Use of Cell-Free Fetal DNA in Maternal Plasma for Noninvasive Prenatal Screening

Amy J. Wagner, MD[a], Michael E. Mitchell, MD[b], Aoy Tomita-Mitchell, PhD[b,*]

KEYWORDS

• Noninvasive prenatal testing • Screening • Cell-free DNA • Aneuploidy

KEY POINTS

- Noninvasive prenatal testing (NIPT) using cell-free fetal (cfDNA) offers potential as a screening tool for fetal anomalies; it is more accurate than maternal serum markers and nuchal translucency tests.
- The accuracy of NIPT using cfDNA, with a lower false-positive rate than previous standard aneuploidy testing, decreases the overall number of invasive tests needed for a definitive diagnosis, subjecting fewer pregnancies to the risk of the invasive procedures.
- Women who undergo NIPT need informed consent before testing and accurate, sensitive counseling after results are available.

INTRODUCTION

Prenatal screening for aneuploidy has been available to pregnant women for more than three decades. Accurate prenatal screening is important for several reasons. It can provide reassurance early in pregnancy in some cases. For those receiving less encouraging news, it allows the opportunity to consider options, have ample time to make difficult decisions, and manage expectations. It also may help to predict the postnatal course and make appropriate delivery plans when needed. An ideal prenatal test is one that is accurate, can be completed early in gestation, and poses minimal or

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[a] Division of Pediatric Surgery, Department of Surgery, Medical College of Wisconsin, 999 North 92nd Street, Suite C320, Milwaukee, WI 53226, USA; [b] Division of Cardiothoracic Surgery, Department of Surgery, Medical College of Wisconsin, 8701 Watertown Plank Road, Milwaukee, WI 53226, USA

* Corresponding author.

E-mail address: amitchell@mcw.edu

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no risk to the fetus and the mother. Over the past several years, the discovery of cell-free fetal DNA (cfDNA) in the maternal circulation has revolutionized prenatal screening and changed the standard of care.

Noninvasive maternal blood testing began in the early 1980s with the maternal serum test for α-fetoprotein (AFP). Since that time, many serum factors have been studied as screening tests for fetal anomalies. Maternal serum markers tested in the second trimester utilized the double (maternal serum beta-human chorionic gonadotropin [hCG] and AFP), triple (maternal serum beta-hCG, AFP, unconjugated estriol), and eventually quadruple (maternal serum beta-hCG, AFP, unconjugated estriol, and inhibin A) screens. The quadruple screen is associated with a false-positive rate of 7% and a sensitivity of less than 80%.[1] In 2007, the American College of Obstetricians and Gynecologists released guidelines that included nuchal translucency (a measurement of the thickness of the back of the fetal neck), serum pregnancy-associated plasma protein A (PAPP-A), and serum beta-hCG in the first trimester in addition to the quadruple screen in the second trimester. The nuchal translucency test has an overall sensitivity of 77% for trisomy 21 and a false-positive rate of 6%.[2] Combining the nuchal translucency and quad screens improves sensitivity, but there continues to be a 3% to 5% false-positive rate.[3]

The maternal serum markers AFP, beta-hCG, unconjugated estriol, and inhibin A (the quad screen) are now routinely utilized in screening pregnancies for trisomy 21. AFP is a major fetal plasma protein and has a structure similar to albumin that is found in postnatal life. AFP is made initially by the yolk sac, gastrointestinal tract, and liver. Fetal plasma levels peak at approximately 10 to 13 weeks gestation and then decline progressively until term, whereas maternal levels peak in the third trimester. Maternal and amniotic fluid levels of AFP are increased in pregnancies in which the fetus has a neural tube defect (ie, anencephaly and open spina bifida) or certain other fetal malformations, such as abdominal wall defects. Screening of maternal blood samples usually is done between weeks 16 and 18 of gestation.[4,5] Although neural tube defects have been associated with elevated levels of AFP, decreased levels have been associated with Down syndrome.

A complex glycoprotein, beta-hCG is produced exclusively by the outer layer of the trophoblast shortly after implantation in the uterine wall. It increases rapidly in the first 8 weeks of gestation, declines steadily until 20 weeks, and then plateaus. Unconjugated estriol is produced by the placenta from precursors provided by the fetal adrenal glands and liver. It increases steadily throughout pregnancy to a higher level than that normally produced by the liver. Unconjugated estriol levels are decreased in Down syndrome and trisomy 18. The last maternal serum marker that makes up the quadruple screen is inhibin-A. Inhibin A, which is secreted by the corpus luteum and fetoplacental unit, is also a maternal serum marker for fetal Down syndrome when levels are reduced.[6]

Another first trimester serum screening test for trisomy 21, or Down syndrome, is PAPP-A. PAPP-A, which is secreted by the placenta, has been shown to play an important role in promoting cell differentiation and proliferation in various body systems. The PAPP-A concentration increases with gestational age until term. Decreased PAPP-A levels in the first trimester (between 10 and 13 weeks) have been shown to be associated with Down syndrome. When used along with free β-hCG and ultrasound measurement of nuchal translucency, serum PAPP-A levels can reportedly detect 82% to 87% of affected pregnancies with a false-positive rate of approximately 5%.[7]

The limitations of these standard prenatal screening tools include high false-positive and false-negative rates. Additionally, the quadruple screen is drawn in the second