GENETICS UPDATE 2019:
FROM ANEUPLOIDY SCREENING TO FETAL EXOME SEQUENCING

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DISCLOSURES

• None
GENETIC SCREENING IN PREGNANCY

• Aneuploidy screening
  – Serum analyte
    • Quad/Integrated
  – cfDNA
• Carrier screening
  – Focused
  – Pan-ethnic approach
ANEUPLOIDY SCREENING RECOMMENDATIONS

• “All women should be offered aneuploidy screening...regardless of maternal age.”
• “Not practical to choose from a large array of screening tests”

Screening for Fetal Chromosome Anomalies, ACOG PB 77, 2007
Reaffirmed: Screening for Aneuploidy ACOG PB 163, 2016
# CURRENT ANEUPLOIDY SCREENING OPTIONS

<table>
<thead>
<tr>
<th></th>
<th>DETECTION RATE</th>
<th>FALSE POSITIVE RATE</th>
</tr>
</thead>
<tbody>
<tr>
<td>Quad</td>
<td>75%</td>
<td>~7%</td>
</tr>
<tr>
<td>First Trimester</td>
<td>85%</td>
<td>~7%</td>
</tr>
<tr>
<td>Integrated</td>
<td>90%</td>
<td>~1-2%</td>
</tr>
<tr>
<td>CFDNA</td>
<td>98%</td>
<td>0.05%</td>
</tr>
</tbody>
</table>
CELL-FREE WHAT?

- **CFDNA**: Cell Free DNA
- **cffDNA**: Cell free fetal DNA
- **NIPT**: Noninvasive Prenatal Testing
- **NIPS**: Noninvasive Prenatal Screening

- Commercial Laboratories’ Brands:
  - Progenity: *Innatal*
  - LabCorp: *InformaSeq*
  - Counsyl: *Prelude Pregnancy Screen*
  - Natera: *Panorama*
  - Ariosa: *Harmony*
CFDNA: BACKGROUND WITH REGARD TO PREGNANCY

- Short, 200 bp DNA fragments in maternal plasma
- Source is both “fetal” and maternal
  - The fetal component is placental
  - Trophoblast apoptosis
- 3-13% of total CFDNA
CURRENT CLINICAL APPLICATIONS FOR THE PRENATAL USE OF CFDNA

• Aneuploidy Screening: 13, 18, 21, X and Y
• Sex determination for X-linked disorders
  – Example: Hemophilia
• RhD status
  – Rh-negative mother
    • Whether or not alloimmunized
      – Rh-Immune globulin prophylaxis
      – Therapy
• Paternally-inherited AD genetic disorder with a known mutation
  – Example: Neurofibromatosis
CFDNA

• Can sample from 9 weeks until term
• Of the three trisomies, the accuracy is best for Down syndrome.
• No risk for miscarriage
• Undetectable at about 2 hours postpartum
ACOG STATEMENTS ON CFDNA SCREENING

• CAN BE USED IN ALL WOMEN
  – Provides less information in younger women than conventional screening

• SHOULD NOT BE USED:
  – In multiple gestations
  – To screen for microdeletions

ACOG Practice Bulletin 163, 2016
CFDNA vs. ANALYTE SCREENING

**CFDNA:**
- Down Syndrome
- Trisomy 13
- Trisomy 18
- Triploidy (one technique)
- Sex Chromosome Aneuploidies
- ? Microdeletions

**Analyte Screening:**
- Down Syndrome
- Trisomy 13
- Trisomy 18
- Triploidy
- Multiple gestations
- Neural tube defects
- Ventral wall defects
- Placental insufficiency
  - IUGR and IUFD
- Smith-Lemli-Opitz
- Steroid Sulfatase Def
MEDICAL COST CONTAINMENT

• Price depends on insurance contracts
• Medicaid contracts
• Many patients have a high out-of-pocket deductible
  • Time of year matters
• Often, analyte screening is more cost-effective (and is still a good screening test!)
IS CFDNA SCREENING REASONABLE REGARDLESS OF MATERNAL AGE?

- If testing is just about aneuploidy screening:
  - Detection rate is the same.
  - False positive rate a bit higher in younger women
- Laboratories have similar results and detection rates
  - Don’t be swayed by different bells and whistles
- Think about cost containment for your patients
UNINTENDED INFORMATION DERIVED FROM CFDNA SCREENING

• Most laboratories cannot distinguish a maternal result from a fetal result!
  – The microdeletion syndrome you identify may be maternal
  – Beware the patient with an organ transplant
  – Maternal oncologic diagnoses
  – 45, X results may reflect older mothers, not fetuses
CFDNA INCIDENTAL FINDINGS, CASE 1

- 40 year old patient
- Nuchal translucency normal
- CfDNA 45, X result
- Serial sonograms: apparently normal female
- Genetic amniocentesis: 46,XX
- Maternal karyotype 45,X[5]/46,XX [45]
  - Interpretation: normal maternal aging
LOSS OF X CHROMOSOMES WITH INCREASING MATERNAL AGE

Regression fit p < 0.001

% cellsXCL

Median age

CFDNA INCIDENTAL FINDINGS, CASE 2

- 35 Year old G1P0
- CfDNA: 45, X; likely maternal result suggested due to volume of 45, X material
- Mosaic 45, X result confirmed by maternal karyotype:
  - 45 X (17), 46 XX (35)
- Maternal phenotype:
  - Mild hearing loss, bone density issues, bicuspid aortic valve, short stature, normal intelligence, thyroid disorder
- Fetal Karyotype could only confirm absence of Y chromosome
MATERNAL MANAGEMENT, CASE 2

• Physical examination/genetic counseling
  – Discussion of medical conditions associated with mosaic 45, X.
  – Discussion of risk of premature ovarian insufficiency
• Referral for medical management
CFDNA INCIDENTAL FINDINGS, CASE 3

• 36 year old G1P0
  – CFDNA: 47, XXX fetal result
    • cfDNA screen results also show multiple chromosomal aberrations (nonspecific)
      – Call to the laboratory:
        • Gain of chromosome X
        • Losses of chromosomes 1p, 14q, 15q, and 19q
REPORT ADDENDUM

• “Bioinformatic review of genome-wide data revealed multiple chromosomal aberrations. Similar patterns of multiple chromosomal aberrations have been observed in cases of known maternal neoplasm... These findings may reflect chromosomal status of the fetus, placenta (confined placental mosaicism), and/or the patient, may be due to a maternal condition, or may be a false positive. Any chromosome aberration(s) found should be followed by further testing”.
CFDNA INCIDENTAL FINDINGS, CASE 3

• Two issues, Fetal and Maternal:
  1. 47, XXX counseling for the fetal result
  2. Multiple fragments suggestive of a maternal neoplasm
     • Could be a myoma!
     • Could be a malignancy?
       – One report:
          » 55 altered genomic profiles
             • 40 neoplasms
                • 18 malignancies
                • 20 myomas

CANCER IN PREGNANCY

• Incidence 1:1000-1:1500 pregnancies
• Most common malignancies:
  – Melanoma
  – Breast
  – Thyroid
  – Colon
  – Cervical
  – Ovarian
  – Hematologic
# Causes of Death Among Reproductive Age Women

<table>
<thead>
<tr>
<th>Age 15-19</th>
<th>Age 20-24</th>
<th>Age 25-34</th>
<th>Age 35-44</th>
</tr>
</thead>
<tbody>
<tr>
<td>Unintentional injuries 39.5%</td>
<td>Unintentional injuries 42.4%</td>
<td>Unintentional injuries 32.6%</td>
<td>Cancer 22.8%</td>
</tr>
<tr>
<td>Suicide 17.5%</td>
<td>Suicide 11.8%</td>
<td>Cancer 12.0%</td>
<td>Unintentional injuries 19.2%</td>
</tr>
<tr>
<td><strong>Cancer 7.9%</strong></td>
<td>Homicide 7.5%</td>
<td>Suicide 9.2%</td>
<td>Heart disease 12.0%</td>
</tr>
<tr>
<td>Homicide 7.1%</td>
<td><strong>Cancer 6.8%</strong></td>
<td>Heart disease 7.8%</td>
<td>Suicide 6.2%</td>
</tr>
<tr>
<td>Heart disease 3.6%</td>
<td>Heart disease 4.3%</td>
<td>Homicide 4.9%</td>
<td>Chronic liver disease 3.8%</td>
</tr>
<tr>
<td>Birth defects 2.9%</td>
<td>Pregnancy complications 2.3%</td>
<td>Pregnancy complications 2.7%</td>
<td>Stroke 2.9%</td>
</tr>
<tr>
<td>Pregnancy complications</td>
<td>Birth defects 1.6%</td>
<td>Diabetes 2.1%</td>
<td>Diabetes 2.7%</td>
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</table>

CDC 2015
SUGGESTED EVALUATION PLAN

Box 1. Stepwise Evaluation of the Patient With More Than One Aneuploidy Detected on Cell-Free DNA

1. Discuss results with performing laboratory

2. History and physical examination with laboratory evaluation
   - Complete blood count with peripheral smear
   - Metabolic panel
   - Pap test
   - Fecal occult blood

3. Chest radiograph

4. Magnetic resonance image of the chest, abdomen, and pelvis

5. Consider annual complete blood count for surveillance
THE BOTTOM LINE

• Don’t ignore test results that are unusual, don’t make sense, or that you don’t understand.

• Don’t be afraid to call the lab that did the report and speak to a genetic counselor or lab director.

• Then, refer to the genetics professional in your region.

• This is an evolving field in which management is not always clear.
WHY IS SCREENING FOR ANEUPLOIDY VALUABLE?

• A way to gain more **information** about the pregnancy
  – Reassurance
  – Planning
  – Patient Autonomy
  – Decision-making
  – Education/preparation
  • May affect antepartum care and delivery management
CONCLUSIONS FOR GENETIC PRENATAL SCREENING

• Offer Laboratory-based aneuploidy screening to everyone!
  – Document it
  – Fine for patients to decline
• Offer Carrier Screening ONCE to Individuals:
  – CF, SMA, ask about intellectual disability and POI re: Fragile X
  – Thalassemia screening if indicated.
PRE- AND POST-TEST COUNSELING GUIDANCE

• ACOG:
  – www.acog.org/More-Info/cfDNA

• National Society of Genetic Counselors:
  – http://nsgc.org/page/non-invasive-prenatal-testing-healthcare-providers
  – http://nsgc.org/page/abnormal-non-invasive-prenatal-testing-results
DIAGNOSTIC TESTING IN PREGNANCY: ARE WE STILL DOING THIS?

- CFDNA will always remain a screening test.
- Patients with a high a priori risk may want direct testing.
  - If tested prenatally, we can guide care
    - Preparation
    - Improve decision-making for newborn therapies
- Newborns with anomalies will get tested eventually
OUR STANDARD MANAGEMENT OF A PRENATAL DIAGNOSIS SAMPLE

FETAL ANOMALY ON ULTRASOUND

GENETIC COUNSELING

DIAGNOSTIC TESTING:

FISH (13, 18, 21, X, Y)

POSITIVE

NEGATIVE

KARYOTYPE

MICROARRAY
LIMITATIONS OF PRENATAL DIAGNOSIS TODAY

- Karyotype
- Microarray (CMA): a lab technique to measure gains or losses of genetic material compared to a standard
  - Adds 6% higher detection rate over karyotype if anomalies are seen
- What’s next if CMA doesn’t produce a diagnosis?
  - Fetal exome sequencing
WHAT IS EXOME SEQUENCING?

• A laboratory technique for sequencing all of the protein-coding genes in the entire genome.
  – These protein-coding genes in the DNA are known as the exome.
    • 3 billion nucleotides (base pairs)
      – Only about 1.5% codes for 20,000 functional genes in the human genome
        » Of these, there are about 4000-5000 disease-associated genes known as “clinical exomes”.
CONGENITAL ANOMALIES

• Affect 2-4% of all newborns
• Responsible for about 20% of perinatal deaths
• 70-80% of anomalous fetuses have a normal chromosomal microarray result
• Fetal exome sequencing:
  – Routine use in adults and children with dysmorphic features
    • About a 30% diagnostic yield
    • May identify a molecular diagnosis where standard genetic testing (karyotype, microarray, and targeted molecular panels) have not.

LIMITATIONS OF EXOME SEQUENCING

- Diagnostic procedure required for sample
- Interpretation challenges
- Turn around time
- Trios needed (mom, dad, fetus)
  - Paternity/consanguinity issues
- Lack of available databases correlating genotype with phenotype
  - The problem of genetic variant interpretation
- Lack of providers for counseling and management
- Cost
BIG TIME Genetics

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