Nephrotic Syndrome and Focal Segmental Glomerulosclerosis (NS/FSGS)

Panel Gene List (55 genes): ACTN4, ADCK4 (COQ8B), ALG1, ANLN, APOL1*, ARHGAP24, ARHGDIA, CD2AP, COL4A3, COL4A4, COL4A5, COQ2, COQ6, CRB2, CUBN, DGKE, EMP2, FAN1, FAT1, FN1, GLA, INF2, ITGA3, ITGB4, KANK1, KANK2, KANK4, LAMB2, LMX1B, MAGI2, MYH9, MYO1E, NEIL1, NPHP1, NPHS1, NPHS2, NUP107, NUP205, NUP93, OCRL, PAX2, PDSS2, PLCE1, PMM2, PTPRO, SCARB2, SGPL1, SMARCAL1, STS, TBC1D8B, TRPC6, TTC21B, WDR73, WT1, XPO5

*G1 and G2 Risk Alleles Only

Clinical Features and Genetics:
Nephrotic syndrome (NS) is a clinically heterogeneous group of disorders characterized by proteinuria, hypertension, hypoalbuminemia, hypercholesterolemia and edema, which can ultimately lead to end-stage renal disease (ESRD). NS affects approximately 1-3 per 100,000 individuals worldwide and is considered one of the most common glomerular kidney diseases in children and adults.\(^1\)\(^-\)\(^2\) Classification of NS may be based on an individual's response to steroids or according to pathologic findings on renal biopsy.\(^3\) Approximately 5-10% of individuals with NS, including approximately 10-15% of all affected children, do not respond to steroid therapy and are classified to have steroid-resistant nephrotic syndrome (SRNS), as opposed to steroid-sensitive nephrotic syndrome (SSNS).\(^4\) Many studies have demonstrated that the incidence of disease in the population varies by age. Approximately 80% of individuals presenting with SRNS under the age of 1 have a genetics diagnosis, while this number drops to 26.5% for individuals presenting in childhood. Onset in adulthood is not as well studied and presentation is highly variable.\(^3\)

On renal biopsy, the most common histopathologic changes observed in individuals with NS are either minimal change disease (MCD, most common in children with NS) or focal segmental glomerulosclerosis (FSGS, most common in adults with NS). FSGS is characterized by scarring in a limited number of kidney glomeruli and accounts for about 40% and 20% of adult and pediatric cases of NS, respectively.\(^2,5\)\(^-\)\(^6\) While most cases of NS are reported to be sporadic due to secondary causes (such as diabetes, autoimmune disease, medication and infections), familial occurrence has been reported.\(^7\)\(^-\)\(^9\) In the literature, the clinical term NS and the pathologic term FSGS are not always separated and might refer to the same disease entity. FSGS is a pattern of renal injury, and while some patients may also have nephrotic syndrome, others may only present with mild proteinuria.\(^10\) In this panel, we have grouped together the NS and FSGS conditions to encompass a wider spectrum of renal disease. Overall, pathogenic variants in the ACTN4, ADCK4 (COQ8B), ARHGAP24, ARHGDIA, CD2AP, INF2, LAMB2, MYO1E, NPHS1, NPHS2, PLCE1, TRPC6 and WT1 genes are well-
established in the literature in association with NS/FSGS. The majority of cases of SRNS are caused by variants in NPHS1, NPHS2 and WT1, while the majority of cases of FSGS are caused by variants in ACTN4 and INF2.11-12

The presence of specific variants in the APOL1 gene (c.1024A>G, p.Ser342Gly, a.k.a. G1 allele; c.1164_1169delTTATAA, p.Asn388_Tyr389del, a.k.a. G2 allele) is associated with increased risk of ESRD among individuals of recent African ancestry. Individuals with two risk alleles (homozygotes or compound heterozygotes) have a 10-17 fold higher odds of developing FSGS type 4 and a 29 fold higher risk of developing HIV-associated nephropathy, an increased risk of sickle cell-associated nephropathy, and shortened graft survival of kidney transplants based on the donor’s genotype.13-20 Of note, an individual positive for both G1 and G2 alleles is assumed to be a compound heterozygous because these alleles are in complete negative linkage disequilibrium.15

NS/FSGS may appear as a renal-only phenotype or may show features that overlap with other multi-system conditions, as in Alport syndrome (AS) and Coenzyme Q10 deficiency (CoQ10 deficiency). AS is characterized by progressive renal disease, mild to moderate sensorineural hearing loss, and ocular features.21 Renal manifestations include hematuria, proteinuria, ultrastructural changes of the glomerular basement membrane and progression to ESRD.22 Ocular symptoms are common and can include anterior lenticonus, central and peripheral fleck retinopathies, temporal retinal thinning and recurrent corneal erosions.23 Recent literature has shown that variants in COL4A3, COL4A4 and COL4A5 genes, which are well-established AS genes, are additionally associated with FSGS and these patients are more likely to present with hematuria, family history and glomerular basement membrane abnormalities.24 Coenzyme Q10 is involved in multiple cellular processes including mitochondrial respiratory activity. Defects in the nuclear encoded ADCK4 (COQ8B), COQ2, COQ6 and PDSS2 genes cause primary CoQ10 deficiency, an autosomal recessive mitochondrial disorder that presents with SRNS, encephalopathy and slowly-progressive multiple system atrophy-like phenotype. Age of onset ranges from birth to the seventh decade and may vary in severity.25 Primary CoQ10 deficiency accounts for up to 2% of NS.26

Other genes previously reported to be associated with NS/FSGS and/or with pathogenic variants causing similar presentation included in this panel are ALG1, ANLN, CRB2, CUBN, DGKE, EMP2, FAN1, FAT1, FN1, GLA, ITGA3, ITGB4, KANK1, KANK2, KANK4, LMX1B, MAGI2, MYH9, NEIL1, NPHP1, NUP107, NUP205, NUP93, OCRL, PAX2, PMM2, PTPRO, SCARB2, SGPL1, SMARCAL1, STS, TBC1D8B, TTC21B, WDR73 and XPO5.

Test Methods:
Genomic DNA is extracted from the submitted specimen. For skin punch biopsies, fibroblasts are cultured and used for DNA extraction. The DNA is enriched for the complete coding
regions and splice site junctions of the genes on this panel using a proprietary targeted capture system developed by GeneDx for next-generation sequencing with CNV calling (NGS-CNVT). The enriched targets are simultaneously sequenced with paired-end reads on an Illumina platform. Bi-directional sequence reads are assembled and aligned to reference sequences based on NCBI RefSeq transcripts and human genome build GRCh37/UCSC hg19. After gene specific filtering, data are analyzed to identify sequence variants and most deletions and duplications involving coding exons at the exon-level; however, technical limitations and inherent sequence properties effectively reduce this resolution for some genes. For the APOL1 gene, only the G1 and G2 risk alleles are evaluated. For the SGPL1 and TBC1D8B genes, sequencing but no copy number testing is performed. For the ALG1 and ARHGDA genes, only whole gene deletions or duplications may be reported. Alternative sequencing or copy number detection methods are used to analyze regions with inadequate sequence or copy number data by NGS. Reportable variants include pathogenic variants, likely pathogenic variants and variants of uncertain significance. Likely benign and benign variants, if present, are not routinely reported but are available upon request.

Technical Test Sensitivity:
The technical sensitivity of sequencing is estimated to be >99% at detecting single nucleotide events, but less for deletions greater than 20 base pairs, insertions or rearrangements greater than 10 base pairs, or low-level mosaicism. The copy number assessment methods used with this test identify most deletions and duplications involving coding exons but are less reliable for detecting copy number variants of less than 500 base pairs. Assessment of copy number events also depends on the inherent sequence properties of the targeted regions, including shared homology and exon size. Mosaicism detection is limited and balanced chromosome aberrations cannot be identified.

Clinical Test Sensitivity:
NS/FSGS is a genetically heterogeneous disorder. The clinical sensitivity of sequencing and deletion/duplication analysis of the genes included in this panel depends in part on the patient’s clinical phenotype and family history. Additional information about the general clinical sensitivity of each gene is included in the table below.

<table>
<thead>
<tr>
<th>Gene</th>
<th>Protein</th>
<th>Inheritance</th>
<th>Disease Association</th>
<th>Sensitivity</th>
</tr>
</thead>
<tbody>
<tr>
<td>ACTN4</td>
<td>Actinin, Alpha-4</td>
<td>AD</td>
<td>FSGS 1</td>
<td>~3% of FSGS^12</td>
</tr>
<tr>
<td>ADCK4</td>
<td>Coenzyme Q8B</td>
<td>AR</td>
<td>NS 9</td>
<td>34/79 individuals with CoQ10 Deficiency^25</td>
</tr>
<tr>
<td>ALG1</td>
<td>Asparagine-Linked</td>
<td>AR</td>
<td>CDG Type 1k</td>
<td>~2-7% of SRNS^27-29</td>
</tr>
</tbody>
</table>

207 Perry Parkway, Gaithersburg, MD 20877 | P: 301-519-2100 | F: 201-421-2010 | E: genedx@genedx.com

www.genedx.com
<table>
<thead>
<tr>
<th>Gene</th>
<th>Protein Name</th>
<th>Disorder Type</th>
<th>Disorder</th>
<th>Allele Frequency</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>ANLN</strong></td>
<td>Actin-Binding Protein Anillin</td>
<td>AD</td>
<td>FSGS 8</td>
<td>~1% of FSGS</td>
</tr>
<tr>
<td><strong>APOL1</strong></td>
<td>Apolipoprotein L-I</td>
<td>AR</td>
<td>G1 and G2 Risk Alleles for ESRD / FSGS</td>
<td>In large population databases, the G1 and G2 alleles account for about 14% and 23% of APOL1 alleles among individuals with African ancestry, respectively.</td>
</tr>
<tr>
<td><strong>ARHGAP24</strong></td>
<td>Rho GTPase-Activating Protein 24</td>
<td>AD</td>
<td>FSGS</td>
<td>Rare</td>
</tr>
<tr>
<td><strong>ARHGDIAD</strong></td>
<td>Rho GDP-Dissociation Inhibitor Alpha</td>
<td>AR</td>
<td>NS 8</td>
<td>Rare</td>
</tr>
<tr>
<td><strong>CD2AP</strong></td>
<td>CD2 Associated Protein</td>
<td>AD/AR</td>
<td>FSGS 3</td>
<td>Rare</td>
</tr>
<tr>
<td><strong>COL4A3</strong></td>
<td>Collagen, type IV, alpha-3</td>
<td>AD/AR</td>
<td>AS</td>
<td>~12-15% of AS</td>
</tr>
<tr>
<td><strong>COL4A4</strong></td>
<td>Collagen, type IV, alpha-4</td>
<td>AD/AR</td>
<td>AS</td>
<td>~5-8% of AS</td>
</tr>
<tr>
<td><strong>COL4A5</strong></td>
<td>Collagen, type IV, alpha-5</td>
<td>XL</td>
<td>AS / AS-DL / AMME</td>
<td>~80-85% of AS</td>
</tr>
<tr>
<td><strong>COQ2</strong></td>
<td>Coenzyme Q2, Prenyltransferase</td>
<td>AR</td>
<td>Primary CoQ10 Deficiency 1</td>
<td>10/79 individuals with CoQ10 Deficiency</td>
</tr>
<tr>
<td><strong>COQ6</strong></td>
<td>Coenzyme Q10 Monooxygenase 6</td>
<td>AR</td>
<td>Primary CoQ10 Deficiency 6</td>
<td>5/79 individuals with CoQ10 Deficiency</td>
</tr>
<tr>
<td><strong>CRB2</strong></td>
<td>Crumbs Cell Polarity Complex Component 2</td>
<td>AR</td>
<td>FSGS 9 / Ventriculomegaly with Cystic Kidney Disease</td>
<td>Rare</td>
</tr>
<tr>
<td><strong>CUBN</strong></td>
<td>Cubilin</td>
<td>AR</td>
<td>Megaloblastic Anemia-1, Finnish type</td>
<td>Rare</td>
</tr>
<tr>
<td><strong>DGKE</strong></td>
<td>Diacylglycerol Kinase Epsilon</td>
<td>AR</td>
<td>NS 7</td>
<td>Rare</td>
</tr>
<tr>
<td><strong>EMP2</strong></td>
<td>Epithelial Membrane Protein 2</td>
<td>AR</td>
<td>NS 10</td>
<td>Rare</td>
</tr>
<tr>
<td><strong>FAN1</strong></td>
<td>FANC and FANC Associated Nuclease 1</td>
<td>AR</td>
<td>Interstitial Nephritis</td>
<td>Rare</td>
</tr>
<tr>
<td><strong>FAT1</strong></td>
<td>FAT Atypical Cadherin 1</td>
<td>AR</td>
<td>CAKUT / NS</td>
<td>Rare</td>
</tr>
<tr>
<td><strong>FN1</strong></td>
<td>Fibronectin 1</td>
<td>AD</td>
<td>Glomerulopathy with Fibronectin Deposits</td>
<td>Rare</td>
</tr>
<tr>
<td>Gene</td>
<td>Description</td>
<td>Allele</td>
<td>Disease</td>
<td>Percentage</td>
</tr>
<tr>
<td>--------</td>
<td>--------------------------------------</td>
<td>--------</td>
<td>---------</td>
<td>------------</td>
</tr>
<tr>
<td>GLA</td>
<td>Galactosidase, Alpha</td>
<td>XL</td>
<td>Fabry disease</td>
<td>~90-100% of Fabry Disease⁵⁶</td>
</tr>
<tr>
<td>INF2</td>
<td>Inverted Formin 2</td>
<td>AD</td>
<td>FSGS 5</td>
<td>~10% of FSGS¹²</td>
</tr>
<tr>
<td>ITGA3</td>
<td>Integrin, Alpha-3</td>
<td>AR</td>
<td>ILNEB</td>
<td>Rare¹¹,⁵⁷-⁵⁸</td>
</tr>
<tr>
<td>ITGB4</td>
<td>Integrin, Beta-4</td>
<td>AR</td>
<td>Epidermolysis Bullosa</td>
<td>Rare¹¹,⁵⁹-⁶⁰</td>
</tr>
<tr>
<td>KANK1</td>
<td>KN Motif and Ankyrin Repeat Domains 1</td>
<td>AR</td>
<td>NS</td>
<td>Rare⁶¹</td>
</tr>
<tr>
<td>KANK2</td>
<td>KN Motif and Ankyrin Repeat Domains 2</td>
<td>AR</td>
<td>NS 16</td>
<td>Rare⁶¹</td>
</tr>
<tr>
<td>KANK4</td>
<td>KN Motif and Ankyrin Repeat Domains 4</td>
<td>AR</td>
<td>NS</td>
<td>Rare⁶¹</td>
</tr>
<tr>
<td>LAMB2</td>
<td>Laminin, Beta-2</td>
<td>AR</td>
<td>NS 5 / Pierson syndrome</td>
<td>Rare¹¹,⁶²-⁶⁴</td>
</tr>
<tr>
<td>LMX1B</td>
<td>LIM Homeobox Transcription Factor 1, Beta</td>
<td>AD</td>
<td>Nail-Patella syndrome</td>
<td>~95% of Nail-Patella syndrome⁶⁵</td>
</tr>
<tr>
<td>MAGL2</td>
<td>Membrane-Associated Guanylate Kinase, WW And PDZ Domains-Containing, 2</td>
<td>AR</td>
<td>NS 15</td>
<td>Rare⁶⁶</td>
</tr>
<tr>
<td>MYH9</td>
<td>Myosin, Heavy Chain 9, Nonmuscle</td>
<td>AD</td>
<td>AS-like syndrome with Macrothrombocytopenia</td>
<td>~98% of MYH9-Related Disorders⁶⁷</td>
</tr>
<tr>
<td>MYO1E</td>
<td>Myosin 1E</td>
<td>AR</td>
<td>FSGS 6</td>
<td>Rare⁶⁸-⁶⁹</td>
</tr>
<tr>
<td>NEIL1</td>
<td>Endonuclease VIII-Like 1</td>
<td>AR</td>
<td>NS</td>
<td>Rare⁶⁹</td>
</tr>
<tr>
<td>NPHP1</td>
<td>Nephrocystin 1</td>
<td>AR</td>
<td>JS 4 / NPHP 1 / SLS 1</td>
<td>~15-20% of NPHP-RC⁷⁰-⁷¹</td>
</tr>
<tr>
<td>NPHS1</td>
<td>Nephrin</td>
<td>AR</td>
<td>NS 1</td>
<td>~5-14% of SRNS⁴,²⁸,⁷²</td>
</tr>
<tr>
<td>NPHS2</td>
<td>Podocin</td>
<td>AR</td>
<td>NS 2</td>
<td>~10-40% of SRNS¹¹,²⁸,⁷²</td>
</tr>
<tr>
<td>NUP107</td>
<td>Nucleoporin, 107-KD</td>
<td>AR</td>
<td>GAMOS 7 / NS 11</td>
<td>Rare⁷³,⁷⁴</td>
</tr>
<tr>
<td>NUP205</td>
<td>Nucleoporin, 205-KD</td>
<td>AR</td>
<td>NS 13</td>
<td>Rare⁷⁵</td>
</tr>
<tr>
<td>NUP93</td>
<td>Nucleoporin, 93-KD</td>
<td>AR</td>
<td>NS 12</td>
<td>Rare⁷⁵</td>
</tr>
<tr>
<td>OCRL</td>
<td>Inositol Polyphosphate 5-Phosphatase OCRL-1</td>
<td>XL</td>
<td>Lowe syndrome / Dent disease</td>
<td>~15% of Dent Disease⁷⁶</td>
</tr>
<tr>
<td>PAX2</td>
<td>Paired Box 2</td>
<td>AD</td>
<td>FSGS 7 / Papillorenal syndrome</td>
<td>~4% of FSGS⁷⁸</td>
</tr>
<tr>
<td>PDSS2</td>
<td>Prenyl Diphosphate Synthase, Subunit 2</td>
<td>AR</td>
<td>Primary CoQ10 Deficiency 3</td>
<td>2/79 individuals with</td>
</tr>
<tr>
<td>Marker</td>
<td>Gene Symbol</td>
<td>Description</td>
<td>Inheritance</td>
<td>Phenotype</td>
</tr>
<tr>
<td>-------</td>
<td>-------------</td>
<td>-------------</td>
<td>-------------</td>
<td>-----------</td>
</tr>
<tr>
<td>PLCE1</td>
<td>Phospholipase C, Epsilon-1</td>
<td>AR</td>
<td>NS 3</td>
<td>29/45 individuals with NS/FSGS&lt;sup&gt;11&lt;/sup&gt;</td>
</tr>
<tr>
<td>PMM2</td>
<td>Phosphomannomutase 2</td>
<td>AR</td>
<td>CDG Type 1a</td>
<td>~90-100% of CDG Type 1a&lt;sup&gt;30,79-80&lt;/sup&gt;</td>
</tr>
<tr>
<td>PTPRO</td>
<td>Protein-Tyrosine Phosphatase, Receptor-Type, O</td>
<td>AR</td>
<td>NS 6</td>
<td>Rare&lt;sup&gt;81&lt;/sup&gt;</td>
</tr>
<tr>
<td>SCARB2</td>
<td>Scavenger Receptor Class B, Member 2</td>
<td>AR</td>
<td>AMRF</td>
<td>~97% of AMRF&lt;sup&gt;82&lt;/sup&gt;</td>
</tr>
<tr>
<td>SGPL1</td>
<td>Sphingosine-1-Phosphate Lyase 1</td>
<td>AR</td>
<td>NS 14</td>
<td>35 families with FSGS&lt;sup&gt;43&lt;/sup&gt;</td>
</tr>
<tr>
<td>SMARCAL1</td>
<td>SWI/SNF-Related, Matrix-Associated, Actin-Dependent Regulator of Chromatin, Subfamily A-Like Protein 1</td>
<td>AR</td>
<td>SIOD</td>
<td>13/18 individuals with FSGS&lt;sup&gt;11&lt;/sup&gt; ~90% of SIOD&lt;sup&gt;84&lt;/sup&gt;</td>
</tr>
<tr>
<td>STS</td>
<td>Steroid Sulfatase</td>
<td>XL</td>
<td>Ichthyosis (with or without NS)</td>
<td>3 individuals with NS and STS deletion&lt;sup&gt;85-87&lt;/sup&gt;</td>
</tr>
<tr>
<td>TBC1D8B</td>
<td>TBC1 Domain Family, Member 8B</td>
<td>XL</td>
<td>NS 20</td>
<td>Rare&lt;sup&gt;88&lt;/sup&gt;</td>
</tr>
<tr>
<td>TRPC6</td>
<td>Transient Receptor Potential Cation Channel, Subfamily C, Member 6</td>
<td>AD</td>
<td>FSGS 2</td>
<td>~2% of FSGS&lt;sup&gt;12&lt;/sup&gt;</td>
</tr>
<tr>
<td>TTC21B</td>
<td>Tetraprecipitate Repeat Domain-Containing Protein 21b</td>
<td>AR</td>
<td>NPHP 12 / FSGS</td>
<td>10/61 individuals with FSGS&lt;sup&gt;89-90&lt;/sup&gt;</td>
</tr>
<tr>
<td>WDR73</td>
<td>WD Repeat-Containing Protein 73</td>
<td>AR</td>
<td>GAMOS 1</td>
<td>~1% of SRNS&lt;sup&gt;91-92&lt;/sup&gt;</td>
</tr>
<tr>
<td>WT1</td>
<td>WT1 Transcription Factor</td>
<td>AD</td>
<td>NS 4 / DD / FS</td>
<td>~2-4% of SRNS&lt;sup&gt;2,11,28&lt;/sup&gt;</td>
</tr>
<tr>
<td>XPO5</td>
<td>Exportin 5</td>
<td>AR</td>
<td>NS</td>
<td>Rare&lt;sup&gt;75&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

AD – Autosomal Dominant; AR – Autosomal Recessive; XL – X-Linked

References:

207 Perry Parkway, Gaithersburg, MD 20877 | P: 301-519-2100 | F: 201-421-2010 | E: genedx@genedx.com
49. Watanabe et al. (2018) Nephrolody (Carlton) : (PMID: 29473663)


