14.2% (FFA) screening for genetic and pre-term delivery

14.2% (FFA) supervision of normal infant death

True Positive

AMA, high risk pregnancy

True Positive

23.3% (FFA)

27.2% (FFA)

5.9% (FFA)

AMA, abnormal maternal serum screen

False Positive

31.7% (FFA)

Pyelectasis on U/S

AMA

True Positive

28w3d

36.4%

Abnormal U/S

CHD detected on U/S

TOF identified by U/S

10w0d

Term delivery

10w5d

Abnormal U/S

TOF noted. Infant died at 29h.

VSD and other heart defects on U/S

11.8% (FFA)

Term delivery;

preeclampsia

Craniosynostosis noted on U/S

6

22.3% (FFA)

AMA, high risk pregnancy

TAP

Multiple heart defects on U/S

7

20.7%

Abnormal U/S

Pre-term delivery

Complex fetal heart defects, IUGR and other abnormalities on U/S. Infant died at 5mo.

8

14.2% (FFA)

CHD detected on U/S

Term delivery

TOF, IUGR on U/S

9

27.3% (FFA)

Hx of previous infant with congenital anomalies

Term delivery

ASD and other heart defects noted on postnatal echo

10

5.9% (FFA)

Screening for genetic and chromosomal anomalies

Infant death

TOF noted. Infant died at 29h.

11

14.2% (FFA)

Abnormal U/S, bilateral UTD, VSD

Term delivery

VSD and other heart defects on U/S

12

36.4% (FFA)

Abnormal U/S, chromosome abnormality suspected in fetus

Pre-term delivery

Multiple CHD, Polyhydramnios, IUGR, multiple congenital anomalies noted on U/S

13

10.4%

Supervision of normal first pregnancy

Fetal demise: 18w5d

Malformed male fetus, immature placenta with ONTD

Introduction

• Prenatal detection of microdeletion syndromes enables pregnancy management and early intervention in affected newborns.
• 22q11.2 deletion syndrome—also known as DiGeorge Syndrome—is the most frequent chromosomal microdeletion disorder, estimated to occur in approximately 1 in 2,500-4,000 births.
• Unlike multiple marker screening, noninvasive prenatal screening (NIPS) technology can detect fetuses affected by microdeletions.
• By increasing fetal fraction (FF) by more than two-fold on average, fetal fraction amplification (FFA) may further enhance NIPS detection of microdeletions.
• Limited studies on the clinical performance of NIPS for 22q11.2 deletion detection report positive predictive values (PPV) between 20%-50%. However, no reports have included clinical performance using FFA.
• Here, we sought to describe the performance of 22q11.2 deletion screening using a whole-genome sequencing (WGS)-based NIPS platform that leverages FFA.

Results

• 13 cases that screened positive for 22q11.2 deletion had completed molecular diagnostic testing.
• FFA was a standard part of the NIPS assay at the time that eleven of these thirteen cases were processed.
• Among these 13 screen-positive cases, twelve were confirmed as true positives, and one false positive.
• The resulting PPV was 92.3% (95 percent CI PPV=64.0% to 99.8%). Among true positives, gestational age at time of NIPS testing ranged from 10 weeks 0 days to 35 weeks 0 days.
• The mean fetal fraction of true positives was 21.15% (range 5.9% to 36.4%), near the mean FF level of all samples tested.
• Among these 13 screen-positive cases, twelve were confirmed as true positives, and one false positive.
• The resulting PPV was 92.3% (95 percent CI PPV=64.0% to 99.8%). Among true positives, gestational age at time of NIPS testing ranged from 10 weeks 0 days to 35 weeks 0 days.
• The mean fetal fraction of true positives was 21.15% (range 5.9% to 36.4%), near the mean FF level of all samples tested.
• For the false positive, the gestational age at time of NIPS testing was 10 weeks 5 days, and the fetal fraction was 10.4% assessed prior to FFA enhancements on the assay for the single false positive.

Figure 1. 22q Deletion Screen Positive Cases with Pregnancy Outcome Data

Table 1. Outcomes of Screen Positives with Molecular Confirmation

<table>
<thead>
<tr>
<th>Case #</th>
<th>Result</th>
<th>GA at Draw</th>
<th>Fetal Fraction</th>
<th>Indication for Screening</th>
<th>Pregnancy Outcome</th>
<th>Clinical Notes</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>True Positive</td>
<td>28w3d</td>
<td>31.7% (FFA)</td>
<td>Abnormal U/S</td>
<td>Term delivery</td>
<td>TOF identified on U/S</td>
</tr>
<tr>
<td>2</td>
<td>True Positive</td>
<td>22w2d</td>
<td>26.2% (FFA)</td>
<td>High Risk Pregnancy</td>
<td>Term delivery</td>
<td>VSD and other heart defects on U/S</td>
</tr>
<tr>
<td>3</td>
<td>True Positive</td>
<td>12w0d</td>
<td>27.2% (FFA)</td>
<td>AMA, Abnormal maternal serum screen</td>
<td>Term delivery</td>
<td>Pylectasis on U/S</td>
</tr>
<tr>
<td>4</td>
<td>True Positive</td>
<td>26w3d</td>
<td>30.0% (FFA)</td>
<td>Abnormal U/S</td>
<td>Pre-term delivery</td>
<td>TOF and other heart defects on U/S</td>
</tr>
<tr>
<td>5</td>
<td>True Positive</td>
<td>19w3d</td>
<td>11.8% (FFA)</td>
<td>AMA</td>
<td>Term delivery, preeclampsia</td>
<td>Craniosynostosis noted on U/S</td>
</tr>
<tr>
<td>6</td>
<td>True Positive</td>
<td>10w0d</td>
<td>23.3% (FFA)</td>
<td>AMA, High Risk pregnancy</td>
<td>TAB</td>
<td>Multiple heart defects on U/S</td>
</tr>
<tr>
<td>7</td>
<td>True Positive</td>
<td>35w0d</td>
<td>20.7%</td>
<td>Abnormal U/S</td>
<td>Pre-term delivery</td>
<td>Complex fetal heart defects, IUGR and other abnormalities on U/S. Infant died at 5mo.</td>
</tr>
<tr>
<td>8</td>
<td>True Positive</td>
<td>23w3d</td>
<td>14.2% (FFA)</td>
<td>CHD detected on U/S</td>
<td>Term delivery</td>
<td>TOF, IUGR on U/S</td>
</tr>
<tr>
<td>9</td>
<td>True Positive</td>
<td>18w5d</td>
<td>27.3% (FFA)</td>
<td>Hx of previous infant with congenital anomalies</td>
<td>Term delivery</td>
<td>ASD and other heart defects noted on postnatal echo</td>
</tr>
<tr>
<td>10</td>
<td>True Positive</td>
<td>11w6d</td>
<td>5.9% (FFA)</td>
<td>Screening for genetic and chromosomal anomalies</td>
<td>Infant death</td>
<td>TOF noted. Infant died at 29h.</td>
</tr>
<tr>
<td>11</td>
<td>True Positive</td>
<td>24w6d</td>
<td>14.2% (FFA)</td>
<td>Abnormal U/S, bilateral UTD, VSD</td>
<td>Term delivery</td>
<td>VSD and other heart defects on U/S</td>
</tr>
<tr>
<td>12</td>
<td>True Positive</td>
<td>34w4d</td>
<td>36.4% (FFA)</td>
<td>Abnormal U/S, chromosome abnormality suspected in fetus</td>
<td>Pre-term delivery</td>
<td>Multiple CHD, Polyhydramnios, IUGR, multiple congenital anomalies noted on U/S</td>
</tr>
<tr>
<td>13</td>
<td>False Positive</td>
<td>10w5d</td>
<td>10.4%</td>
<td>Supervision of normal first pregnancy</td>
<td>Fetal demise: 18w5d</td>
<td>Malformed male fetus, immature placenta with ONTD</td>
</tr>
</tbody>
</table>

Conclusions

• Early analysis of outcomes for 22q11.2 deletion performance using a WGS-based NIPS with FFA suggests a PPV substantially higher than previous studies have reported.
• This improvement in PPV likely arises from higher FF levels from FFA, which facilitates detection of small deletions in cell-free fetal DNA.
• These initial results suggest that 22q11.2 screening performance with FFA in WGS-based NIPS is consistent with that of other reportable anomalies recommended by guidelines (e.g., for trisomy 13), warranting possible inclusion of 22q11.2 deletion screening in routine NIPS.
• Outcomes collection for additional cases is ongoing, which is expected to improve the certainty of these results.