C1. DESCRIPTION FOR GENERAL PUBLIC

Endocrine disorders have become more and more frequent diagnosed diseases in both, human and veterinary medicine. Horses overfed with high-energy and high-starch diet, whose physical activity is reduced usually develop metabolic syndrome (equine metabolic syndrome, EMS). Those individuals are characterized by insulin resistance, hyperleptinemia, hyperinsulinemia, chronic inflammation, accumulation of harmful reactive oxygen species (ROS) and excessive cortisol production. Moreover, inflammatory state within adipose tissue disturbs its homeostasis and alters the profile of secreted adipokines. All of that negatively affects stem cell population residing in adipose tissue (adipose-derived stem cells, ASC). Because of their unique features, including ability to differentiate into multiple cell lineages and immunomodulatory properties, interest in its application in regenerative medicine has been enormous. Multiple studies performed all over the World proved their beneficial effects in the treatment of musculoskeletal, metabolic and neurodegenerative disorders.

However, ASC' residing in unfriendly, pro-inflammatory microenvironment of EMS horses adipose tissue, lose their therapeutic potential. Our previous studies have shown that, ASC_{EMS} are characterized by decreased proliferation rate, accumulation of senescence-related factors, increased apoptosis and mitochondrial dysfunction. Mitochondria are small structures localized within cell body which role can be compared to power station- their most important role is to supply energy in the form of adenosine-5'-triphosphate ATP). Although, in ASC_{EMS} significant aberrations of those organelles morphology (vacuoles formation in matrix, cristae disintegration) and dynamics (increased fission) have been observed. Impairment of mitochondria functionality leads to disturbance of energy homeostasis and to excessive accumulation of reactive oxygen species. These in turn, through lipid and protein oxidation initiate the process of programmed cell death- apoptosis.

In presented project we aim to "renovate" and "rejuvenate" ASC_{EMS} cells through mitochondria transfer. Functional, undamaged organelles will be first isolated from ASC of healthy horses, next using author's method introduced to $ASC_{EMS.}$ We expect that conducting our procedure will improve metabolism, bring back multipotent properties and reverse senescence of $ASC_{EMS.}$ Moreover, we would like to investigate how long introduced mitochondria remain within host cells and if endogenous, damaged mitochondria will be preferably removed from cells by mitophagy. This will allow to answer the question whether cells will pick healthy mitochondria over damaged ones and maybe investigate the mechanism beyond that process. It is especially important in the context of the newest research which postulate that stem cells fulfil their therapeutic role *in vitro*, by naturally occurring mitochondria transfer. That's why mitochondria condition is crucial in the cells intended for therapy, for the sake of its effectiveness. "Exchange" in deteriorated mitochondria for "healthy" in ASC_{EMS} may become a milestone in enhancing their therapeutic potential as well as development of effective EMS and other disorders therapies. We expect that after mitochondrial transfer, ASC_{EMS} will recover their stemness and increase expression of multipotency-related genes. Furthermore, we assume that amount of reactive oxygen species will decrease with simultaneous increase of antioxidant proteins activity. What is especially important, we will investigate, for the first time fate and dynamics of transferred mitochondria in recipient cells.

Nowadays, when autologous grafts and cell therapies are still most desirable, "improvement" of cells designed for therapies by mitochondria transfer seems to be tempting perspective. Especially because patients health status significantly affects therapeutic utility of his stem cells. Age, metabolic diseases, lifestyle and diet, all of that influence cell' and its organelles metabolism. So, therefore presented in this research project method may found wide application in tissue engineering when condition of patients mitochondria is unsatisfactory and may significantly limit the quality of therapy. Moreover, obtained results will deliver valuable, preliminary data for *in vivo* experiments.