Noninvasive Prenatal Testing for Fetal Aneuploidy, With or Without Microdeletions

Indications for Ordering

- First- or second-tier screening test for the most common fetal aneuploidy disorders: trisomy 13, trisomy 18, trisomy 21 (Down syndrome), Turner syndrome, sex chromosome aneuploidies (XXX, XXY, XYY), and triploidy
- Test utilizes placental cell-free DNA (cfDNA) found in the maternal blood to identify women with a fetus at increased risk for the targeted disorders
- Screening test for pregnant women at 9w0d gestation to term

Test Description

PCR followed by next generation sequencing

Tests to Consider

Primary tests

Non-Invasive Prenatal Testing for Fetal Aneuploidy 2007537

- Screening for the most common fetal aneuploidy disorders in pregnant women (9w0d-term)
- Disorders screened for include:
  - Trisomy 13 (T13)
  - Trisomy 18 (T18)
  - Down syndrome (trisomy 21 or T21)
  - Turner syndrome (TS)
  - Triploidy
  - Sex chromosome aneuploidies (XXX, XXY, XYY) will be reported if identified
- Test may be ordered for
  - Singleton and twin gestations
  - Pregnancies achieved using an egg donor or surrogate
- Women at increased risk should be offered diagnostic testing (CVS or amniocentesis) instead of screening
- Increased risk includes
  - Women with a previous fetus with autosomal aneuploidy
  - Women ≥35 years at delivery
  - Women with an increased risk for aneuploidy based on multiple marker screening
  - Women with fetal ultrasound findings suggestive of T13, T18, T21, or TS
  - Either parent is a carrier of a Robertsonian translocation involving chromosomes 13 or 21
- Not recommended when the woman or her partner
  - Is a known carrier of a translocation or other chromosome rearrangement that will not result in a fetus with one of the above disorders
  - Has a known numerical abnormality in one of the targeted chromosomes (eg, mosaic T21 or TS)
- Not recommended for women
  - Who have a known twin demise (vanished twin)
  - Who have had an allogenic bone marrow transplant
  - Who are carrying twins *and* used an egg donor/surrogate

Non-Invasive Prenatal Testing for Fetal Aneuploidy with 22q11.2 Microdeletion 2013142

- Screening for whole chromosome fetal aneuploidy involving chromosomes 13, 18, 21, X, Y, and triploidy in pregnant women (9w0d-term)
- Also screens for microdeletions causing 22q11.2 deletion syndrome (DiGeorge/velocardiofacial syndrome [VCFS])
- Useful when the fetus is identified as having a heart defect and/or other findings suggestive of del22q11.2
- May identify presence of 22q11.2 deletion in the patient
- Test may be ordered for
  - Singleton and monozygotic twin gestations
- Not recommended for women
  - Who have a known twin demise (vanished twin)
  - Who are carrying dizygotic twins
  - Who have used an egg donor
  - Surrogates who have not used their own egg
  - Who are carrying twins *and* used an egg donor/surrogate

Non-Invasive Prenatal Testing for Fetal Aneuploidy with Microdeletions 2010232

- Screening for whole chromosome fetal aneuploidy involving chromosomes 13, 18, 21, X, Y, and triploidy in pregnant women (9w0d-term)
- Also screens for microdeletions causing
  - 22q11.2 deletion syndrome (DiGeorge/VCFS)
  - 1p36 deletion syndrome
  - Angelman syndrome
  - Prader-Willi syndrome
  - Cri-du-chat (5p-) syndrome

- Not recommended for women
  - Carrying more than one fetus or who have a known twin demise (vanished twin)
  - Who have used an egg donor
  - Surrogates who have not used their own egg

Related tests (screening for low-risk individuals)

Maternal Serum Screening, Integrated, Specimen #1 0081062 (first trimester) **AND**

Maternal Serum Screening, Integrated, Specimen #2 0081064 (second trimester)

- Screening tests for T21, T18, and open neural tube defect (ONTD)
  - Risks determined using a combination of first- and second-trimester serum markers, with or without first-trimester nuchal translucency (NT) measurement
- Risks provided after second-trimester specimen is received

Maternal Screening, Sequential, Specimen #1 0081293 (first trimester) **AND**

Maternal Screening, Sequential, Specimen #2 0081294 (second trimester)

- First trimester – screens for T21 and T18
- Second trimester – screens for T21, T18, and ONTD
- Requires NT measurement performed by an ultrasonographer certified by the Fetal Medicine Foundation (FMF) or the Nuchal Translucency Quality Review (NTQR)
- Risks provided in both first and second trimesters

Maternal Serum Screen, First Trimester 0081150

- First trimester – screens for T21 and T18
- Does not include AFP for ONTD screening
- Requires NT measurement performed by an ultrasonographer certified by FMF or NTQR

Maternal Serum Screen, Alpha Fetoprotein, hCG, Estriol, and Inhibin A 0080269

- Second trimester – screens for T21, T18, and ONTD

Related tests (diagnostic testing)

Chromosome Analysis, Amniotic Fluid 2002293

- Prenatal chromosome analysis on amniotic fluid when individual
  - Is at increased risk for fetal aneuploidy based on maternal age, abnormal noninvasive prenatal testing (NIPT), abnormal multiple marker screening, or abnormal fetal ultrasound
  - Has a family history of chromosome abnormality or genetic disorder
  - Desires diagnostic testing instead of screening

Chromosome Analysis, Chorionic Villus 2002291

- Prenatal chromosome analysis on chorionic villi when individual
  - Is at increased risk for fetal aneuploidy based on maternal age, abnormal NIPT, abnormal multiple marker screening, or abnormal fetal ultrasound
  - Has a family history of chromosome abnormality or genetic disorder
  - Desires diagnostic testing instead of screening

Cytogenomic SNP Microarray – Fetal 2002366

- Diagnostic test designed to identify genomic abnormalities (eg, aneuploidy and microdeletions)
- Performed on direct or cultured amniotic fluid and chorionic villus sampling (CVS) specimens

Disease Overview

Incidence at birth

- T13 – 1/5,000
- T18 – 1/3,000
- T21 – 1/600
- TS – 1/2,500 female births
- 1p36 deletion – 1/5,000
- 5p deletion – 1/20,000
- 22q11.2 deletion – 1/2,000
- Angelman – 1/12,000
- Prader-Willi – 1/10,000
- Sex chromosome aneuploidies – 1/250-300
- Triploidy – 1/50-100 conceptuses (very rare at birth)

Diagnostic issues

- Traditional noninvasive screening methods (maternal serum biochemical markers with or without fetal NT)
  - Detect only 70-95% of fetal T21
  - Detect 60-90% of fetal T18
  - Do not detect microdeletions
  - Can have screen positive rate >20% in women >35 years
- High false-positive rates, associated with traditional prenatal aneuploidy screening methods, result in unnecessary invasive diagnostic procedures
  - Invasive procedures (eg, amniocentesis, CVS) carry a small risk of pregnancy loss
- cfDNA screening combines very high sensitivity (>99%) with very high specificity (>99%) for fetal aneuploidy
  - Should not be used in place of routine ultrasound or diagnostic testing for chromosomal aneuploidies

- Positive predictive value (PPV) is the likelihood that a positive NIPT screen will be confirmed as positive through a diagnostic test, which varies based on chromosome abnormality, maternal age, and gestational age
  - Average PPV by chromosome (Dar, 2014)
    - T13 – 38%
    - T18 – 93%
    - T21 – 91%
    - TS – 50%
- Microdeletion risk is very low (1/1,000) in most pregnancies and does not increase with maternal age
  - Pretest genetic counseling should be considered to help women fully understand the benefits and limitations of microdeletion screening

Test Interpretation

Sensitivity/specificity

- Aneuploidy
  - Clinical sensitivity/specificity – >99%
- Microdeletions
  - Clinical sensitivity – >99%
  - Clinical specificity – >94%
- While the sensitivity and specificity are very high, both false-positive and false-negative test results can still occur due to confined placental mosaicism, fetal mosaicism, low fetal fraction, and the rarity of some of the disorders
- Please see positive predictive value above, or contact an ARUP genetic counselor if you have questions

Results

Each risk assessment includes the fetal fraction (FF), fetal sex (unless patient history form indicates this information is not desired), and the age-related/pretest risk

- High risk – significantly increased risk for fetus to have trisomy of chromosomes 13, 18, or 21; monosomy X; triploidy; or a deletion causing 22q11.2 microdeletion; 1p36 deletion; Angelman, cri-du-chat, or Prader-Willi syndromes
  - Although not specifically targeted, results that suggest fetal 47,XXX, 47,XXY, or 47,XYY will be reported
  - **As these are screening tests, diagnostic testing (eg, fetal karyotype by CVS or amniocentesis) is required to confirm abnormal results before irreversible action is taken**
- Low risk – very low risk for fetus to have abnormality of chromosomes 13, 18, 21, X, Y; triploidy; or a deletion causing 22q11.2 microdeletion (DiGeorge/VCFS); 1p36 deletion; Angelman, cri-du-chat, or Prader-Willi syndromes
  - Fetal karyotype or other test may still be appropriate if
    - Fetal anomalies are detected by ultrasound
    - Other concerns exist regarding the health of the fetus
- No result – unable to confidently report "high risk" or "low risk" test result (~3% of specimens) and may be due to
  - Insufficient fetal DNA in maternal specimen (low fetal fraction)
  - Presence of mosaicism in one of the targeted chromosomes in the fetus, placenta, or mother
  - Parents of fetus are blood relatives
- Unchanged (microdeletion test results only)
  - Unable to determine if the risk to have a child with a deletion is either increased or decreased, usually due to a low (<7%) fetal fraction
    - Population risk will be reported

Limitations

- Unless otherwise specified in the test descriptions above, inappropriate for women who
  - Are carrying more than one fetus
  - Are not carrying their biological offspring (eg, pregnancies resulting from an egg donor, or when the woman is a surrogate)
  - Have undergone allogeneic bone marrow transplant
  - In any of the above circumstances, cfDNA screening by alternative method may be available
- Aneuploidy for chromosomes other than 13, 18, 21, X, and Y will not be detected
- Fetal mosaicism may not be detected
- Low FF may occur normally in some pregnancies and can affect the ability to report a result
  - Individuals with elevated body mass index (BMI) are at increased risk of having a low FF
    - May result in increased chance of a false-positive, false-negative, or no-call test result
- Maternal factors (eg, BMI or current cancer diagnosis) may affect FF or cfDNA analysis

References

- Dar P, Curnow KJ, et al. Clinical experience and follow-up with large scale single-nucleotide polymorphism-based noninvasive prenatal aneuploidy testing. Am J Obstet Gynecol. 2014; 211(5):527.e1-527.e17
- Pergament E, Cuckle H, et al. Single-nucleotide polymorphism-based noninvasive prenatal screening in a high-risk and low-risk cohort. Obstet Gynecol. 2014;124:210-218