Podocyte-specific proteins of extracellular vesicles excreted in the urine as potential markers of graft function after kidney transplantation

Chronic kidney disease as a result of many systemic diseases such as diabetes, hypertension, atherosclerosis, and kidney disease, e. g. glomerulonephritis, in the late phase of the disease, may lead to renal failure requiring renal replacement therapy – dialysis or kidney transplantation, which, taking into account the quality of life and mortality of patients, is currently the optimal method of the therapy. Nevertheless, the renal graft survival - short (occurring up to a year after transplantation) and long-term, remains a constant problem in management of transplant recipients. The short-term renal graft survival has significantly improved over the past three decades. However, the long-term survival is still unsatisfactory and it is estimated that approximately 3% of kidney graft transplant recipients return to dialysis and require retransplantation. The long-term function loss of the renal graft can be attributed to a number of mechanisms, including non-immune damage to the function of the glomerulus, resulting from a decrease in the number of podocytes up to 30-40 % of normal values, and importantly, the loss of podocyte may be initiated in the early posttransplant period.

The contemporary challenge for transplantology is to prolong the survival of patients and grafts and to improve the function of the transplanted kidney, as well as to search for markers that precede the occurrence of decline in graft function. Currently, the assessment of renal graft function is based on the serum creatinine value, estimated glomerular filtration rate (eGFR) and proteinuria. Therefore, the search for new factors enabling the early diagnosis of the transplanted kidney function, combined with the assessment of the risk of graft loss, seems to be justified. Non-invasive markers that can help in monitoring renal graft function and determining the cause of graft loss are of great interest. These searches could be based on reports showing that losing podocytes - glomerular cells - with urine is a more specific marker of glomerular damage than proteinuria. Furthermore, the fragments of the cell membrane from the podocytes attached to glomeruli can be encapsulated in extracellular vesicles and be excreted into the urine as extracellular vesicles. Preliminary studies done by our team have shown that the creatinine-standardized number of extracellular vesicles excreted in the urine was 1.5-fold higher in patients in the early period after transplantation compared to healthy volunteers. There are also indications that these vesicles also differ in their protein composition – podocin and nephrin. Thus, we think that the quantitative as well as qualitative characterization of urinary extracellular vesicles of podocyte origin will allow us to determine the extent of urinary podocyte loss in the early period after kidney transplantation and, in consequence, to predict the distant function of the transplanted kidney.

The project will conduct experiments to elucidate whether proteins of urinary extracellular vesicles of podocyte origin may be used as potential early markers of renal graft function in patients after kidney transplantation. For this purpose, the extracellular vesicles will be isolated from the urine of patients in the early period after kidney transplantation (1, 2, 4 and 12 weeks) and healthy volunteers. Their presence will be confirmed by detecting proteins specific for extracellular vesicles and proteins indicative of podocyte origin. The dynamics of their changes will be compared with indicators of early posttransplant period graft function (after 1 year). Moreover, the urine excretion of total protein, creatinine, albumin, solubilized nephrin and podocin will be determined.

The results obtained from the presented experiments will allow for more precise assessment of the function of renal transplantation, which may enable the introduction of new diagnostic procedures that may contribute to the prolongation of renal graft survival.