# Genetic testing of *WT1, NPHS2* and *NPHS1* genes in Czech pediatric patients with steroid-resistant nephrotic syndrome identified the genetic background in 25% cases

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## Introduction and aims

Steroid-resistant nephrotic syndrome (SRNS) is the second most frequent chronic kidney disease in the first three decades of life. Idiopathic forms may respond to calcineurin inhibitors, whereas genetic forms are usually resistant to this treatment. The risk of recurrence after renal transplant is 30-40% in patients with idiopathic SRNS, this risk is minimal in patients with genetic forms. Recessive or dominant mutations in over than 40 genes were identified as causes of monogenic forms of SRNS. *WT1* and *NPHS2* are the most frequent genes mutated in children with SRNS older than 3 months at the time of manifestation, whereas mutations in *NPHS1* are the most common reason in children with manifestation in the first 3 months of life (congenital NS). The aim of the study was to test patients from Czech Republic and by molecular-genetic methods.

## Methods and patients

The DNA samples from 82 pediatric patients (0-19 years of age) from Czech Republic were tested by direct Sanger sequencing for genes *WT1* (exon 8 and 9) and *NPHS2* (whole gene). In 8 patients with infantile SRNS without mutation of *WT1* or *NPHS2* the *NPHS1* gene was tested.

## Results - mutations

| NPHS2 | | |
|---|---|---|
| Nucleotide change | Effect on protein | Number of patients |
| c.[413G>A];[413G>A] | p.[(Arg138Gln)];[(Arg138Gln)] | 6 |
| c.[59C>T];[59C>T] | p.[(Pro20Leu)];[(Pro20Leu)] | 1 |
| c.[523C>A];[523C=] | p.[(Pro175Thr)];[Pro175=] | 1* |
| c.[868G>A];[868G=] | p.[(Val290Met)];[Val290=] | 2* |
| c.[952G>A];[952G=] | p.[(Ala318Thr)];[Ala318=] | 1* |
| c.[948delT];[948T=] | | 1* |
| c.413G>A(;)868G>A | p.(Arg138Gln)(;)(Val290Met) | 1 |
| c.413G>A(;)1150T>C | p.(Arg138Gln)(;)(Term384Gln) | 1 |
| c.413G>A(;)419delG | p.(Arg138Gln)(;)? | 2 |
| | | Total number 16 |
| **NPHS1** | | |
| Nucleotide change | Effect on protein | Number of patients |
| c.[-475_-468del];[-475_-468=] | | 1* |
| c.[3230A>G];[3230A=] | p.[(Asn1077Ser)];[Asn1077=] | 2* |
| c.397+3A>G(;)1103C>T | | 1 |
| c.[1627+34G>A];[1627+34G=] | | 1* |
| | | Total number 5 |
| **WT1** | | |
| Nucleotide change | Effect on protein | Number of patients |
| c.[1406A>G];[1406A=] | p.[(His469Arg)];[His469=] | 1 |
| c.[1283G>A];[1283G=] | p.[(Cys428Tyr)];[Cys428=] | 1 |
| c.[1372C>T];[1372C=] | p.[(Arg458Term)];[Arg458=] | 1 |
| c.[1340-28C>T];[1340-28C=] | | 1 |
| c.[1301G>C];[1301G=] | p.[(Arg434Pro)];[Arg434=] | 2 |
| c.[1432+4C>T ];[1432+4C=] | | 1 |
| c.[1384C>T];[1384C=] | p.[(Arg462Trp)];[Arg462=] | 1 |
| c.[1325A>C];[1325A=] | p.[(Gln442Pro)];[Gln442=] | 1 |
| c.[1390G>A];[1390G=] | p.[(Asp464Asn)];[Asp464=] | 1 |
| | | Total number 10 |

*1 heterozygous mutation

## Results

We found heterozygous mutations of *WT1* gene in 9/82 patients (11%). These were 7 unrelated patients with different mutations and 2 twin sisters with the identical mutation. Seven patients progressed to end-stage renal disease (ESRD), 4 patients presented with Wilms tumour. We found homozygous or compound heterozygous mutations of *NPHS2* gene in 10/82 patients (12%). One patient carried a causal heterozygous mutation in *NPHS2* and the R229Q polymorphism. One patient had a combination of a heterozygous mutation in *WT1* (H469) and a homozygous mutation in the *NPHS2* gene (P20L). Six patients progressed to ESRD. Compound heterozygous mutation in *NPHS1* gene (c.397+3A>G(;)1103C>T) was found in one of eight patients with congenital NS.

## Conclusion

Mutations in WT1, NPHS2 and NPHS1 genes are responsible for 25% of cases of childhood SRNS, this finding corresponds with results of other populations published in literature. Immunosuppressive treatment is not indicated in patients with genetic SRNS. Molecular genetic analysis is thus essential both for proper management of patients with SRNS and for outcome estimation.


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