidant properties have been demonstrated however, increasing evidence indicates that EGCG produces reactive oxygen species. The aim of this study was to evaluate the effect of EGCG intake during 90 days in hematological cardiovascular risk factors, vitamins A and E, DNA damage and oxidative damage in human blood. Peripheral blood from 30 healthy individuals (10 males and 20 females; 18 – 45 years), was collected at time 0 (T0) and time 90 (T90). During 90 days, participant's ingested capsules of green tea extract (225mg EGCG) daily. Hematological cardiovascular risk factors including lipid profile and liver function parameters were assessed using colorimetric methods. Vitamins A and E in serum were quantified by HPLC and analysis of DNA damage and oxidative damage was performed by comet assay. Our results showed that lipid profile and liver function parameters are not affected by EGCG and serum levels of vitamin E increased, but not vitamin A. An increase of DNA damage and DNA oxidative damage after 90 days of EGCG consumption was also reported. The results suggest that EGCG can induce DNA damage, possibly due to ROS induction, with associated increase of the antioxidant vitamin E, however without alteration of hematological cardiovascular risk factors.

104. Expression of the novel Apolipoprotein o is linked to triglyceride accumulation and oxidative stress in chicken and human hepatoma cells

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Non-alcoholic fatty liver disease (NAFLD) reflects a spectrum of chronic liver diseases characterized by hepatic fat accumulation. Apolipoprotein O (ApoO) is a new member of the apolipoprotein family that may play a role in lipid metabolism and mitochondrial electron transport activity.
We hypothesized that hepatic expression of ApoO is tightly linked not only to diet-induced hepatosteatosis, but also to oxidative stress and hormones.
Therefore, we compared the effects of lipid loading on ApoO regulation in chicken (LMH) with those in human (HepG2) hepatoma cells. Incubation with oleic acid (OA) induced triglyceride accumulation, but did not affect cell viability. RT-qPCR and Western blot analyses showed significant increase in ApoO transcript and protein levels in both cell lines. Oxidative stress applied by H_2O_2 revealed induction of ApoO in the same or even higher extent as monitored by OA. ApoO increased upon treatment with estrogen supporting the assumption that estrogen affects lipoprotein metabolism. Furthermore, both cell lines showed a significant decrease of the mitochondrial membrane potential upon incubation with OA.
We assume that our findings support a role of ApoO as an effector of compromised mitochondrial function that likely accompanies the onset of NAFLD.

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SFRR-E 2019 ANNUAL MEETING
"REDOX HOMEOSTASIS: FROM SIGNALING TO DAMAGE"

Abstracts of poster presenters in alphabetical sequence of first author

9. Unravelling the effect of N(ε)-(carboxymethyl)lysine (CML) and N(ε)-(carboxyethyl)lysine (CEL) on the ability of α-Synuclein to reduce the formation of Cu^{2+}-catalyzed reactive oxygen species

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α-Synuclein (αS) is a protein found in the neurons of the substantia nigra, where plays a role in neurotransmission and maintains the redox balance [1]. Moreover, αS is well-known to be involved in the development of Parkinson's disease (PD) since it forms the PD-associated intraneuronal fibrillary deposits, known as Lewy bodies (LB). Aggregation of αS is stimulated by mutations, metal chelation, oxidative damage and post-translational modifications. In fact, most LB isolated from post-mortem brains are modified through the glycation process. Although the effect of the advanced glycation end products (AGEs) on the aggregation of αS has been studied, there are not data reporting how the AGEs formed on αS affect the neuronal redox equilibrium. Consequently, we have studied how N(ε)-(carboxymethyl)lysine (CML) and N(ε)-(carboxyethyl)lysine (CEL) -two AGEs found in the neurons of PD models [2]- affected the capacity of αS to inhibit the formation of reactive oxygen species (ROS) from Cu^{2+}-catalysed ascorbic acid (AA) degradation, and to be oxidized by ROS. The obtained data clarify the effect on glycation on the capacity of αS to protect against oxidative damage.

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10. Functional evaluation of oxidative stress parameters in hypoxic premature born infants

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Objective: was to evaluate the intensity of oxidative stress in hypoxic premature born infants.

Material and method: Study group: 43 premature infants born in 2017, in „Bega" Hospital Timisoara.The inclusion criteria: gestational age under 32 weeks, Apgar score ≤ 6/ 5 min, lab analysis for metabolic acidosis ratio and clinical signs for hypoxia. The oxidative stress and antioxidant capacity evaluation was perfomed by d-ROM and BAP parameters (Panel Carratelli, Diacron Italy). Results: Values above 300 unit Carr for d-ROM are indicative for oxidative stress, 76% of patients had these values (the maximal level was 465 U Carr). The optimum value for BAP is higher than 2200 microM/l. At birth 28% of cases had a border-line condition for BAP. At 72 hours of life, d-ROM increased in 45% of the cases, but in parallel manner with the increase of BAP. In 8% of the patients, d-ROM remains high and BAP- under the normal level, in these patients the death occurred. Conclusion: This study pointed out a strong correlation between high d-ROM and/or low BAP level and the prognostic for the patient's disease.

11. Protective effects of L-carnitine against radiation-induced surface and gland epithelial degeneration in rat endometrium

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Purpose: The aim of the study was to detect the effects of L-carnitine (LC) on radiation-induced uterine injury.
Materials and methods: Thirty Wistar albino rats were divided into five groups. The control group received physiological saline intraperitoneally. Radiation-1 and radiation-2 groups received whole-body X-irradiation of 8.3 Gy as a single dose. These groups were sacrificed at the 6th hour and on the 4th day after irradiation, respectively. The radiation-1+LC and the radiation-2+LC groups received the same dose irradiation plus a daily dose of 200 mg/kg LC.
Results: The levels of serum MCP-1 and IFN-γ were significantly higher in the radiation groups than the control groups. Treatment with LC decreased the serum MCP-1 and IFN-γ levels considerably. Radiation induced flattening of the endometrial surface and glandular epithelium, depletion of deep glands, effects that were partially blocked by LC treatment. The expression of proinflammatory cytokines in uterine tissue were markedly stimulated in irradiated rats. In the radiation groups, PARP-1, IL-1β, IL-6, TNF-α and NFκB expression in the uterine tissue were

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