DESCRIPTION FOR THE GENERAL PUBLIC

A glomerulus is a tuft of small blood vessels (capillaries) located at the beginning of a nephron in the kidney. The unique, highly specialized and terminally differentiated glomerular visceral epithelial cells (podocytes) are an essential and integral part of the glomerular filter. These cells are the final barrier that restricts entry of plasma proteins from the circulation into the urine. Blood enters the glomerular capillaries and is filtered across the endothelium and the glomerular basement membrane (GBM) and through the slit diaphragms (SD) between podocyte foot processes to produce the primary urinary filtrate. In healthy glomeruli, this barrier restricts the passage of macromolecules but is permeable to water and small solutes. Injury to podocytes leads to proteinuria, a hallmark of most glomerular diseases. Because podocytes, similarly as neuronal cells, are not able to proliferate, they are the most vulnerable component of glomerular filter. The loss of podocytes from the kidney glomerulus is an early and key event in the development of diabetic nephropathy (DN), a chronic progressive disease that affects up to 40% of patients with diabetes mellitus. In advanced stage of the disease dialysis and even transplantation are necessary, which is related to growing costs of the treatment. In the last decade podocytes were found to be a target for insulin action, thus can be modulated by factors increasing or decreasing their sensitivity to insulin. Their insulin sensitivity is critical for glomerular function and the loss of correct insulin signaling in consequence leads to alterations and disorders featuring DN. Underlying mechanism of insulin resistance of podocytes induced by high glucose concentrations still remains unclear, thus the proposed project will undertake the attempt to explain the mechanism of impairment of glomerular podocytes function and induction of insulin resistance in podocytes exposed to hyperglycemic medium, mimicking diabetic conditions.

Although phosphorylation is probably the most extensively studied and ubiquitous protein modification in numerous biological pathways, the list of proteins that undergo acetylation–deacetylation cycle is increasing rapidly. Growing evidence is accumulating for a role of acetylation as a key coordinator of overall cellular metabolism. Up to now, more than 2000 acetylated proteins have been identified, and protein acetylation, a highly reversible process, has emerged as a key post-translational modification regulating the activity of multitude cellular proteins. Sirtuins are a conserved family of a deacetylases that are associated with numerous cellular signaling pathways. The most studied member of sirtuins family among the other members is deacetylase SIRT1. Many studies have shown a role of SIRT1 in the regulation of cellular insulin sensitivity. Our research demonstrated that in podocytes exposed to high glucose concentrations, SIRT1 protein amount and activity are decreased, with concomitant deterioration of podocytes insulin responsiveness and functions. SIRT1 protein expression and activity may be regulated by nitric oxide (NO). Recent studies indicated that phosphodiesterase 5 (PDE5) inhibition results in enhancement of NO/cGMP-dependent signaling pathway, activating its downstream cGMP-dependent protein kinase G (PKG) activity, leading in consequence to activation of SIRT1 and improvement of insulin sensitivity *in vitro* in murine pre-adipocytes, myoblasts and hepatocytes, and *in vivo* in diet-induced obese mice, however there is no data demonstrating the regulatory function of NO/cGMP/PKG/PDE5 pathway towards deacetylase SIRT1 in podocytes.

We will examine the possibility of restoration of SIRT1 protein level and activity, and then improvement of insulin responsiveness in podocytes cultivated in hyperglycemic conditions as a result of treatment with compounds modulating activities of PDE5 and PKG. We will also determine if SIRT1 activity influences activities of enzymes involved in cGMP-dependent pathway. Obtained results will approach us to novel knowledge about mechanisms occurring in the development of high glucose-induced insulin resistance of glomerular podocytes in diabetes. Characterizing pathways leading to insulin resistance would help in designing strategies to treat this problem.