"Searching for mutations in *COL4A3, COL4A4* and *COL4A5* genes and defining their impact on clinical outcome in a national cohort of Polish families with hematuria with the use of next-generation sequencing technology"

Hereditary glomerulopathies are manifested by glomerular hematuria present in several members of the same family. Two main renal diseases associated with glomerular familial hematuria are Alport Syndrome (AS) and thin basement membrane nephropathy (TBMN). AS is classified as a rare disease. Affected families encounter typical problems and frustrations of a disease which only few specialists have gained expertise in. AS has been described many years ago as a progressive glomerulopathy with proteinuria, hypertension and loss of renal function requiring dialysis and renal transplantation, accompanied by hearing loss and ocular changes. In contrast individuals with TBMN in the majority of cases demonstrate only isolated hematuria, sometimes with slight proteinuria. TBMN is looked upon as a mild disease.

Both diseases are caused by anomalies in the structure of collagen IV α chains – a basic component of basement membranes. In glomeruli, inner ear and eye structures, basement membranes have an unique collagen IV composition of α3, α4 and α5 chains. The presence of abnormal or dysfunctional chains leads to renal and ocular damage and hearing loss. Abnormalities of α chains are caused by mutations in several genes. The gene encoding α5 (*COL4A5*) is present on the chromosome X and its mutations are responsible for X-link mode of inheritance of AS. The genes encoding α4 and α3 chains (*COL4A4* and *COL4A3*) are located on chromosome 2 and cause autosomal recessive AS (when both parents pass one mutation to a child). Mutations in these two last genes are responsible also for TBMN, which is transmitted in autosomal dominant pattern. Individuals with TBMN are considered carriers of autosomal recessive AS. In addition, there are contradictory opinions about occurrence of autosomal dominant mode of inheritance of AS, which demand further studies.

Early distinction beetween mild and serious causes of familial hematuria is not easy on the basis of clinical symptoms alone or in combination with renal biopsy findings. The characteristic clinical symptoms are seen in the second and third decade of life; whereas biopsy findings are frequently not characteristic and rather unreliable for diagnosis. During the first decade of an affected individual's life the only symptom is usually microscopic hematuria. Today next generation sequencing (NGS) studies enable an early and accurate diagnosis in 40-50% of families. NGS studies are relatively cheap, precise and available quickly. The recognition of the type of mutation and inheritance pattern in a family in correlation with clinical presentations, allows the differentiation between AS and TBMN. It may also assist in identifying individuals with a worse prognosis who may benefit from early start of nephroprotective therapy.

In Poland the diagnosis of AS and TBMN has been based on clinical findings and renal biopsy results. The final diagnosis has been made therefore very late, after many years of anxiety concerning the future prognosis of the child. Diagnostic genetic studies have not been to date available. The proposed multicentre study (involving the majority of paediatric nephrology medical centers in Poland) organised by the Medical University of Gdansk aims at recognizing the *COL4A3, COL4A4* and *COL4A5* mutations present in Polish population with familial hematuria. A national Registry for AS and TBMN has been set up for on-line clinical and histopathological data collection. A pilot study for the presence of *COL4A3, COL4A4* or *COL4A5* mutations has been performed in 38 children with familial hematuria and has yielded positive results in 89% tested. Further genetic studies are planned to allow recognition of the existing profile of mutations in Polish families. This should enable precise genetic counselling and assessment of long term prognosis. The genetic studies may also aid in the selection of patients benefiting from early interventional treatment. In addition, realisation of the project will select a group of families with no recognised mutation - for further genetic studies to be performed through the ERKNet (European Reference Network for rare kidney diseases), to which the Gdansk centre belongs.