Preimplantation Genetic Diagnosis for inherited renal diseases

54th ERA-EDTA Congress
Madrid, Spain
June 3-6, 2017

Nine Knoers
Department of Genetics

University Medical Center Utrecht
Chronic kidney disease frequently has genetic aetiology

Genetic renal disease:

- > 50% of children progressing to renal-replacement therapy
- >10% of adults progressing to renal-replacement therapy

Mutations in > 200 genes associated with inherited kidney diseases
Cumulative numbers of genes discovered for monogenic kidney disease over time
Next Generation Sequencing

From one test per gene to one test for all (involved) genes

- Disease-specific multi-gene panels
- Whole Exome sequencing (all genes)
- Whole Genome sequencing (WGS: complete DNA)
Hereditary nephropathies

Diagnostic yield by systemic screening of all known genes using NGS techniques:

- 15-20% Severe CAKUT
- 30% SRNS
- 60-70% aHUS
- 50-80% Hereditary tubulopathies

Table 1 | Defined rare genetic renal disorders

<table>
<thead>
<tr>
<th>Genetic disorder</th>
<th>Clinical disease entities</th>
<th>Mendelian disease transmission</th>
<th>Genetic disease entities</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Genetic disorders of renal growth and structure</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>CAKUT</td>
<td>14</td>
<td>7 Autosomal dominant</td>
<td>10</td>
</tr>
<tr>
<td></td>
<td></td>
<td>5 Autosomal recessive</td>
<td>12</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1 X-linked</td>
<td>3</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1 Mixed*</td>
<td>1</td>
</tr>
<tr>
<td>Ciliopathies</td>
<td>12</td>
<td>2 Autosomal dominant</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td></td>
<td>8 Autosomal recessive</td>
<td>65</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1 X-linked</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1 Mixed*</td>
<td>1</td>
</tr>
<tr>
<td><strong>Genetic disorders of renal function</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Glomerular diseases</td>
<td>14</td>
<td>7 Autosomal dominant</td>
<td>12</td>
</tr>
<tr>
<td></td>
<td></td>
<td>4 Autosomal recessive</td>
<td>15</td>
</tr>
<tr>
<td></td>
<td></td>
<td>2 X-linked</td>
<td>4</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1 Mixed*</td>
<td>1</td>
</tr>
<tr>
<td>Renal tubular diseases and metabolic diseases</td>
<td>24</td>
<td>4 Autosomal dominant</td>
<td>9</td>
</tr>
<tr>
<td></td>
<td></td>
<td>10 Autosomal recessive</td>
<td>22</td>
</tr>
<tr>
<td></td>
<td></td>
<td>3 X-linked</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td></td>
<td>7 Mixed*</td>
<td>10</td>
</tr>
<tr>
<td>Nephrolithiasis</td>
<td>5</td>
<td>3 Autosomal recessive</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1 X-linked</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td></td>
<td>1 Mixed*</td>
<td>2</td>
</tr>
</tbody>
</table>

CAKUT, congenital anomalies of the kidney and urinary tract. *Mixed mode of transmission, depending on disease and/or syndrome subtype and affected gene.

Increased diagnostic possibilities before and during pregnancy
Pre-pregnancy advice in CKD: do not forget genetic counseling

In 20% of patients with CKD onset before 25 years of age, a monogenic disease can be identified.

Half of these genetic diagnoses constitute revisions of the original clinical primary renal diagnosis (van Eerde et al., 2015)
In 20% of patients with CKD onset before 25 years of age, a monogenic disease can be identified.

Half of these genetic diagnoses constitute revisions of the original clinical primary renal diagnosis.

- Both male and female patients with genetically determined renal disease should be informed about the risks for their offspring and what their options are as prospective parents.

- Nephrologists are advised to arrange pre-pregnancy genetic workup and genetic counseling.

van Eerde et al., KI 2016
Reproductive options for couples confronted with an increased risk of a child with a genetic disease

- Get pregnant together, no tests

- Get pregnant together, **prenatal diagnostic test** potentially followed by termination of pregnancy (TOP)

- Sperm or oocyte donor

- Adoption

- **Preimplantation Genetic Diagnosis (PGD)**
Methods of prenatal diagnosis

**Invasive**
- Chorionic villus sampling
- Amniocentesis
- Cordocentesis

**Non-invasive**
- Ultrasonography
- Maternal serum screen
- Cell free fetal DNA in maternal circulation

Non-Invasive Prenatal Diagnosis (NIPD) / Testing (NIPT)
Non-invasive prenatal diagnosis (NIPD)

Genetic testing of cell-free fetal DNA in maternal circulation

Cell free fetal DNA (cffDNA) shed from the placenta
CffDNA fragments represent the entire fetal genome

Applications:

- Fetal Rhesus D determination
- Fetal sex determination for X-linked disorders
- Aneuploidy testing (screening: NIPT)
- Detection specific fetal mutations in monogenic disorders
NIPD for monogenic disorders

Promising but introduction into clinical practice slow

- Technical challenges (confirm presence of cffDNA and accurately estimate the percentage of cffDNA in sample)
- Costs high

Ethical issues

- Ease of access and safety of procedure may engender feelings of pressure to have testing
- Potential of broadening the scope of testing (whole genome?)
Preimplantation Genetic Diagnosis (PGD)

Introduced in 1989, in UK

Test **before** pregnancy:

- Identifying unaffected embryos
  - ex vivo/*before implantation*

- Preventing genetic disease to be transferred to following generation(s)
  - without termination of pregnancy

**In-vitro fertilisation (IVF)/Intracytoplasmic sperm injection (ICSI) plus genetic testing**
PGD procedure: 3-12 months

- Preparation time
- Hormone treatment, IVE/I
- Embryo biopsy (only possible when mutation(s) known!)
- Genetic testing of 1 cell
- Embryo transfer

Technically difficult
Offered in specialized centres

Day 1
Fertilized egg cell

Day 3
8 cells embryo

Day 3
Biopsy
PGD principle

- Biopsied cell
- Affected
- Affected
- Affected

Transfer only unaffected embryos to the patient

Embryo transfer day 4-5
Wide variation in PGD policy in Europe

**Switzerland & Austria**: only recently legalized for very severe disorders

Very limited in **Germany & Italy**: only for very severe early-onset untreatable disorders

**France**: strictly regulated; each request to use PGD reviewed by Centre Pluridisciplinaire de Diagnostic Prénatal

**UK**: detailed list of diseases for which PGD is permitted; licence committees decide about new indications

**USA**: no regulations; PGD can be used for variety of indications, including sex selection, selection of “saviour siblings”, selection for children with disabilities such as deafness ➔ reproductive tourism?

Bayefsky, Reprod Biomed Soc 2017
PGD in The Netherlands (NL)

- Since 1995
- 4 University Medical Centres: “PGD Nederland”
- 1 license: Maastricht University Medical Centre
- Counseling, IVF/ICSI also in
  - Utrecht
  - Groningen
  - Amsterdam
  - > “Transport PGD”: blastocyst to Maastricht for testing

- PGD for a “new disease” requires approval of:
  - working group Maastricht
  - national indication committee

ADPKD = approved
PGD outcomes general

Success?
- Clinical pregnancy rate 20% per cycle
- Smoking, obesity, increased age significantly decrease chances

Risks?
- For women those of IVF (bleeding, infection, hyperstimulation)
- Misdiagnosis: 2-5% (PND recommended)
- Pregnancies after PGD do not bear a higher risk for congenital anomalies or adverse perinatal outcome.
- PGD-children do not differ in physical nor neuropsychological measurement outcome from IVF/ICSI or NC-children

But....
- Oldest PGD children are now ~25 years of age
- Epigenetic changes may develop later in life or in next generations
<table>
<thead>
<tr>
<th>Expanding list of Nephrogenetic PGD indications in various European centers</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Alport Syndrome</strong></td>
</tr>
<tr>
<td><strong>ADPKD/ARPKD</strong></td>
</tr>
<tr>
<td><strong>Bardet-Biedl syndrome</strong></td>
</tr>
<tr>
<td><strong>Bartter syndrome</strong></td>
</tr>
<tr>
<td><strong>Branchio-Oto-Renal S.</strong></td>
</tr>
<tr>
<td><strong>Cystinosis</strong></td>
</tr>
<tr>
<td><strong>dRTA + hearing loss</strong></td>
</tr>
<tr>
<td><strong>Fabry disease</strong></td>
</tr>
<tr>
<td><strong>Fraser syndrome</strong></td>
</tr>
<tr>
<td><strong>Joubert syndrome</strong></td>
</tr>
<tr>
<td><strong>Tuberous sclerosis</strong></td>
</tr>
</tbody>
</table>
### PGD (NL) for genetic renal disorders up to 2015

<table>
<thead>
<tr>
<th>Disease</th>
<th>Genes</th>
<th>Inheritance</th>
<th>Couples referred (n)</th>
<th>Couples started cycle (n)</th>
<th>Cycles (n)</th>
<th>Ongoing pregnancies (n)</th>
</tr>
</thead>
<tbody>
<tr>
<td>ADPKD</td>
<td>PKD1, PKD2</td>
<td>AD</td>
<td>26</td>
<td>4</td>
<td>6</td>
<td>2</td>
</tr>
<tr>
<td>aHUS</td>
<td>CFH</td>
<td>AD</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Alagille syndrome</td>
<td>JAG1</td>
<td>AD</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Alport disease</td>
<td>COL4A5</td>
<td>XL</td>
<td>17</td>
<td>7</td>
<td>10</td>
<td>5</td>
</tr>
<tr>
<td>ARPKD</td>
<td>PKHD1, FCYT</td>
<td>AR</td>
<td>10</td>
<td>1</td>
<td>6</td>
<td>0</td>
</tr>
<tr>
<td>Bardet Biedl syndrome</td>
<td>BBS7</td>
<td>AR</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>BOR syndrome</td>
<td>EYA1</td>
<td>AD</td>
<td>2</td>
<td>4</td>
<td>5</td>
<td>1</td>
</tr>
<tr>
<td>Congenital nephrotic</td>
<td>NPHS1</td>
<td>AR</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Cystinosis</td>
<td>CTNS</td>
<td>AR</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>FSGS</td>
<td>INF2</td>
<td>AD</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>HNF1B related</td>
<td>HNF1B</td>
<td>AD</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Disease</td>
<td>Genes</td>
<td>Inheritance</td>
<td>Couples referred (n)</td>
<td>Couples started cycle (n)</td>
<td>Cycles (n)</td>
<td>Ongoing pregnancies (n)</td>
</tr>
<tr>
<td>----------------------------------</td>
<td>---------</td>
<td>-------------</td>
<td>---------------------</td>
<td>---------------------------</td>
<td>------------</td>
<td>--------------------------</td>
</tr>
<tr>
<td>Lowe syndrome</td>
<td>OCRL</td>
<td>XL</td>
<td>5</td>
<td>2</td>
<td>8</td>
<td>0</td>
</tr>
<tr>
<td>Nail patella syndrome</td>
<td>LMX1B</td>
<td>AD</td>
<td>3</td>
<td>3</td>
<td>2</td>
<td>1</td>
</tr>
<tr>
<td>Nephrogenic diabetes insipidus</td>
<td>AVPR2</td>
<td>XL</td>
<td>6</td>
<td>4</td>
<td>3</td>
<td>0</td>
</tr>
<tr>
<td>Renal coloboma syndrome</td>
<td>PAX2</td>
<td>AD</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Simpson-Golabi-Behmel syndrome</td>
<td>SGBS1</td>
<td>XL</td>
<td>5</td>
<td>6</td>
<td>9</td>
<td>2</td>
</tr>
<tr>
<td></td>
<td>SGBS2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Tuberous sclerosis complex</td>
<td>TSC1</td>
<td>AD</td>
<td>24</td>
<td>17</td>
<td>22</td>
<td>5</td>
</tr>
<tr>
<td></td>
<td>TSC2</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Von Hippel-Lindau disease</td>
<td>VHL</td>
<td>AD</td>
<td>16</td>
<td>5</td>
<td>8</td>
<td>2</td>
</tr>
<tr>
<td>Wilms tumor</td>
<td>WT</td>
<td>AD</td>
<td>1</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Fabry disease</td>
<td>GLA</td>
<td>XL</td>
<td>2</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Wilson disease</td>
<td>ATP7B</td>
<td>AR</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>Sensenbrenner syndrome</td>
<td>IFT43</td>
<td>AR</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>28 genes</strong></td>
<td></td>
<td><strong>134 couples</strong></td>
<td><strong>58 couples</strong></td>
<td><strong>85</strong></td>
<td><strong>21 ongoing pregnancies</strong></td>
</tr>
</tbody>
</table>

PGD (NL) for genetic renal disorders up to 2015
Case

- 37 yr. old patient
- Hypertension age 25
- Diagnosis ADPKD age 31
- PKD1 mutation c.3866delT
- MDRD eGFR >60
- **Actively requested** information was referred for PGD (2010)
After counseling and nephrological evaluation, the PGD procedure was performed.

- 2nd cycle: pregnancy
- Ultrasound at 20 weeks: no abnormalities
- Follow up uneventful

1st ADPKD PGD pregnancy in NL
Attitudes toward PGD

Attitudes in Patients with Autosomal Dominant Polycystic Kidney Disease Toward Prenatal Diagnosis and Preimplantation Genetic Diagnosis

Oscar Swift, Enric Vilar, Belinda Rahman, Lucy Side, and Daniel P. Gale

FIG. 3. Attitudes to PGD in ADPKD patients. PGD, preimplantation genetic diagnosis.

Swift et al., Genet Test Mol Biomarkers, 2016
PGD for inherited kidney diseases in perspective

- Low numbers of referrals
  - PGD not the preferred option/ethical concerns
  - Initially in couples at risk of disorders with severe neonatal/infantile morbidity

- Approved indications for PGD now also include disorders with adult onset and variable expression
- Growing number of genes identified for nephrogenetic disorders
- Increasing capacity

PGD can now be offered to couples at high risk of having offspring with genetic kidney disease
Implications for nephrologic care

- Consider the **possibility of a genetic disease** in your patients, especially when they are in reproductive age.

- Consider **identifying gene defect** in 1 patient/family, even if there is no direct clinical use for that patient.

- Inform patients **on time**:
  - Discuss options every 2-3 years between 20 and 35+ years.
  - Local clinical geneticist can further inform interested couples.
Patients with genetically determined renal disease should be informed about the risks for their offspring and what their options are as prospective parents; timely genetic work-up and genetic counseling is key!

- PGD is an important alternative to PND. In many European countries PGD for (severe) inherited kidney disorders is possible and can be offered to couples at high risk of having offspring with these genetic kidney diseases after proper genetic counseling.