Non-Invasive Prenatal Screening

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CLINICAL APPLICATION
Non-Invasive prenatal screening relies on a single maternal blood draw. It is the newest methodology in the detection of fetal aneuploidies for Chromosomes 21, 18 and 13 and sex chromosomes. It offers a safer screening option prior to an invasive diagnostic procedure of amniocentesis and CVS. The test can be performed in the first trimester of pregnancy at a minimum of 10 weeks gestational age. The test sequences cell free DNA from the mother’s blood using massively parallel sequencing technology (MPS) on a next generation sequencing platform. This offers remarkable sensitivity for prenatal screening.

Current American College of Obstetrics and Gynecology (ACOG) recommendations are that testing be offered to women under the following conditions:
- Advanced maternal age
- Positive results of prenatal aneuploidy screening
- Presence of ultrasound abnormalities
- Previous affected pregnancy for fetal aneuploidy

CLINICAL BACKGROUND
Trisomy 21 or Down syndrome is caused by a complete or partial third copy of chromosome 21 in an individual’s cells. It is estimated to occur in 1 in 691 live births. T21 is associated with characteristic features and health conditions that will require management. These conditions include reduced intellectual capacity and congenital heart defects in up to 50% of patients.

Trisomy 18 or Edwards syndrome is caused by the presence of a third copy of chromosome 18. It is the most common autosomal trisomy after T21. It is estimated to occur in 1 in 6,000 live births. Approximately 90% of live births do not survive the first year of life.

Trisomy 13 or Patau syndrome is caused by a complete or partial extra copy of chromosome 13. It is the third highest live birth autosomal trisomy. It affects approximately 1 in 10,000 live births. Greater than 80% of live births die within the first year of life.

Monosomy X (45,X) or Turner syndrome occurs between 1:2000 to 1:5000 live births. It is believed that 99% of prenatal cases succumb by spontaneous abortion. Monosomy X is characterized by a prevalence of cardiovascular abnormalities. In addition, individuals are characterized with short stature, kidney abnormalities, thyroid disorders, risk for diabetes, infertility and cognitive learning difficulties can be present.

XXX (47,XXX) is characterized by the presence of an extra X chromosome in females. It is thought to occur in 1:1000 live female births. Because of lyonization, symptoms are generally mild to nonexistent, but can include tall stature, learning disabilities, dyslexia, microcephaly, delayed motor skills and weak muscle tone.

XXY (47,XXY) or Klinefelter syndrome is a genetic disorder in which male cells contain an extra X chromosome copy. It is present in 1:500 to 1:1000 live male births. Males can have less muscle mass and weaker bones when younger. Gynecomastia and Microorchidism may be present and individuals tend to be infertile.

XYY (47,XYY) or Jacob’s syndrome is the presence of an extra Y chromosome in male cells. It occurs at a rate of approximately 1:1000 live male births. The clinical phenotype is normal.

LAB TEST CONNECT

Quick Facts
- Recommended if patient is of advanced maternal age, has an abnormal prenatal screen, an abnormal ultrasound or a previous aneuploidy pregnancy
- Screens for trisomy 21, 18 and 13
- Optional sex chromosome analysis for aneuploidy and risk stratification
- Optional trisomy 9 and 16 with microdeletions
- Test failure rate 0.1% - lowest in the industry
- Egg donors and IVF are acceptable for testing

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Non-Invasive Prenatal Screening

TEST INFORMATION

Non-Invasive Prenatal Screening

DESCRIPTION

The PAML NIPT test offering has three options...

1. **BASIC VERIFI® BY PAML TEST (PNA1):**
   - Detects
     - T21 (Down syndrome)
     - T18 (Edwards syndrome)
     - T13 (Patau syndrome)

2. **BASIC TEST WITH SEX CHROMOSOME ANEUPLOIDY ANALYSIS (PNA2) – NO EXTRA CHARGE:**
   - Monosomy X (MX, Turner syndrome)
   - XXX (Trisomy X)
   - XXY (Klinefelter syndrome)
   - XYY (Jacob’s syndrome)
   - Fetal sex (XX or XY) – aids in stratifying the risk for X-linked disorders such as hemophilia, Duchenne muscular dystrophy or cases of ambiguous genitalia, such as congenital adrenal hyperplasia

3. **EXPANDED ANEUPLOIDY WITH SEX CHROMOSOME (PNA3):**
   - The basic test with sex chromosome plus Trisomies 9 and 16 with microdeletions.
   - Limited to singleton pregnancies only.

Test Performance*

<table>
<thead>
<tr>
<th>BASIC VERIFI TEST</th>
<th>CHR</th>
<th>N</th>
<th>OBSERVED SENSITIVITY</th>
<th>95%CI</th>
<th>OBSERVED SPECIFICITY</th>
<th>95%CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>21</td>
<td>577</td>
<td>99.14%</td>
<td>98.0-99.7</td>
<td>99.94%</td>
<td>99.90-99.97</td>
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</tr>
<tr>
<td>18</td>
<td>175</td>
<td>98.31%</td>
<td>95.0-99.6</td>
<td>99.9%</td>
<td>99.86-99.93</td>
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</tr>
<tr>
<td>13</td>
<td>53</td>
<td>98.15%</td>
<td>90.0-99.9</td>
<td>99.95%</td>
<td>99.91-99.97</td>
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</table>

<table>
<thead>
<tr>
<th>SEX CHROMOSOME ANEUPLOIDY ANALYSIS</th>
<th>CHR</th>
<th>N</th>
<th>OBSERVED SENSITIVITY</th>
<th>95%CI</th>
<th>OBSERVED SPECIFICITY</th>
<th>95%CI</th>
<th>ACCURACY</th>
<th>95%CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>MX</td>
<td>508</td>
<td>95.0%</td>
<td>75.1-99.9</td>
<td>99.0%</td>
<td>97.6-99.7</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>XX</td>
<td>508</td>
<td>97.6%</td>
<td>94.8-99.1</td>
<td>99.2%</td>
<td>97.2-99.9</td>
<td>98.4%</td>
<td>96.9-99.3</td>
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</tr>
<tr>
<td>XY</td>
<td>508</td>
<td>99.1%</td>
<td>96.9-99.9</td>
<td>98.3%</td>
<td>96.9-99.8</td>
<td>99.0%</td>
<td>97.7-99.7</td>
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**SEX CHROMOSOME ANEUPLOIDY ANALYSIS**

<table>
<thead>
<tr>
<th>XXX</th>
<th>XXX</th>
<th>XY</th>
<th>XXXY</th>
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</thead>
<tbody>
<tr>
<td>Limited data on these rare aneuploidies preclude performance calculations</td>
<td></td>
<td></td>
<td></td>
</tr>
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</table>

*Test performance data on file with Verinata Health, Inc., a wholly owned subsidiary of Illumina, Inc.*

METHOD

Cell-free DNA is isolated from a 7 ml blood draw from the mother. DNA is enriched and subjected to massive parallel sequencing analysis. Chromosomes of interest are compared to optimized reference chromosomes within the DNA sample. This minimizes sample-to-sample and run-to-run variation.

ORDER CODE

<table>
<thead>
<tr>
<th>PNA1</th>
<th>Prenatal Aneuploidy without sex chromosome</th>
</tr>
</thead>
<tbody>
<tr>
<td>PNA2</td>
<td>Prenatal Aneuploidy with Sex Chromosome</td>
</tr>
<tr>
<td>PNA3</td>
<td>Expanded Aneuploidy with Sex Chromosome</td>
</tr>
</tbody>
</table>

CPT CODE

| 81420 |

SPECIMEN REQUIREMENTS

Whole Blood (7 ml minimum, 10 ml preferred)
Room temp 5 days

RANGES

No aneuploidy detected

REFERENCES

3. For test metrics from the MELISSA validation study, please see Bianchi DW, Platt LD, Goldberg JD, et al. Genome-wide fetal aneuploidy detection by maternal plasma DNA sequencing. Obstet Gynecol. 2012;119:890-901. In accordance with medical societies’ requests, the observed metrics shown are provided to reflect more recent clinical experience.

DISCLAIMER

The verifi test was developed by, and its performance characteristics were determined by Verinata Health, Inc., a wholly owned subsidiary of Illumina, Inc. The VHI laboratory is CAP-accredited and certified under the Clinical Laboratory Improvement Amendments (CLIA) as qualified to perform high complexity clinical laboratory testing. It has not been cleared or approved by the U.S. Food and Drug Administration.

For more information, please contact your local sales representative.