The Most Trusted Name in Genetics
Current limitations w/status quo

GENETIC TESTING IS NOT ACCESSIBLE TO EVERYONE DUE TO:

- Insurance contracts
- Slow turnaround
- Limited office and counseling resources
- High costs
Solutions:

- In-Network/Affordable
- Quick results
- Highest standard of care
- Easy on/off-line integration
Rethinking the clinical lab
What BMGL has to offer:

- In-Network w/nearly ALL payors including Medicaid
- Human Genome Sequencing Center
  - Largest genome center in the US
- Baylor’s years of variant curation
- Expert analysis from Baylor faculty members
- Test development input from clinical specialists
- Broad testing menu across multiple specialties

BMGL, Your One Stop for Family Planning
In-Network w/nearly ALL Payors
Human Genome Sequencing Center

- Established in 1996
- The Center was chosen in 1999 as one of three sites to complete the Human Genome Project
- In June 2000, scientists deciphered the human genome, the blueprint for human life
- In April 2003, the HGP produced a complete, high-quality human DNA reference
- Responsible for determining the DNA sequence of chromosomes 3, 12, and part of X.
Baylor’s expertise and experience in the field:

<table>
<thead>
<tr>
<th>Thought Leaders</th>
<th>Baylor’s expertise and experience in the field:</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dr. Arthur Beaudet</td>
<td>Hundreds of thousands of Molecular Tests</td>
</tr>
<tr>
<td>Dr. Jim Lupski</td>
<td>10+ HiSeq Units</td>
</tr>
<tr>
<td>Dr. Richard Gibbs</td>
<td>3 Ion Torrents</td>
</tr>
<tr>
<td>Dr. Sharon Plon</td>
<td>3 MiSeqs</td>
</tr>
<tr>
<td>Dr. Brendan Lee</td>
<td>Illumina X10</td>
</tr>
<tr>
<td>Dr. Christine Eng</td>
<td></td>
</tr>
<tr>
<td>Dr. Lee-Jun Wong</td>
<td></td>
</tr>
</tbody>
</table>
“Although Mendelian diseases are individually rare, they are quite common when viewed as a group, and their burden on patients is great.”

1 in every 100 children is born with a genetic disorder*

*World Health Organization
Dietz HC, New Therapeutic Approaches to Mendelian Disorders. NEJM 2010;363(9):852-862
Reproductive Carrier Screening
Knowledge is power. Plan ahead with GeneAware.

Learn more
Expanded carrier screen
• Screens for 132 genes
• Patients can be screened before conception or during pregnancy

GeneAware

Panel Listing
Four GeneAware panels are offered for your convenience. Females are screened for X-linked Duchenne and Becker muscular dystrophies and Fragile-X syndrome in all four panel options.

Download Gene Aware Panel Listing
Depth and Breath

› Alpha-Thalassemia

› DMD – Duchenne/Becker Muscular Dystrophy

› SCID – Severe Combined Immunodeficiency

25% Positive Rate

<1% Carrier Couple Rate
Turnaround time at or below industry standard

![Bar chart showing turnaround time (TAT) in days for Q3'14, Q4'14, and Q1'15. The chart indicates a decrease in TAT from Q3'14 to Q1'15, with Q1'15 at 9.7 days.]
Pre-test education
# Reporting

**Patient Name:** Jane Doe  
**Date of Birth:** 1/6/1982  
**Gender:** F  
**Lab Number:** 128456  
**Barcode #:** A12345678  
**Ethnicity:** African American  
**Indication for Study:** HISTORY OF TWO STILLBIRTHS  
**Provider Name:**  
**Test Code:** 60100

### GeneAware Complete Reproductive Carrier Screening  
**CARRIER**

### Results Summary

<table>
<thead>
<tr>
<th>Disease</th>
<th>Gene</th>
<th>Inheritance Pattern</th>
<th>Variant</th>
<th>Patient Status</th>
<th>Partner Status</th>
<th>Reproductive Risk</th>
</tr>
</thead>
<tbody>
<tr>
<td>Glucose-6-Phosphate Dehydrogenase Deficiency</td>
<td>G6PD</td>
<td>X-linked</td>
<td>c.202G&gt;A (p.V68M)</td>
<td>Carrier</td>
<td>Not Submitted</td>
<td>1 in 4</td>
</tr>
<tr>
<td>Glucose-6-Phosphate Dehydrogenase Deficiency</td>
<td>G6PD</td>
<td>X-linked</td>
<td>c.376A&gt;G (p.N126D)</td>
<td>Carrier</td>
<td>Not Submitted</td>
<td>1 in 4</td>
</tr>
<tr>
<td>Beta Hemoglobinopathies (Beta-thalassemia and Sickle Cell)</td>
<td>HBB</td>
<td>Autosomal recessive</td>
<td>c.20A&gt;T (p.E7V; Sickle Cell S allele)</td>
<td>Carrier</td>
<td>Not Submitted</td>
<td>1 in 40</td>
</tr>
</tbody>
</table>

### Interpretation

**Glucose-6-Phosphate Dehydrogenase Deficiency:**  
This individual carries one copy of p.V68M and one copy of p.N126D. While phase has not been established in this individual, p.V68M is nearly always in cis to p.N126D, which is designated as p.[V68M; N126D]. The p.[V68M; N126D] allele is also known as G6PD (A-), and is associated with reduced enzyme activity (10-60% activity, PMID: 17036616). Most individuals with the p.[V68M; N126D] allele are asymptomatic, but mild to moderate hemolysis may occur and adverse consequences are associated with administration of primaquine. This individual has a 1 in 2 chance for transmitting the p.[V68M; N126D] allele. There is a ¼ chance for a pregnancy to produce a son with the p.[V68M; N126D] genotype. Assuming a healthy, untested African American partner, there is also a 1 in 32 chance of producing a daughter who is homozygous for the p.[V68M; N126D] allele.

**Sickle Cell Disease:**  
This individual is a heterozygous carrier of the sickle cell trait. Please see the Sickle Cell counseling section of this report for an explanation of the significance of this finding.

### Comments and Recommendations:
Results delivery

SIGN IN

Please enter your username and password. Don't have an account? Click here to Register.
You may also use your GeneAware username and password to access the portal

Account Information

Username: 
Password*: 

*Password is case sensitive. Forgot password?

Sign In
Genetic counseling

After discussing your test results with your health care provider, a complimentary consult with a certified Genetic Counselor is available by phone. You can schedule a phone appointment by contacting Client Service at 713-798-6555. If you would prefer to see a local genetic counselor in person, we can help assist in locating the nearest Genetic Counselor.
Average patient cost

Average patient out of pocket by quarter

<table>
<thead>
<tr>
<th>Quarter</th>
<th>2014</th>
</tr>
</thead>
<tbody>
<tr>
<td>Q1</td>
<td>$90.00</td>
</tr>
<tr>
<td>Q2</td>
<td>$90.00</td>
</tr>
<tr>
<td>Q3</td>
<td>$60.00</td>
</tr>
<tr>
<td>Q4</td>
<td>$30.00</td>
</tr>
</tbody>
</table>
Why expanded carrier screening?

An empirical estimate of carrier frequencies for 400+ causal Mendelian variants: results from an ethnically diverse clinical sample of 23,453 individuals

Gabriel A. Lazarin, MS1, Imran S. Haque, PhD1, Shivani Nazareth, MS1, Kevin Ioni, BS2, A. Scott Patterson, MA1, Jessica J. Jacobson, MD1, John R. Marshall, MD1, William K. Seltzer, PhD, FACMG2, Pasquale Patrizio, MD3, Eric A. Evans, PhD3 and Balaji S. Srinivasan, PhD1,4

Original Research Article  Genetics in Medicine  Open

Purpose: Recent developments in genetics have led to expanded carrier screening panels capable of assessing hundreds of causal mutations for genetic diseases. This new technology enables simultaneous measurement of carrier frequencies for many diseases. As the resultant test ordering of carrier frequencies impacts the design and prioritization of screening programs, the accuracy of this testing is a public health concern.

Methods: A total of 24,816 individuals from many ethnicities, genetics, and infertility clinics were enrolled for routine recessive disease carrier screening. Multiple carrier screening was performed and results were aggregated for this study.

Results: Twenty-four percent of individuals were identified as carriers for at least one of 108 disorders, and 3.2% were carriers for multiple disorders. We report estimates of carrier frequency by self-defined ethnicity and disease.

Conclusions: To our knowledge, this study of a large, ethnically diverse clinical sample provides the most accurate measurements to date of carrier frequencies for hundreds of recessive disorders. The study also shows that there are cases of individuals with expanded panels and provides support for a panel that prioritizes variants in “higher risk” categories by the physician, as recommended by national guidelines.

Genet Med advance online publication 13 September 2012

Key Words: carrier frequency, carrier screening, genetic testing, panel, ethnic, recessive disease

77%
Of carriers Counsyl identified would have been missed by ACOG guidelines

69%
Of carriers Counsyl identified would have been missed by ACMG guidelines

100%
Of the disorders on our panel are actionable

—Lazarin, et al., Genetics in Medicine, 2012
Candidates for carrier screening

The ideal population is individuals and couples considering having children in the near future.
45% of individuals that do not report Jewish ancestry are carriers of diseases related to Jewish ancestry.

*Other carriers detected reported African-American, Asian, Hispanic and Native American ancestry.

* Source: Counsyl, internal data, 2009 - 2011.
Are these diseases rare?

#1 cause of infant deaths each year

20% of all infant deaths

12% of pediatric hospitalizations were due to birth defects and/or genetic disease

Total healthcare cost: $2.5-5.0 billion
Disease incidence

1/800
Neural Tube Defect

1/700
Down Syndrome

1/400
Conditions on GeneAware
2013 ACMG guidelines

ESTABLISHES:

- Requirements for informed consent
- Disease inclusion criteria

Now it is possible, using new technologies, to screen for mutations in many genes for approximately the same cost as previously required to detect mutations in a single gene or a relatively small number of population-specific mutations in several genes.
Ethnicity based on screening

- 1 in 7 marriages are between spouses of different ethnicities
- Multiracial children have increased 50% over the last decade
- Publications support the pitfalls of relying on self-reported ethnicity
OB/GYN adoption of expanded carrier screening

~67% of respondents support Universal ECS

~52% provide ECS at patient’s request

~80% consider preconception screening to be optimal

Only 15% offer ECS to all patients


**Benn P et al. Obstetricians’ and gynecologists’ practice and opinions of expanded carrier testing and non-invasive prenatal testing. Prenatal Diagnosis (2013)
# Higher carrier frequencies

<table>
<thead>
<tr>
<th>Disease</th>
<th>Rate</th>
<th>Ethnicity</th>
</tr>
</thead>
<tbody>
<tr>
<td>Alpha Thalassemia</td>
<td>1/16</td>
<td>East Asians</td>
</tr>
<tr>
<td>Hermansky Pudlak Syndrome</td>
<td>1/21</td>
<td>Puerto Ricans</td>
</tr>
<tr>
<td>Duchene Muscular Dystrophy</td>
<td>1/3,764</td>
<td>Pan-Ethnic</td>
</tr>
<tr>
<td>MECP-2 Duplication Syndrome</td>
<td>Unknown 1% prevalence in males w/intellectual disability/MR</td>
<td>Pan-Ethnic</td>
</tr>
<tr>
<td>POLG-Related Disorders</td>
<td>1/50</td>
<td>Pan-Ethnic</td>
</tr>
<tr>
<td>Sandhoff Disease</td>
<td>1/279</td>
<td>Pan-Ethnic</td>
</tr>
</tbody>
</table>
### GeneAware Carriers Found

<table>
<thead>
<tr>
<th>ACOG</th>
<th>ACMG</th>
<th>Not ACOG</th>
<th>ACMG</th>
</tr>
</thead>
<tbody>
<tr>
<td>✓ Cystic Fibrosis</td>
<td>✓ Spinal Muscular Atrophy</td>
<td>✓ Medium Chain Acyl-CoA Dehydrogenase Def</td>
<td></td>
</tr>
<tr>
<td>✓ Sickle Cell Disease and Hemoglobin E</td>
<td>✓ Tay-Sachs Disease</td>
<td>✓ Very Long Chain Acyl-CoA Dehydrogenase Def</td>
<td></td>
</tr>
<tr>
<td>✓ PKU</td>
<td>✓ Metachromatic Leukodystrophy</td>
<td>✓ Carnitine Palmitoyltransferase II Deficiency</td>
<td></td>
</tr>
<tr>
<td>✓ Methylmalonic Aciduria</td>
<td>✓ Atelosteogenesis Type 2</td>
<td>✓ POLG Related Disorder</td>
<td></td>
</tr>
<tr>
<td>✓ Congenital Myasthenic Syndrome</td>
<td>✓ Juvenile Nephronophthisis</td>
<td>✓ GJB2-Related Deafness</td>
<td></td>
</tr>
<tr>
<td>✓ Alpha-1 Antitrypsin Deficiency</td>
<td>✓ G6PD Deficiency</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

GeneAware Carriers Found by Baylor Miraca Genetics Laboratories.
NIPT

• NIPT Background & Technology
• Overview of *Illumina*’s NIPT Solution
  ▪ *Product Evolution & Clinical Evidence*
    • verifi® *test*
    • verifi® *Twins Panel*
    • verifi® *Microdeletions Panel*
• Illumina Sequencing & NIPT Technology Enablement
Overview

NIPT Background & Technology
Prenatal Prevalence of Reported Chromosomal Abnormalities

Data adapted from Wellesley, D, et al., Rare chromosome abnormalities, prevalence and prenatal diagnosis rates from population-based congenital anomaly registers in Europe. *Eur J of Hum Gen*, 11 January 2012.
### Conventional Prenatal Screening Options

**Detection Rates for Trisomy 21**

<table>
<thead>
<tr>
<th>Screening Option</th>
<th>Detection Rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1st Trimester Blood Screen</td>
<td>64-70</td>
</tr>
<tr>
<td>NT Ultrasound</td>
<td>64-70</td>
</tr>
<tr>
<td>1st Trimester Blood Screen NT Ultrasound</td>
<td>82-87</td>
</tr>
<tr>
<td>NT Ultrasound</td>
<td>64-70</td>
</tr>
<tr>
<td>Integrated Screen</td>
<td>69</td>
</tr>
<tr>
<td>2nd Trimester Blood Screen Serum</td>
<td>85-88</td>
</tr>
<tr>
<td>Quadruple Screen</td>
<td>81</td>
</tr>
<tr>
<td>Integrated Screen</td>
<td>94-96</td>
</tr>
<tr>
<td>Triple Screen</td>
<td>69</td>
</tr>
<tr>
<td>1st Trimester Blood Screen NT Ultrasound 2nd Trimester Blood Screen</td>
<td>94-96</td>
</tr>
<tr>
<td>False Positive Rate:</td>
<td>5%</td>
</tr>
</tbody>
</table>

ACOG Practice Bulletin No. 77, January 2007
Serum Screening

Combined Serum Screens, NT, Ultra-sound

CVS

Amnio

**Spectrum of Prenatal Testing**

- *SCREENING*
  - Risk scores are generated and modified based on biochemical analysis and population statistics

- *DIAGNOSTIC*
  - Results are based entirely on genetic factors

*Not meant to represent percentage of accuracy*
Goals of NIPT

*Superior test*

- Reduce exposure of risk to fetus
- Reduce false positives
- Testing that can easily be offered to pregnant women
- Enable a high detection rate
Cell Free DNA (cfDNA)

- cfDNA released through apoptosis
  - Released into bloodstream as small DNA fragments (150-200bp)
  - 2–20% of total cfDNA in maternal blood is fetal (from placenta)

- Ideal analyte for aneuploidy testing
  - Detected after 7 weeks gestation
  - Undetectable after 6 hours postpartum
Massively Parallel Sequencing (MPS)

Fetal DNA fragments in maternal blood.

Cell-free DNA fragments are then sequenced.

Compare the individual sequenced chromosomes against a reference for analysis.

Cell-free DNA sequenced via MPS

Alignment of reads

Chromosome 21

Reference chromosomes
Massively Parallel Sequencing (MPS)

1. Extract and Prepare cfDNA

2. Next-Gen DNA Sequencing
Massively Parallel Sequencing (MPS)

Alignment of Unique cfDNA Sequences

3

Alignment

CGATTTAACT

AGGTACCGAT

GACTTCCAGG

...ACCACGATTAACTGGAGTAAAGACTTCCAGGTACCGATCTAGCCT...

⇠ Human Genome ⇝

Millions of “counts” per sample

NOT TO SCALE
Detection of Fetal Aneuploidy

Fetal cfDNA (20%)

Maternal cfDNA

Chromosomes: 1 2 3 …… 21

Trisomy 21

4 Counting

10% more Chr21 cfDNA in T21

NOT TO SCALE

VS
The verifi® Prenatal Test Uses Massively Parallel Sequencing (MPS) *With whole-genome coverage: A superior approach*

**MPS provides precise, across-the-genome coverage**

- **Benefits**
  - Lowest assay failure rates <1%
  - Faster analysis time (faster TAT)

**Targeted sequencing is limited to few chromosomes, loci**

- **Drawbacks**
  - High assay failure rates of 4–12%
  - Slower analysis time (slower TAT)
verifi® Minimizes Test Failures

NIPT Test Failure Rates

Targeted Sequencing

Whole Genome Sequencing

- Natera¹: 6.4%
- Ariosa²: 4.5%
- Ariosa²: 4.6%
- Sequenom³: 5.5%
- Requires retest of sample; Marketed data

Only looked at SCA

Per submitted redraw

Why do test failures matter in NIPT?

• Increase risk for real false positive, false negative results
  – High rate of aneuploidy in test failures
  – Thus a “high risk” result potentially leading to increased invasive test utilization
  – Potential to increase false negative results if no action taken

• Redraw for NIPT is usually ineffective
  – High published redraw failure rates
  – Leads to increased TAT, office visits

Overview of Illumina’s NIPT Solution

Product Evolution & Clinical Evidence
Illumina’s NIPT (verifi® test) Portfolio

- **verifi** prenatal test
  - Monosomy X Option (FEB, 2012)
  - Sex Chromosomes Option (DEC, 2013)
  - Twins Option (OCT, 2014)
  - Microdeletion Panel T9, T16 (JUL, 2014)
verifi® Test Supported by Publications in Most Prestigious Journals in Field
### verifi® Test Performance

#### 2012 Clinical Validation

<table>
<thead>
<tr>
<th>Chromosome</th>
<th>Sensitivity</th>
<th>95% CI</th>
<th>Specificity</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>21</td>
<td>&gt;99.9% (90/90)</td>
<td>96.0–100.0</td>
<td>99.8% (409/410)</td>
<td>98.7–100.0</td>
</tr>
<tr>
<td>18</td>
<td>97.4% (37/38)</td>
<td>86.2–99.9</td>
<td>99.6% (461/463)</td>
<td>98.5–100.0</td>
</tr>
<tr>
<td>13</td>
<td>87.5% (14/16)</td>
<td>61.7–98.5</td>
<td>&gt;99.9% (485/485)</td>
<td>99.2–100.0</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Sex Chromosomes</th>
<th>Sensitivity</th>
<th>95% CI</th>
<th>Specificity</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>XX</td>
<td>97.6% (243/249)</td>
<td>94.8–99.1</td>
<td>99.2% (257/259)</td>
<td>97.2–99.9</td>
</tr>
<tr>
<td>XY</td>
<td>99.1% (227/229)</td>
<td>96.9–99.9</td>
<td>98.9% (276/279)</td>
<td>96.9–99.8</td>
</tr>
<tr>
<td>Monosomy X</td>
<td>95.0% (19/20)</td>
<td>75.1–99.9</td>
<td>99.0% (483/488)</td>
<td>97.6–99.7</td>
</tr>
</tbody>
</table>

- Clinical trial data from MELISSA study
  - Karyotype available for all samples ran

Original publication: Obstet Gynecol. 2012 May; 119(5): 890-901
Clinical Experience Publication

2013 Clinical Experience

- Commercial test accuracy emulates the clinical validation study
  - False positives: 0.2%
  - False negatives: 0.08%

- Average turn-around time of 5.1 days

- Test failure rate of 0.7%

verifi® Test Performance

2014 Clinical Experience (Autosomes)

Overall Test Performance

<table>
<thead>
<tr>
<th>n=34,306</th>
<th>Negative</th>
<th>Positive</th>
<th>False Negative (FN)</th>
<th>False Positive (FP)</th>
<th>Prevalence</th>
<th>Sensitivity*</th>
<th>Specificity*</th>
</tr>
</thead>
<tbody>
<tr>
<td>21</td>
<td>33710</td>
<td>596</td>
<td>5</td>
<td>19</td>
<td>1.70%</td>
<td>99.14%</td>
<td>99.94%</td>
</tr>
<tr>
<td>18</td>
<td>34098</td>
<td>208</td>
<td>3</td>
<td>33</td>
<td>0.52%</td>
<td>98.31%</td>
<td>99.90%</td>
</tr>
<tr>
<td>13</td>
<td>34235</td>
<td>71</td>
<td>1</td>
<td>18</td>
<td>0.16%</td>
<td>98.15%</td>
<td>99.95%</td>
</tr>
<tr>
<td>All</td>
<td>33431</td>
<td>875</td>
<td>9</td>
<td>70</td>
<td>2.37%</td>
<td>98.89%</td>
<td>99.79%</td>
</tr>
</tbody>
</table>

- For chromosome 21 PPV is 0.9681 and NPV is 0.9999
- Overall False Positive (FP) % is 0.2 and False Negative (FN) % is 0.026
  *Limited outcome data

- Over 34,000 patient cohort
- 0.1% technical failure rate
- 2-4 business day average turn around time

Positive Predictive Value and Negative Predictive Value

2014 Clinical Experience

- Positive Predictive Value (PPV)
  - Proportion of positive results that are true positive
- Negative Predictive Value (NPV)
  - Proportion of negative results that are true negative

PPV and NPV are population specific metrics and should not be compared between different populations.

<table>
<thead>
<tr>
<th>Chr</th>
<th>PPV</th>
<th>NPV</th>
</tr>
</thead>
<tbody>
<tr>
<td>21</td>
<td>0.97</td>
<td>0.9999</td>
</tr>
</tbody>
</table>
DNA Sequencing versus Standard Prenatal Aneuploidy Screening

Diana W. Bianchi, M.D., R. Lamar Parker, M.D., Jeffrey Wentworth, M.D., Rajeevi Madankumar, M.D., Craig Saffer, M.D., Anita F. Das, Ph.D., Joseph A. Craig, M.D., Darya I. Chudova, Ph.D., Patricia L. Devers, M.S., C.G.C., Keith W. Jones, Ph.D., Kelly Oliver, B.S., Richard P. Rava, Ph.D., and Amy J. Sehnert, M.D., for the CARE Study Group*
False Positive Rates Significantly Lower by NIPT
Across all trimesters

<table>
<thead>
<tr>
<th></th>
<th>NIPT%*</th>
<th>Standard Screen</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Trisomy 21</td>
<td>0.3% (6/1909)</td>
<td>3.6% (69/1909)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Trisomy 18</td>
<td>0.2% (3/1905)</td>
<td>0.6% (11/1905)</td>
<td>0.0325</td>
</tr>
</tbody>
</table>
Specificity and PPV Higher by NIPT

<table>
<thead>
<tr>
<th></th>
<th>Trisomy 21 (n = 5)</th>
<th></th>
<th>Trisomy 18 (n = 2)</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>NIPT % (95% CI)</td>
<td>Standard Screen % (95% CI)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Sensitivity</td>
<td>&gt;99.9 (47.8 – 100.0)</td>
<td>&gt;99.9 (29.2 – 100.0)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Specificity</td>
<td>99.7 (99.3 – 99.9)</td>
<td>96.4 (95.4 – 97.2)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>PPV</td>
<td>45.4 (16.7 – 76.6)</td>
<td>4.2 (0.9 – 11.7)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>NPV</td>
<td>&gt;99.9 (99.8 – 100.0)</td>
<td>&gt;99.9 (99.8 – 100.0)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Conclusions

- cfDNA testing merits serious consideration as a primary screening method for fetal autosomal aneuploidy compared to currently available aneuploidy screening markers
  - Reduced false positive rate
  - Higher positive predictive value
  - Minimize screening complexity and time to results

89% reduction in diagnostic invasive procedures necessary to confirm a positive screen result
verifi® Twins Panel
Twin Test Option Overview

- Available for monozygotic and dizygotic twin pregnancies
- Two options available for twins:
  - verifi® test for 21, 18, 13
  - verifi® test for 21, 18, 13, and the presence of Y
- Sex chromosome aneuploidy analysis not available for twins

Clinical Validation Data on Twins

- Improved algorithm validated with 115 twin samples
- Blinded analysis of twin samples in R&D lab**:
  - 3/3 T21 cases correctly classified
  - 1/1 T18 cases correctly classified
  - 91/91 cases with at least one male correctly classified
  - No T13 cases identified or collected
  - No false positive and no false negative results
- Samples collected from MELISSA and CARE studies

**Illumina (formerly Verinata Health) publication “Accurate Aneuploidy Detection in Twin Pregnancies using the SAFER Algorithm”, Data on File.
verifi® Microdeletion Panel, T9 & T16
Chromosomal Microarray versus Karyotyping for Prenatal Diagnosis

Findings from the extensive prenatal (NICHD) study to date:

- Wapner et al. found clinically relevant deletions/duplications in 6% of samples with ultrasound anomalies and normal karyotype
  - Highest incidence, 22q11 deletion (DiGeorge, ~3 Mb deletion in chromosome 22)
  - 17q12 deletion (~1–2 Mb) associated with autism and developmental delay
  - Many more aberrations —size range 0.05–10 Mb associated with developmental delay

These abnormalities can be determined prenatally.

As a group, microdeletions/microduplications are seen frequently in prenatal diagnosis, and are secondary to whole chromosome aneuploidies.
Noninvasive Detection of Fetal Subchromosome Abnormalities via Deep Sequencing of Maternal Plasma

Anupama Srinivasan,1 Diana W. Bianchi,2 Hui Huang,1 Amy J. Sehnert,1 and Richard P. Rava1,*

The purpose of this study was to determine the deep sequencing and analytic conditions needed to detect fetal subchromosome abnormalities across the genome from a maternal blood sample. Cell-free (cf) DNA was isolated from the plasma of 11 pregnant women carrying fetuses with subchromosomal duplications and deletions, translocations, mosaicism, and trisomy 20 diagnosed by metaphase karyotype. Massively parallel sequencing (MPS) was performed with 25-mer tags at approximately 10^9 tags per sample and mapped to reference human genome assembly hg19. Tags were counted and normalized to fixed genome bin sizes of 1 Mb or 100 kb to detect statistically distinct copy-number changes compared to the reference. All seven cases of microdeletions, duplications, translocations, and the trisomy 20 were detected blindly by MPS, including a microdeletion as small as 300 kb. In two of these cases in which the metaphase karyotype showed additional material of unknown origin, MPS identified both the translocation breakpoint and the chromosomal origin of the additional material. In the four mosaic cases, the subchromosomal abnormality was not demonstrated by MPS. This work shows that in nonmosaic cases, it is possible to obtain a fetal molecular karyotype by MPS of maternal plasma cfDNA that is equivalent to a chromosome microarray and in some cases is better than a metaphase karyotype. This approach combines the advantage of enhanced fetal genomic resolution with the improved safety of a noninvasive maternal blood test.
Common Microdeletion Syndromes

- Typically include developmental delay/intellectual disability & birth defects
- Usually de novo
- Equal risk for all pregnant women

<table>
<thead>
<tr>
<th>Syndrome</th>
<th>Incidence</th>
</tr>
</thead>
<tbody>
<tr>
<td>22q11 Deletion Syndrome</td>
<td>1 in 4,000</td>
</tr>
<tr>
<td>1p36 Deletion Syndrome</td>
<td>1 in 4,000-10,000</td>
</tr>
<tr>
<td>Prader-Willi Syndrome (15q11.2)</td>
<td>1 in 10,000-25,000</td>
</tr>
<tr>
<td>Angelman Syndrome (15q11.2)</td>
<td>1 in 12,000</td>
</tr>
<tr>
<td>Cri-du-chat Syndrome (5p-)</td>
<td>1 in 20,000-50,000</td>
</tr>
<tr>
<td>Wolf-Hirschhorn Syndrome (4p-)</td>
<td>1 in 50,000</td>
</tr>
</tbody>
</table>

Compare incidence to the common aneuploidies:
- Down syndrome – 1/740
- Trisomy 18 – 1/6000
- Trisomy 13 – 1 in 12000
- MX – 1/5000
- Other SCAs – 1/2000 each
verifi® Microdeletion Panel

**Rationale for Product Use**

Microdeletion Panel is an optional screening panel used in a clinical context such as abnormal ultrasound in current pregnancy, family history of microdeletion syndrome, or other indications.

- Microdeletions are not part of routine serum screening
- Microdeletions are common and more severe than Down syndrome
- Can occur spontaneously without family history
- Prenatal incidence is independent of maternal age or ethnicity
verifi® Microdeletion Panel Test Performance

91.6% Sensitivity, 99.84% Specificity in Clinical Samples

Table 1: Performance Specifications of the verifi microdeletion panel

<table>
<thead>
<tr>
<th>Syndrome</th>
<th>22q11.2 deletion syndrome (DiGeorge)</th>
<th>1p36 deletion syndrome</th>
<th>Prader-Willi/ Angelman syndrome</th>
<th>Cri du chat syndrome</th>
<th>Wolf-Hirschhorn syndrome</th>
</tr>
</thead>
<tbody>
<tr>
<td>Min. Syndrome Region Size</td>
<td>2.7 Mb</td>
<td>5 Mb</td>
<td>5.8 Mb</td>
<td>9.8 Mb</td>
<td>3.6 Mb</td>
</tr>
<tr>
<td><strong>Sensitivity</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No. Affected Samples Tested</td>
<td>8</td>
<td>0</td>
<td>0</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>No. Samples Detected</td>
<td>7</td>
<td>0*</td>
<td>0*</td>
<td>2</td>
<td>2</td>
</tr>
<tr>
<td>% Sensitivity [95% CI]</td>
<td>87.5% [47–99]</td>
<td>1</td>
<td>1</td>
<td>100% [15–100]</td>
<td>100% [15–100]</td>
</tr>
<tr>
<td><strong>Specificity</strong></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>No. Putative Unaffected Samples Tested</td>
<td>1797</td>
<td>1797</td>
<td>1797</td>
<td>1797</td>
<td>1797</td>
</tr>
<tr>
<td>No. Samples Detected</td>
<td>0</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>% Positive Call Rate</td>
<td>0% [0–0.2%]</td>
<td>0% [0–0.1%]</td>
<td>0.05% [0.01–0.31%]</td>
<td>0% [0–0.2%]</td>
<td>0.11% [0.01–0.4%]</td>
</tr>
<tr>
<td>% Specificity [95% CI]</td>
<td>&gt; 99.8%</td>
<td>&gt; 99.9%</td>
<td>&gt; 99.7%</td>
<td>&gt; 99.8%</td>
<td>&gt; 99.6%</td>
</tr>
</tbody>
</table>

* Titration of fragmented genomic DNA derived from cell lines containing either a 1p36 or 15q11.2 deletion demonstrated a linear dose response and confirmed the assay's ability to measure copy number change at these loci.

† No estimates of confidence intervals or sensitivity were performed for sample sizes < 2.
# verifi® Prenatal Test

**Product Profile**

| Chromosomes Analyzed | Singletons: 21, 18, 13, and Optional X and Y  
| Optional Microdeletion Panel  
| Optional 9, 16  
| Twins: 21, 18, 13  
| Optional Y |

| Blood draw requirement | 1 blood tube (7–10mL) |

| Patient Eligibility | Validated in high-risk pregnancies and general population  
| Singletons and twins at ≥10 weeks gestation  
| Egg donors accepted |

| Sample collection | On-site collection kits, ambient shipping |

| Time to Result | 2-4 business days |

| Test Failure Rate | 0.1%* |

---

Why verifi®?

- Highest performance backed by NGS leader
  - Lowest assay failure rates of 0.1%
  - Fastest time to report, 2 - 4 business days
  - Best PPV with a low failure rate
  - Can detect aneuploidy at fetal fractions as low as 1.4%

- Broad coverage of genetic conditions seen in clinical setting
  - Can be performed in pregnancies where an egg donor was used and twins, bone marrow, etc.
  - Microdeletion panel, with superior performance
  - Ability to rapidly add new content

- Clinical results you can trust
  - Published clinical experience on over 34,000 patients
    - 0.02% False Negative %
    - 0.2% False Positive %

- Genetic Counseling Support
Genetic Counseling NIPT Flipbook

- 10 pages with illustrations to help patients understand the NIPT testing process, conditions tested, result interpretation
- Used by healthcare providers prior to NIPT
- verifi® prenatal test specific
- Available in 9 languages: English, Spanish, Portuguese, French, German, Italian, Korean, Japanese, Chinese
- Download at www.verifitest.com Tools For Your Practice
Patient Education Video

- 12 minute video providing an overview of the benefits and limitations of various prenatal testing options
  - Prenatal screening (e.g. first trimester combined screen)
  - CVS/Amniocentesis
  - NIPT (verifi® prenatal test specific)
    - Includes description of conditions tested
- Healthcare providers can direct their patients to watch this video in the clinic or at home prior to their OB appointment
- Available in 9 languages: English, Spanish, Portuguese, French, German, Italian, Korean, Japanese, Chinese
- Can be viewed at www.verifitest.com
- Tools For Your Practice
  - downloadable to other practices’ websites
Hereditary Cancer Panels
Test Designer
Lee-Jun Wong, PhD – Lab Director

• Experience
  – World renowned geneticist in hereditary disorders and technology development
  – 22 years in diagnostics of genetic diseases
  – Published >250 peer reviewed papers
  – First to develop NGS-based test for simultaneous detection of point mutations and CNVs in dual genomes
  – First to validate NGS for quantitative mutation analysis
Technology

Next-generation sequencing and deletion/duplication analysis
## Hereditary Cancer Tests

<table>
<thead>
<tr>
<th>Test</th>
<th># Genes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Comprehensive Hereditary Test</td>
<td>61</td>
</tr>
<tr>
<td>Hereditary Brain/CNS/PNS Cancer</td>
<td>17</td>
</tr>
<tr>
<td>Hereditary Reproductive Cancer</td>
<td>23</td>
</tr>
<tr>
<td>Hereditary Renal Cell Carcinoma</td>
<td>12</td>
</tr>
<tr>
<td>Hereditary Endocrine Cancer</td>
<td>15</td>
</tr>
<tr>
<td>Hereditary Leukemia/Lymphoma</td>
<td>13</td>
</tr>
<tr>
<td>Comprehensive Hereditary GI Cancer</td>
<td>22</td>
</tr>
<tr>
<td>Hereditary Melanoma</td>
<td>4</td>
</tr>
<tr>
<td>Hereditary High Risk Breast</td>
<td>7</td>
</tr>
<tr>
<td>Paraganglioma &amp; Pheochromocytoma</td>
<td>9</td>
</tr>
<tr>
<td>Hereditary High Risk Colorectal</td>
<td>11</td>
</tr>
<tr>
<td>Hereditary Pancreatic</td>
<td>16</td>
</tr>
<tr>
<td>Hereditary Prostate</td>
<td>5</td>
</tr>
</tbody>
</table>
Why Order Panels?

BREAST CANCER RISK (PENETRANCE)

- General Population
- Moderate
- High

MUTATION FREQUENCY

Very Rare  Rare  Common

BRCA1  BRCA2  CDH1  PALB2  PTEN  STK11  TP53
ATM  MSH6  BARD1  MUTYH  BRIP1  NBN  CHEK2  PMS1  EPCAM  PMS2  MLH1  RAD50  MRE11A  RAD51C  MSH2  RAD51D
Unique Features of BCM Panels

- 100% deep coverage of all coding regions of genes.
- Detection of single nucleotide mutations, small insertion/deletion mutations, and simultaneously detect exonic deletions.
- Less variability and better consistency in results allow for more accurate gene coverage and copy number analysis
- Board certified, leading experts in clinical and diagnostic mutations.
Our Confirmation Steps

• All mutations detected in the panels will be confirmed using a secondary method
• All copy number changes will be assessed by next-generation sequencing and a custom hereditary cancer array
• BMGL is the ONLY clinical laboratory that confirms mutations and copy number changes by two separate methods
Reporting - Five Tier Classification System

Class 5 - Definitely Pathogenic

Class 4 - Likely Pathogenic

Class 3 – Uncertain (VUS)

Class 2 - Likely Not Pathogenic or of Little Clinical Significance

Class 1 - Not Pathogenic or of No Clinical Significance
Class 5

Definitely Pathogenic

- Frameshift and nonsense variants
- Intron variants occurring in the consensus splice acceptor or donor sequence sites
  - Sequence >20 base pairs into intronic region
- Missense variants, in-frame deletion, insertion and indels that have been conclusively demonstrated
Classes 2 - 4

- **Likely pathogenic (class 4):** Database/literature suggest pathogenic; family history and additional clinical information suggest pathogenic; probability analysis is deleterious

- **Likely benign (class 2):** Family history and additional clinical notes do not fit as pathogenic; insufficient database/literature information; probability prediction is benign

- **Uncertain (class 3):** Cannot be classified as Class 2 or 4
Class 1

Not Pathogenic/Benign variants or of no Clinical Significance

- Variants of common polymorphisms seen in greater than 1% of alleles in the general population and rare variants that display little or no association with cancer risk in families
Curation pipeline

Evidence
- Databases
- Literature search
- Experimental evidence
- Computational algorithms

Curation
- BMGL designed system

Review
- GCs
- Ph.D.s
- Lab directors

Variant classification
- based on
  - Molecular & functional damage
  - Clinical pathogenicity

Yearly re-curation
Breast and ovarian cancer

Breast Cancer
- Sporadic: 15-20%
- Familial: 5-10%
- Hereditary: 70-80%

Ovarian Cancer
- Sporadic: 85-90%
- Hereditary: 10-15%
Red Flags

• Breast cancer before the age of 50
• Ovarian cancer at any age
• Male breast cancer at any age
• Two primary breast cancers in an individual
• Two or more breast cancer in a family, with one diagnosed under 50
• Ashkenazi Jewish ancestry
• Previously identified *BRCA* mutation in a family
Why test?

- Increased screening
- Risk reducing surgery
- Chemoprevention
- Identifying other family members who may be at an increased risk
Screening and Prevention

• Screening Guideline Recommendations
  – Breast:
    • Screening:
      – 25-29 years old, annual breast MRI (preferred or mammogram if MRI is unavailable)
      – 30-75 years old, annual mammogram and breast MRI screening
    • Risk reducing surgery:
      – Prophylactic mastectomy
      – Prophylactic BSO
Screening and Prevention

– Ovarian:
  • Screening:
    – Beginning at age 30 (or 5-10 y prior to earliest diagnosis)
    – Transvaginal ultrasound and CA-125 blood test every six months
  • Risk reducing surgery:
    – Prophylactic TAH/BSO
Results delivery

SIGN IN

Please enter your username and password. Don't have an account? Click here to Register.
You may also use your GeneAware username and password to access the portal.

**Account Information**

Username: 
Password*: 

*Password is case sensitive.  Forgot password?
The people you do business with are as important, if not more important than the products that they sell.

— Jack Welch