Accurate Gender Determination Using a Single-Nucleotide Polymorphism-based Non-invasive Prenatal Test

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Abstract

Background: Accurate gender determination is required in the management of certain X-linked diseases, including medical fetal therapy. In addition, discrepant ultrasound and NIPT gender results may result in unnecessary invasive testing and further unnecessary follow up. Objective: To determine the ability of single nucleotide polymorphism (SNP)-based non-invasive prenatal testing (NIPT) to determine fetal gender.

Methods: Gender was determined as part of a validation study of a SNP-based NIPT method for fetal whole-chromosomal aneuploidy. Whole-free DNA was isolated from maternal blood samples, amplified using a 19,488-plex PCR approach, and sequenced. Sequencing results were analyzed using the NATUS algorithm to determine the fetal chromosomal copy number. Male was defined as presence of the Y chromosome and female defined as lack of the Y chromosome. The SNP-based NIPT-determined gender was compared to karyotype results obtained from invasive procedures or at birth.

Results: Gender determination was made on 1,005 samples; Y chromosome was detected in 534 cases, no Y chromosome was detected in 471 cases. All gender determinations (100%; 1,005/1,005) made by NIPT matched karyotype. The cohort contained 12 Monosomy X cases correctly identified as female, including one false negative 45,X case called as 46,XX, and one false positive 46,XX case called 45,X.

Conclusions: This SNP-based NIPT can determine gender with high accuracy. The ability to determine fetal gender early in pregnancy can alter clinical management, particularly in cases of sex-linked disorders. Furthermore, reducing discrepancy between NIPT and ultrasound will result in less unwanted invasive procedures and unnecessary follow up testing.

Methods

Visualization of SNP Data Used in Fetal Sex Determination

Figure 2: Y-Chromosome SNP data for female and male fetuses. Y-axis indicates the magnitude of measured Y chromosome SNPs. A male fetus is characterized by the presence of a cluster of black dots (SNPs) on the Y chromosome. The absence of Y chromosome dots indicates a female fetus. Representative SNP plots shown at 5% (A), 10% (B), and 20% (C) fetal fractions; the exclusive presence of Y-chromosome SNPs in male fetuses is evident across a range of fetal fractions. For male fetuses, as fetal fraction increases, the spread of the Y-chromosome dots increases and they migrate further up from the X-axis. Note that this is not how the algorithm makes ploidy calls, but is one method for visualizing the data.

Validation Studies: 100% Accuracy for Fetal Sex

Table 2: Comparison of NIPT fetal sex results with fetal karyotype.

<table>
<thead>
<tr>
<th>NIPT Call</th>
<th>Male</th>
<th>Female</th>
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<tbody>
<tr>
<td>Male</td>
<td>46,XY; 47,XX +21/+18/or +13</td>
<td>534</td>
</tr>
<tr>
<td>Female</td>
<td>45,X; 46,XX; 47,XX +21/+18/or +13</td>
<td>0</td>
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Results

Conclusions

This SNP-based NIPT accurately determined fetal sex, as defined by the presence or absence of the Y chromosome. Accurate sex determination is required in the management of X-linked disorders. Incorrect fetal sex determination can lead to incorrect identification of sex chromosome abnormalities and gender, resulting in unnecessary diagnostic follow-up testing and potential diagnostic odyssey.

References

6. Natera internal data.