Prenatal Screening and Diagnostic Testing: Current Status

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Maternal Fetal Medicine and Reproductive Genetics

Disclosures

- Principal Investigator of clinical trial on cfDNA supported by Ariosa Diagnostics
- Research support from Natera
- No personal financial disclosures

Detection rate of prenatal screening for Down syndrome has improved over time
Cell free DNA tests performed through 2014

Traditional Serum Screening

<table>
<thead>
<tr>
<th>Test Type</th>
<th>Time Period</th>
</tr>
</thead>
<tbody>
<tr>
<td>1st trimester biochemistry</td>
<td>10-14 weeks</td>
</tr>
<tr>
<td>2nd trimester biochemistry</td>
<td>15-20 weeks</td>
</tr>
<tr>
<td>Nuchal translucency</td>
<td></td>
</tr>
</tbody>
</table>

Down Syndrome:
93% detection, 4.5% screen positive rate

Title: Pregnancy: Prepare for unexpected prenatal test results.

Title: cfDNA screening for T21: meta-analysis (Gil et al, Ultrasound Obstet Gynecol, 2015)

<table>
<thead>
<tr>
<th>Study</th>
<th>DR (%)</th>
<th>FPR (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>(98.5 - 99.6)</td>
<td>(0.05 - 0.14)</td>
</tr>
</tbody>
</table>

Down Syndrome:
93% detection, 4.5% screen positive rate
cfDNA vs traditional screening

Pros
- Simpler protocol
  ◦ Results more straightforward
  ◦ Not as gestational age dependent
- More accurate
  ◦ Fewer invasive tests
- Potentially earlier results

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Cons
- Need for more complete pre-test consent
  ◦ Advantages of a two-step process
- Fewer invasive tests = less expertise
- 2-4% test failure
- Expensive

What percentage of all chromosome abnormalities will be detected by cfDNA screening?

A. 99%
B. 75%
C. 50%
D. 12%

Disorder | Prevalence  
---|---
Common trisomies (13,18,21) | 0.2%
Other chromosome abnormalities | 0.2%
Microdeletions and duplications | 1.5%
Mendelian Genetic Disorders | 0.4%
Congenital heart defects | 0.8%
Other structural defects | 3%
Adverse OB outcomes | 15-20%
Total | ~25%

Causes of Birth Defects and Other Adverse Perinatal Outcomes: It's Not All Down Syndrome
Aneuploidies Present in LOW RISK Women

- Tri 21: 49.2%
- Sex chromosomal: 9.9%
- Tri 13: 5.5%
- Tri 18: 12.9%
- Other*: 20.8%

*Not detected by cfDNA

Norton et al, SMFM, 2015

Congenital disorders by maternal age

Increasing maternal age ➔

cfDNA is a very precise test for a rare condition

cfDNA is more precise for T13, 18, 21

Increasing maternal age ➔
**cfDNA is more precise for T13, 18, 21**

**cfDNA screening**
- Trisomy 13, 18, 21
- Sex chromosomes
- +/- microdeletions

**Traditional screening**
- Trisomy 18, 21, +/-13
- Other chromosomal
- Early dx fetal anomalies, esp cardiac (NT)
- Spina bifida and ventral wall defects (MSAFP)
- Adverse obstetric outcomes
  - Preeclampsia, preterm birth, fetal growth restriction

**Other abnormalities**
- Trisomy 13, 18, 21
- Sex chromosomes
- +/- microdeletions

**Other aneuploidies**
The performance of cfDNA for other aneuploidies is NOT AS GOOD as for trisomy 21

<table>
<thead>
<tr>
<th>Trisomy</th>
<th>Detection Rate</th>
<th>False Positive Rate</th>
</tr>
</thead>
<tbody>
<tr>
<td>Trisomy 21</td>
<td>99%</td>
<td>0.1%</td>
</tr>
<tr>
<td>Trisomy 18</td>
<td>97%</td>
<td>0.3%</td>
</tr>
<tr>
<td>Trisomy 13</td>
<td>87%</td>
<td>0.6%</td>
</tr>
<tr>
<td>Sex chromosomes</td>
<td>86%</td>
<td>0.6%</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>86%</strong></td>
<td><strong>1.6%</strong></td>
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</table>

-Brief Report- Genetics in Medicine

Discordant noninvasive prenatal testing and cytogenetic results: a study of 109 consecutive cases

Jia-Chi Wang, MD, PhD1, Tribochan Sahoo, MD2,3, Steven Schornberg, PhD, Kimberly A. Kopita, MS1, Leslie Rossi, MS1, Kyla Patek, MS2 and Charles M. Storoni, MD, PhD
Wang et al, *Genetics in Medicine*, 2014

<table>
<thead>
<tr>
<th>Aneuploidy</th>
<th>No. of positives</th>
<th>No (%) confirmed</th>
</tr>
</thead>
<tbody>
<tr>
<td>T21</td>
<td>41</td>
<td>38/41 (93%)</td>
</tr>
<tr>
<td>T18</td>
<td>25</td>
<td>16/25 (64%)</td>
</tr>
<tr>
<td>T13</td>
<td>16</td>
<td>7/16 (44%)</td>
</tr>
<tr>
<td>45X</td>
<td>16</td>
<td>6/16 (38%)</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>98</strong></td>
<td><strong>67 (67%)</strong></td>
</tr>
</tbody>
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Your 25 y.o. patient has cfDNA screening that returns positive for trisomy 13. What is the chance that this is a TRUE positive results?

A. >99%
B. 75%
C. 50%
D. <10%

PPV Calculator: www.perinatalquality.org

NIPT/Cell Free DNA Screening Predictive Value Calculator

www.perinatalquality.org
Cell free DNA limitations

1. Limited number of abnormalities (T13,18,21, sex chromosomes)
   - 75-80% of aneuploidies
   - Missed aneuploidies can be associated with intellectual and other disabilities
   - Important for patients who request screening

2. Test failure
   - 1-8% of tests fail to provide a result
   - This increases the risk for abnormality
Fetal fraction and maternal weight

Failed testing increases aneuploidy risks

**Author** | **OR for aneuploidy**
--- | ---
Norton et al, 2015 | 6.2x
Pergament et al, 2014 | 2.5x
Turocy et al, 2015 | 5.7x

- Primarily T13, 18, triploidy
- These are essentially screen positive results

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If you consider detection of all chromosomal abnormalities, and consider all the failed tests as “screen positive”, what is the performance of traditional screening vs cfDNA screening?

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<tr>
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<th>Cohort Detection Rate</th>
<th>False Positive Rate</th>
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<tbody>
<tr>
<td>Sequential screening</td>
<td>81.6%</td>
<td>4.5%</td>
</tr>
<tr>
<td>cfDNA if “no results” cases = high risk</td>
<td>77.1%</td>
<td>3.7%</td>
</tr>
<tr>
<td>cfDNA if “no results” have no follow up</td>
<td>70.7%</td>
<td>0.7%</td>
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ACOG/SMFM September 2015

- Conventional screening is most appropriate first line screen for most patients
- Ethically any patient may choose cfDNA screening, but should be counseled regarding limitations and benefits
- Diagnostic testing is required to confirm abnormal results before irreversible decisions

- Amniocentesis loss rate now estimated at 0.11% (1/900)

Ultrasound in Obstetrics & Gynecology; 2016
Pregnancy: Prepare for unexpected prenatal test results

Diana W. Bianchi

Women are learning about their own health problems through fetal screening. Revise consent forms and raise awareness, urges Diana W. Bianchi.

Table 2. Association of Maternal Cancers With Different Types of Aneuploidies Detected at Noninvasive Prenatal Testing

<table>
<thead>
<tr>
<th>Type of Aneuploidy Detected by NIPT</th>
<th>Total No. of Samples</th>
<th>No. of Known Maternal Cancers (%) [95% CI]</th>
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<tr>
<td>Single trisomy*</td>
<td>2650</td>
<td>2 (0.08) [0-0.27]</td>
</tr>
<tr>
<td>Single SCAb</td>
<td>950</td>
<td>0 (0) [0-0.39]</td>
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<tr>
<td>Single trisomy + SCA</td>
<td>30</td>
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The Public Health Evidence for FDA Oversight of Laboratory Developed Tests: 20 Case Studies

Office of Public Health Strategy and Analysis
Office of the Commissioner
Food and Drug Administration
November 16, 2015

"...these products may have caused or have caused actual harm to patients."

Office of Public Health Strategy and Analysis
Office of the Commissioner
Food and Drug Administration
November 16, 2015
Should all women be offered cfDNA screening?

C. Tests with the Potential to Yield Both Many False-Positive and False-Negative Results

i. Noninvasive Prenatal Testing (A.K.A. cell-free DNA testing)

<table>
<thead>
<tr>
<th>Category</th>
<th>LDT Characteristics</th>
</tr>
</thead>
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<td>LDT Name</td>
<td>Noninvasive prenatal cell-free DNA testing (NIPT, or cfDNA)</td>
</tr>
<tr>
<td>Description</td>
<td>Blood test to identify traces of fetal chromosomes in maternal blood</td>
</tr>
<tr>
<td>Purpose</td>
<td>To detect a range of fetal chromosomal abnormalities</td>
</tr>
<tr>
<td>Target Population</td>
<td>Pregnant women concerned about a fetal chromosomal abnormality</td>
</tr>
<tr>
<td>Alternatives</td>
<td>Invasive testing, including amniocentesis and chorionic villi sampling; “quad testing” of multiple substances combined with ultrasound imaging</td>
</tr>
<tr>
<td>LDT Problem 1</td>
<td>Lack of clinical validation that tests detect and predict fetal abnormalities at an appropriate rate</td>
</tr>
<tr>
<td>LDT Problem 2</td>
<td>Many false-positive results when used in the general population</td>
</tr>
<tr>
<td>Clinical Consequence</td>
<td>Women with false positive results may abort a normal pregnancy, women with false-negative results may deliver a child with an unanticipated genetic syndrome</td>
</tr>
<tr>
<td>Potential Impact of FDA Oversight</td>
<td>Assurance the test meets minimum performance standards; evaluation of manufacturer claims</td>
</tr>
<tr>
<td>Cost Impact of Inaccuracy</td>
<td>Not estimated</td>
</tr>
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Should all women be offered cfDNA screening?
Should all women be offered cfDNA screening?

Issues with cfDNA screening mean the clinical utility is lower than carefully curated data would lead you to believe.

Risk Profiler and Test Detection Tool

gem.perinatalquality.org
What is a provider to do?

- Follow ACOG/SMFM guidelines
- Use PPV calculator for any test positive cases
- Explain to patients that cfDNA is NOT a “noninvasive amnio”
- Counsel patients with failed tests
- Know the performance characteristics of the lab that you use
  - Test failure rate
  - Incidental findings reporting

Thank you!!