C1. DESCRIPTION FOR GENERAL PUBLIC

The entry into the 20th century is considered to be the beginning of the plague of diseases related to insulin resistance (IR) and pathological obesity - set of symptoms termed as metabolic syndrome (MS).

Increased frequency of MS and IR occurrence become a serious issue not only in human, but also in veterinary medicine. MS was found to affect horses (equine metabolic syndrome, EMS) overfed with high energy fodder concomitant together with reduced physical activity. Similarly to humans, typical EMS symptoms are as follow: pathological obesity (including abnormal accumulation of adipose tissue above eyes and tail base) IR, hyperinsulinemia and chronic, systemic inflammatory state. Unfortunately, no effective therapy for EMS treatment exists, which sometimes leads to the necessity of animal's euthanasia due to development of laminitis. What is important, additional mechanisms beyond the IR development in peripheral tissues of EMS horses have not been elucidated yet, which strongly limits the advancement of therapy and understanding of disease' aetiology at molecular level.

Our team possesses a thorough experience in studying EMS evidenced in scientific papers. So far, we have described inflammation-driven dysfunction of adipose tissue and impairment of adipose-derived stem cells (ASC) in EMS individuals.

In presented project, for the first time, we aim to determine the influence of EMS on functionality, fat infiltration and inflammation of the liver - phenomena widely reported to occur in human MS. EMS horses are characterized by increased levels of aspartate aminotransferase (AST), alanine transaminase (ALT) and γ-glutamyl transferase (GGT). What is more, liver cells display IR, inflammation, apoptosis and decreased chaperone mediated autophagy (CMA). Histological liver analysis revealed its infiltration with immune cells and excessive accumulation of lipid droplets within hepatocytes. Although no comprehensive data regarding liver condition in obese, insulin resistant horses with EMS exist. Our preliminary data strongly indicates that liver may play a central and crucial role in the development of EMS and related IR. Liver lipids overload, especially saturated free fatty acids and accumulation of their metabolites might be closely correlated with the development of hyperinsulinemia and IR. Thus improving functionality of EMS horses' hepatocytes through increasing insulin sensitivity and chaperone mediated autophagy may become effective therapeutic strategy in the course of EMS. For that reason, in presented project we aim to investigate in depth the IR development at molecular level and examine therapeutic utility of inhibition of two enzymes involved in insulin signalling - low molecular weight tyrosine phosphatase (LMPTP) and protein tyrosine phosphatase 1B (PTP1B) as their activity is increased during IR. Functional evaluation of components of insulin receptor signalling pathway may contribute to the development of innovative therapeutic strategies which would improve insulin sensitivity. The project consists of several distinct stages encompassing basic research and clinical veterinary practice, it will reveal mechanism of IR development in EMS horses and deliver the potential drug candidate for enhancement of insulin sensitivity during EMS. Classification of horses and biopsy harvesting (liver) are assigned to the task one. In the next step, proteins of insulin receptor signalling pathway and inflammation are going to be investigated in obtained tissue homogenates. Special emphasis will be put on pathological changes of the liver- accumulation of lipids and its infiltration with immune cells. Next stage will test and select an inhibitor with the best insulin sensitizing, anti-inflammatory and chaperone mediated autophagy modulation properties for further application in EMS-suffering horses. To test this, horse hepatocytes and HepG2 cell line will be cultured in the presence of PTP1B and LMPTP inhibitors: MSI-1436 (Trodusquemine) and "compound 23" (N,N-diethyl-4-(4-((3-(piperidin-1-yl)propyl)amino)quinolin-2-yl) benzamide) respectively. Based on obtained data, selected inhibitor will be applied to horses with EMS to further confirm its therapeutic usefulness.

Proposed research tasks will be carried out by a multidisciplinary team composed of specialized research groups, which include molecular biologists, veterinarians, chemists, immunologists and histologists. We are convinced that our combined experience will enable reliable and professional realisation of set goals. Data generated during the project will deliver important information regarding EMS aetiology at molecular level and extend our knowledge in the field of veterinary endocrinology in general. Most importantly, the project may help in developing innovative methods for MS treatment in humans.