Genetic Diagnosis of Disorders/Differences of Sex Development (DSD): The DSD-Translational Research Experience

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Disorders of Sex Development (DSD)

Congenital conditions in which development of chromosomal, gonadal or anatomic sex is atypical
DSD: A Paradigm for Management of Complex Conditions

- Affects ~1% of the population
- Congenital
- Treatable but not curable
- Public health significance (fertility, cancer)
- Pervasive challenge to quality of life
- Requires interdisciplinary care
- Chronic conditions
- Transition from pediatric to adult care
Consensus Statement on Management of Intersex Disorders

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“Disorders of Sex Development:
Platform for Basic and Translational Research”

For more information, please contact the DSD-TRN coordinator,
Dr. Emmanuèle Délot: edelot@childrensnational.org (202) 476 6011.
The DSD-TRN:

- 9 new clinical sites joined the original 4 sites since inception

- Seattle Children’s
- OHSU
- UC, San Francisco
- UC, Los Angeles
- Lurie (Chicago, IL)
- UM, Ann Arbor
- Cohen/Long Island (New York)
- National Children’s (Washington D.C.)
- Cincinnati Children’s (OH)
- Le Bonheur, Memphis (TN)
- Mercy Children’s, Oklahoma City
- Washington U. (St Louis, MO)
The Power of Standardized Registries
The **DSD-TRN registry** documents current clinical practice and captures **standardized, quantitative, longitudinal** data on the **genetic** basis of the condition, deep **endocrine** and **anatomic** phenotyping and **psychosocial** adaptation of patients and families.

Web-enabled, searchable, scalable data bank
~2000 unique data points, including genomic variants
Currently 800 patients and family members enrolled.

The **DSD-TRN biobank** centrally collects biological samples from all sites (DNA, gonadal biopsies)
DSD-TRN MODEL OF CARE:

❖ Goal: optimize health and quality of life outcomes by
  ❖ Reducing variability of practice across providers
  ❖ Investigation in areas where no consensus exists
  ❖ Use of gathered evidence to elaborate best practices
  ❖ Involvement of patient stakeholders in all activities

❖ Interdisciplinary team meets with patient to ensure families receives coherent information. Decision-making is shared between all providers and patient family.

❖ Standardization of practice:
  ❖ Commitment to early and comprehensive diagnosis, including genetic
  ❖ Reliable and sensitive description of anatomy
  ❖ Commitment to routine psychosocial screening and to putting into place structure for follow-up, support & education for families.
  ❖ Use of standardized clinical forms for genetic, anatomy, endocrine and psychosocial assessment, customized for DSD diagnosis and management.
Diagnostic effort by DSD-TRN teams increased the percentage of patients with a firm diagnosis from 24% to 46%

<table>
<thead>
<tr>
<th>(TOTAL = 144 forms)</th>
<th>Yes</th>
<th>No</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patient had Dx prior to 1\textsuperscript{st} visit</td>
<td>35</td>
<td>101</td>
</tr>
<tr>
<td>Patient currently has Dx</td>
<td>65</td>
<td>71</td>
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</table>

DSD-TRN registry data
Délot et al. 2017 (Endo Metabolism Clinics)
Genetic diagnostic effort varies greatly by site

<table>
<thead>
<tr>
<th>Site</th>
<th>1</th>
<th>2</th>
<th>3</th>
<th>4</th>
<th>5</th>
<th>6</th>
<th>7</th>
<th>8</th>
<th>9</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dx</td>
<td>15</td>
<td>11</td>
<td>10</td>
<td>13</td>
<td>5</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>9</td>
</tr>
<tr>
<td>no Dx</td>
<td>8</td>
<td>21</td>
<td>5</td>
<td>12</td>
<td>14</td>
<td>1</td>
<td>10</td>
<td>4</td>
<td>5</td>
</tr>
<tr>
<td>% Dx</td>
<td>66%</td>
<td>37%</td>
<td>67%</td>
<td>52%</td>
<td>26%</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
<td>64%</td>
</tr>
<tr>
<td># Reported</td>
<td>23</td>
<td>30</td>
<td>15</td>
<td>25</td>
<td>19</td>
<td>1</td>
<td>10</td>
<td>4</td>
<td>14</td>
</tr>
<tr>
<td># Enrolled</td>
<td>54</td>
<td>31</td>
<td>15</td>
<td>39</td>
<td>19</td>
<td>17</td>
<td>43</td>
<td>72</td>
<td>14</td>
</tr>
<tr>
<td>% Reported</td>
<td>43%</td>
<td>97%</td>
<td>100%</td>
<td>64%</td>
<td>100%</td>
<td>6%</td>
<td>23%</td>
<td>6%</td>
<td>100%</td>
</tr>
<tr>
<td>% with Dx</td>
<td>28%</td>
<td>36%</td>
<td>67%</td>
<td>33%</td>
<td>26%</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
<td>64%</td>
</tr>
</tbody>
</table>

DSD-TRN registry data
Délot et al. 2017 (Endo Metabolism Clinics)
Diagnostic success varies greatly by method

<table>
<thead>
<tr>
<th>Method</th>
<th>Karotype/FISH</th>
<th>Single gene</th>
<th>CMA</th>
<th>Exome</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dx achieved</td>
<td>19</td>
<td>36</td>
<td>6</td>
<td>4</td>
</tr>
<tr>
<td>Test performed</td>
<td>129</td>
<td>47</td>
<td>43</td>
<td>9</td>
</tr>
</tbody>
</table>

Single gene test efficient only for suspicion of CAH

Still very rarely performed
Very high yield: 60% in trios; 25% in singletons

DSD-TRN recommends exome as first-tier Dx tool

*DSD-TRN registry data - Délot et al. 2017 (Endo Metabolism Clinics)*
**46, XY GONADAL DYSGENESIS**

- **15%** mutations in SRY
- **15%** mutations in SF1
- **<1%** rare genetic causes (SOX9, DAX1)
- **70%** no known genetic etiology

**46, XX TESTICULAR DSD**

- **90%** translocations of SRY
- **10%** no known genetic etiology

**46, XX OVOTESTICULAR DSD**

- **10%** translocations of SRY
- **A few case reports of mutations of dup22, RSPO1**
- **90%** no known genetic etiology
The Power of Sequencing
THE COST OF SEQUENCING IS DRAMATICALLY DECREASING AND THE SPEED IS INCREASING.
Human Exome

30 million bases (about 1% of the genome),
~20,000 genes, ~240,000 exons

ONLY CAPTURE THE REGIONS OF THE GENOME OF INTEREST

CLINICAL EXOME SEQUENCING
<table>
<thead>
<tr>
<th>Variant of Unknown Significance</th>
<th>Likely Pathogenic</th>
</tr>
</thead>
<tbody>
<tr>
<td>• To report or not to report?</td>
<td>• Gene known to be involved in sex development</td>
</tr>
<tr>
<td>• Our philosophy: report when clinically relevant and/or actionable</td>
<td>• Mutation previously reported as causative</td>
</tr>
<tr>
<td></td>
<td>• OR likely to be damaging (frameshift, splice-site)</td>
</tr>
<tr>
<td></td>
<td>• Predicted to be damaging by either SIFT, Polyphen, or Condel</td>
</tr>
<tr>
<td></td>
<td>• Minor allele frequency &lt;0.1%</td>
</tr>
<tr>
<td></td>
<td>• Not reported</td>
</tr>
</tbody>
</table>

**Variants from Next Generation Sequencing**
COHORT OF 40 46,XY PATIENTS WITH DSD

Research and Clinical cases (UCLA, Seattle, UM and many other clinics)

Many already had array CGH and all usual suspects sequenced

Wide phenotypic variability

>>>>> EXOME SEQUENCING
RESULTS

Convincing variant found in half (20/40)
Pathogenic variant found in 25%
Likely pathogenic variant found in 10%
Potential Variants of DSD-related Clinical Significance found in 15%

Genetic Diagnosis Rate:
~50% of cases

Baxter et al., 2015, JCEM : 35% to 50%
Eggers et al., 2016, Genome Biology : 43%
PATIENT 1

- 46XY patient presenting at birth with ambiguous genitalia, normal male hormonal profile. Male gender assigned.
- Familial case with at least three other cases in a sibship of 14
- 1 testis, 1 ovotestis with Fallopian tubes

PATIENT 2

- Patient with typical female external genitalia presenting at 15 for primary amenorrhea
- 46, XY with complete gonadal dysgenesis

2 VERY DIFFERENT PRESENTATIONS
EXOME

Both have the same mutation in Sex Determination gene MAP3K1

Same variant previously reported in a family of 5 XY females with complete gonadal dysgenesis

Variant call: Pathogenic

None had ovotestis or ambiguous genitalia:

Expansion of the phenotypic spectrum
2 MORE VARIANTS FOUND IN MAP3K1 IN OUR COHORT
not previously reported

**p.Arg339Gln**
(46XY female with complete gonadal dysgenesis)
de novo
predicted to be damaging (SIFT, PolyPhen, Condel)
not present in population

**p.Pro257Leu**
(46XY, ambiguous genitalia, gonadal dysgenesis, Müllerian remnants)
predicted to be damaging (SIFT, PolyPhen)
not present in Exome Variant Server database

Variant call: Likely Pathogenic
PATIENT 3

- Newborn with penoscrotal hypospadias, chordee, micropenis, descended testes.

**p.Tyr791His in BNC2**
(gene associated with hypospadias)
- Predicted to be damaging
- Not reported previously

AND

**p.Ser107Leu in FGFR1**
(gene associated with HH)
- Predicted to be tolerated
- Not reported previously

Variant call: VUS
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