Nephron is the basic structural and functional unit of the kidney. Each nephron is composed of an initial filtering component and a tubule specialized for reabsorption and secretion (proximal, distal convoluted tubules). Glomerular capillary pressure, and thus glomerular filtration rate, can be influenced by constriction or relaxation of the afferent arteriole, resulting in decreases or increases in pressure. The glomerular filtration barrier is composed of the fenestrated endothelium of the glomerular capillaries, the fused basal lamina of the endothelial cells and podocytes, and the filtration slits of the podocytes.
Diabetic kidney disease and its most severe manifestation, end-stage renal disease, remains one of the leading causes of reduced lifespan in people with diabetes. However, we currently lack in-depth understanding of the timing of onset and progression of kidney disease. Moreover, there are currently no reliable biomarkers for early detection of impaired kidney function, which can enable therapeutic interventions to prevent or slow disease progression. Two universally accepted hallmarks of kidney disease progression are decreased glomerular filtration rate and progressive albuminuria as a consequence of damaged glomerular filtration barrier.

The podocytes with their foot processes constitute an important cellular layer of glomerular barrier involved in regulation of glomerular permeability. Disturbance of podocytes function has a central role in the development of proteinuria in diabetic nephropathy. Retraction of podocyte foot processes, which form a slit diaphragm, is a common feature of proteinuria. At present, the correlation of retraction with the development of proteinuria in not well understood.

Early stages of type 2 diabetes are characterized by elevated insulin and glucose concentration. We demonstrated that both factors stimulate reactive oxygen species production, which leads to impairment of podocyte actin cytoskeleton, dysregulation of insulin signaling and disruption of the glomerular filtration barrier. Moreover, insulin resistance correlate with dysregulation of phosphate and calcium homeostasis and vascular calcification, but this mechanism is not known in podocytes. It has been demonstrated that pyrophosphate is an inhibitor of calcification routinely present in urine. Disturbance in in the balance between pyrophosphate and phosphate extracellularly causing hydroxyapatite deposition.

It should be mention that podocytes cell body is mostly located in glomerular ultrafiltrate fluid, devoid of fetuin, osteopontin and albumin, the inhibitors of calcification. Therefore, the pyrophosphate could, at least in part, contribute to the balance of inorganic phosphate in ultrafiltrate fluid, preventing hydroxyapatite formation. We hypothesized that NPP1 and αKlotho regulate the balance between pyrophosphate and inorganic phosphate concentration, and in consequence protect podocyte barrier against calcification and that this mechanism could be disrupted in diabetes.
The principal goal of the project is to define the role of αKlotho and NPP1 in the development of pathological changes in renal filtration barrier observed in type 2 diabetes, which eventually lead to diabetic nephropathy and kidney failure. The research project is divided into two parts: *in vivo* and in *vitro*. The experiments *in vitro* will be focused on investigation: cellular and molecular mechanism of Klotho and NPP1 interplay in podocyte, phosphate-calcium homeostasis, permeability to albumin across the podocytes monolayer, glomerular permeability to albumin. The major part of this project will be searching for new biomarkers of early diabetic nephropathy and podocytes damage in urine samples from rat ZDSD model which displays type 2 diabetes progression and from patients with type 2 diabetes. Realization of this project might help to understand the molecular mechanism of proteinuria and to further develop novel diagnostic markers to test early diabetic nephropathy and podocytes damage in diabetes.