



Noninvasive Prenatal Testing to Determine Fetal Aneuploidies, Microdeletions, Single-Gene Disorders, and Twin Zygosity

Effective: May 1, 2025

Next Review: January 2026

Last Review: March 2025

IMPORTANT REMINDER

Medical Policies are developed to provide guidance for members and providers regarding coverage in accordance with contract terms. Benefit determinations are based in all cases on the applicable contract language. To the extent there may be any conflict between the Medical Policy and contract language, the contract language takes precedence.

PLEASE NOTE: Contracts exclude from coverage, among other things, services or procedures that are considered investigational or cosmetic. Providers may bill members for services or procedures that are considered investigational or cosmetic. Providers are encouraged to inform members before rendering such services that the members are likely to be financially responsible for the cost of these services.

DESCRIPTION

Fetal cell-free DNA fragments and fetal cells present in the plasma of pregnant women can be used for fetal screening, including testing for fetal sex chromosome aneuploidies (e.g., Turners, Klinefelter syndrome), fetal sex determination, twin zygosity, and microdeletion syndromes (e.g., Prader-Willi/Angelman syndrome).

MEDICAL POLICY CRITERIA

Note: This policy does not address reproductive carrier screening (see Cross References).

- I. Genetic testing of maternal plasma for fetal trisomies 13, 18, and 21 may be considered **medically necessary**.
- II. For *member contracts subject to Washington's State Board of Health Rule* (WAC 246-680), genetic testing of maternal plasma for fetal sex chromosome aneuploidies (e.g., sex chromosome aneuploidy (SCAs) or sex chromosome aneuploidy panel (SCAP) testing) may be considered **medically necessary**.
- III. For *all other member contracts*, genetic testing of maternal plasma for fetal sex chromosome aneuploidies (e.g., sex chromosome aneuploidy (SCAs) or sex chromosome aneuploidy panel (SCAP) testing) is considered **investigational**.

- IV. Genetic testing of maternal plasma for fetal sex determination is considered **not medically necessary**.
- V. Genetic testing of maternal plasma for fetal microdeletion syndromes, fetal single-gene disorders, and twin zygosity is considered **investigational**, including combination tests that include one or more of these components (see Policy Guidelines).

NOTE: A summary of the supporting rationale for the policy criteria is at the end of the policy.

POLICY GUIDELINES

COMBINATION TESTS

Combination tests that include investigational test components (such as microdeletion or single-gene testing) in addition to the standard trisomy testing include, but are not limited to the following tests:

- MaterniT® 21 PLUS + ESS (Labcorp)
- Panorama™ (Natera)
- Unity™ (BillionToOne)

TESTING RESULTS

Karyotyping would be necessary to exclude the possibility of a false-positive, nucleic acid sequencing– based test. Before testing, women should be counseled about the risk of a false-positive test. In a 2015 committee opinion, the American College of Obstetricians and Gynecologists recommended that all patients receive information on the risks and benefits of various methods of prenatal screening and diagnostic testing for fetal aneuploidies, including the option of no testing.

Studies published to date on noninvasive prenatal screening for fetal aneuploidies have reported rare but occasional false positives. False-positive findings have been found to be associated with factors including placental mosaicism, vanishing twins, and maternal malignancies. Diagnostic testing is necessary to confirm positive cell-free fetal DNA tests, and management decisions should not be based solely on the results of cell-free fetal DNA testing. The American College of Obstetricians and Gynecologists further recommended that patients with indeterminate or uninterpretable (i.e., “no call”) cell-free fetal DNA test results be referred for genetic counseling and offered ultrasound evaluation and diagnostic testing because “no call” findings have been associated with an increased risk of aneuploidy.

Cell-free fetal DNA screening does not assess risk of neural tube defects. Patients should continue to be offered ultrasound or maternal serum α -fetoprotein screening.

LIST OF INFORMATION NEEDED FOR REVIEW

It is critical that the list of information below is submitted for review to determine if the policy criteria are met. If any of these items are not submitted, it could impact our review and decision outcome.

- Name of the genetic test(s) or panel test

- Name of the performing laboratory and/or genetic testing organization (more than one may be listed)
- The analyses included in the test (e.g., trisomies, sex chromosome aneuploidies, etc.)
- Relevant billing codes
- Blood draw date
- Brief description of how the genetic test results will guide clinical decisions that would not otherwise be made in the absence testing
- Medical records related to this genetic test

CROSS REFERENCES

1. [Evaluating the Utility of Genetic Panels](#), Genetic Testing Policy No. 64
2. [Fetal RHD Genotyping Using Maternal Plasma](#), Genetic Testing No. 74
3. [Invasive Prenatal \(Fetal\) Diagnostic Testing Using Chromosomal Microarray Analysis \(CMA\)](#), Genetic Testing, Policy No. 78
4. [Genetic Testing for the Evaluation of Products of Conception and Pregnancy Loss](#), Genetic Testing, Policy No. 79
5. [Reproductive Carrier Screening for Genetic Diseases](#), Genetic Testing, Policy No. 81
6. [Maternal Serum Analysis for Risk of Adverse Obstetric Outcomes](#), Laboratory, Policy No. 75

BACKGROUND

Historically, karyotype testing was an optional test used to examine chromosomes in a sample of fetal cells to help identify genetic disorders. Karyotype testing is an invasive and requires either an amniocentesis or a chorionic villi sampling test (CVS). Newer non-invasive prenatal screening tests have been developed that analyzes fetal cell-free DNA (cfDNA) or fetal cells circulating in maternal blood. Most DNA is contained within cells, but a small amount circulates freely in the bloodstream, called cfDNA. This non-invasive prenatal screening test (NIPT) analyzes the maternal serum for fetal trisomy aneuploidies and can also include testing for fetal sex chromosomes aneuploidies, microdeletions, twin zygosity, and fetal sex determination.

FETAL TRISOMY ANEUPLOIDY TESTING

National guidelines recommend that all pregnant women be offered screening for fetal chromosomal abnormalities, the majority of which are aneuploidies (an abnormal number of chromosomes). Fetal chromosomal abnormalities occur in approximately 1 in 160 live births. The trisomy syndromes are aneuploidies involving three copies of one chromosome. Trisomies 21 (Down syndrome, T21), 18 (Edwards syndrome, T18) and 13 (Patau syndrome, T13) are the most common forms of fetal aneuploidy that survive to birth. The most important risk factor for Down syndrome is maternal age, with an approximate risk of 1/1500 in young women that increases to nearly 1/10 by age 48.^[1]

Standard aneuploidy screening involves combinations of maternal serum markers and fetal ultrasound done at various stages of pregnancy. The detection rate for various combinations of noninvasive testing ranges from 60% to 96% when the false-positive rate is set at 5%. When tests indicate a high risk of a trisomy syndrome, direct karyotyping of fetal tissue obtained by amniocentesis or CVS is required to confirm that T21 or another trisomy is present. Both amniocentesis and CVS are invasive procedures and have procedure-associated risks of fetal injury, fetal loss and infection. A new screening strategy that reduces unnecessary amniocentesis and CVS procedures or increases detection of T21, T18, and T13 could improve outcomes. Confirmation of positive noninvasive screening tests with amniocentesis or

CVS is recommended; with more accurate tests, fewer women would receive positive screening results.

SEX CHROMOSOME ANEUPLOIDY

Some of the NIPT prenatal tests also include testing for sex chromosome aneuploidies (SCAs) or sex chromosome aneuploidy panel (SCAP) testing. Abnormalities in the number of X or Y chromosomes result in the following syndromes:

- Turner syndrome (Monosomy X or 45, X)
- Klinefelter syndrome (47, XXY)
- Triple X syndrome (47, XXX)
- Jacob syndrome (47, XYY)
- XYY syndrome (48, XYY)

Sex chromosome aneuploidies occur in approximately 1 in 400 live births. These aneuploidies are typically diagnosed postnatally, sometimes not until adulthood, such as during an evaluation of diminished fertility. Alternatively, sex chromosome aneuploidies may be diagnosed incidentally during invasive karyotype testing of pregnant women at high risk for Down syndrome. Potential benefits of early identification (e.g., the opportunity for early management of the manifestations of the condition), must be balanced against potential harms that can include stigmatization.

MICRODELETION SYNDROMES

Microdeletion syndromes are defined as a group of clinically recognizable disorders characterized by a small (< 5Mb) deletion of a chromosomal segment spanning multiple disease genes, each potentially contributing to the phenotype independently. The phenotype is defined as the set of observable characteristics of an individual resulting from the interaction of its genotype with the environment. Microdeletion testing can include, but is not limited to the following conditions or syndromes:

- 22q deletion syndrome (DiGeorge)
- 22q11 deletion syndrome (Shprintzen syndrome)
- 15q11.2 (Prader-Willi/Angelman syndromes)
- 5p deletion (Cri du chat syndrome)
- 1p36 deletion syndrome
- 4p deletion (Wolf-Hirschhorn syndrome)

Clinical implications of prenatal testing for microdeletions are not well defined. It is unclear whether prenatal diagnosis is appropriate given the inherent difficulty in accurately predicting the [phenotype](#) for the myriad of microdeletion syndromes. Though laboratories may offer screening for microdeletion syndromes, screening for these microdeletion syndromes is not currently the main intent of NIPT screening tests.

SINGLE-GENE DISORDERS

Single-gene disorders (also known as monogenic disorders) are caused by a variation in a single gene. Individually, single-gene disorders are rare, but collectively are present in approximately 1% of births. The Vistara Single-Gene Disorder Test panel screens for 25 conditions that result from variants across 30 genes, which have a combined incidence of 1 in

600 (0.17%).^[2] These include Noonan syndrome and other Noonan spectrum disorders, skeletal disorders (e.g., osteogenesis imperfecta, achondroplasia), craniosynostosis syndromes, Cornelia de Lange syndrome, Alagille syndrome, tuberous sclerosis, epileptic encephalopathy, *SYNGAP1*-related intellectual disability, CHARGE syndrome, Sotos syndrome, and Rett syndrome. The clinical presentation and severity of these disorders can vary widely. Some, but not all, can be detected by prenatal ultrasound examination.

FETAL SEX DETERMINATION

Sequencing-based testing of maternal serum for determination of fetal sex in the first trimester of pregnancy is possible. However, the current standard of care for fetal sex is ultrasound. Fetal sex includes:

- Male (XX)
- Female (XY)

TWIN ZYGOSITY TESTING

Twin gestations occur in approximately 1 in 30 live births in the United States and have a 4- to 10-fold increased risk of perinatal complications.^[3] Dizygotic or "fraternal" twins occur from ovulation and fertilization of two oocytes, which results in dichorionic placentation and two separate placentas. In contrast to dichorionic twins, monochorionic twin pregnancies share their blood supply. Monochorionic twins account for about 20% of twin gestations and are at higher risk of structural defects, miscarriage, preterm delivery, and selective fetal growth restriction compared to dichorionic twins.^[3] Up to 15% of monochorionic twin pregnancies are affected by twin-to-twin transfusion syndrome (TTTS), a condition characterized by relative hypovolemia of one twin and hypervolemia of the other.^[4] According to estimates from live births, TTTS occurs in up to 15% of monochorionic twin pregnancies. In these twin pregnancies, serial fetal ultrasound examinations are necessary to monitor for development of TTTS as well as selective intrauterine growth restriction because these disorders have high morbidity and mortality and are amenable to interventions that can improve outcomes.^[4] NIPT using cell-free fetal DNA to determine zygosity in twin pregnancies could potentially inform decisions about early surveillance for TTTS and other monochorionic twin-related abnormalities. In particular, determining zygosity with NIPT could potentially assist in the assessment of chorionicity when ultrasound findings are not clear.

REGULATORY STATUS

None of the commercially available sequencing assays listed above have been submitted to or reviewed by the U.S. Food and Drug Administration (FDA). Clinical laboratories may develop and validate tests in-house and market them as a laboratory service. Laboratory-developed tests (LDTs) must meet the general regulatory standards of the Clinical Laboratory Improvement Act (CLIA). Laboratories offering LDTs must be licensed by CLIA for high-complexity testing. The NIPT panels vary significantly in the base components and additional options a provider may choose on the requisition form. Commercial tests include, but are not limited to, the following:

- **Harmony™** Prenatal Test (Ariosa Diagnostics, now Roche).
Tests for fetal trisomies.

Additional options for testing fetal sex chromosome aneuploidies, fetal sex, monosomy X, and 22q11.2 microdeletion.

- **InformaSeqSM Prenatal Test** (Integrated Genetics, a division of LabCorp)
Tests for fetal trisomies.
Optional testing includes fetal sex chromosome and fetal sex.
- **MaterniT Genome** (Sequenom Laboratories, now LabCorp)
Tests for genome wide aneuploidies
- **MaterniT21TM Plus** (Sequenom Laboratories, now LabCorp).
Tests for fetal trisomies and fetal sex.
Additional items that may be added include testing for microdeletions, other chromosomes (T16, T22), and sex chromosomes aneuploidies.
- **PanoramaTM** (Natera).
Tests for fetal trisomies, fetal sex chromosome aneuploidies, triploidy, microdeletions, and fetal sex.
- **PrequelTM Prenatal Screen** (Myriad)
Tests for fetal trisomies, with options for sex chromosome and microdeletion testing.
- **Progenity Innatal[®] Prenatal Screen** (Progenity)
Tests for fetal trisomies, may include fetal sex chromosome aneuploidies and fetal sex.
- **UnityTM (BillionToOne)**
Tests for fetal trisomies, sex chromosome aneuploidy, fetal sex (optional), fetal RhD status (optional), as well as maternal carrier screening for cystic fibrosis, spinal muscular atrophy, sickle cell disease, alpha and beta-thalassemia, and fragile x syndrome (optional). Fetal screening via single-gene non-invasive prenatal testing is done reflexively for identified maternal carriers.
- **Verifi[®] Prenatal Test** (Illumina, formerly Verinata Health).
There are two options for these tests which may include fetal trisomies, fetal sex chromosomes aneuploidies, microdeletions, and fetal sex.
- **VistaraTM Single Gene NIPT** (Natera)
Tests for 25 autosomal dominant and X-linked conditions across 30 genes.

EVIDENCE SUMMARY

Human Genome Variation Society (HGVS) nomenclature^[5] is used to describe variants found in DNA and serves as an international standard. It is being implemented for genetic testing medical evidence review updates starting in 2017. According to this nomenclature, the term “variant” is used to describe a change in a DNA or protein sequence, replacing previously-used terms, such as “mutation.” Pathogenic variants are variants associated with disease,

while benign variants are not. The majority of genetic changes have unknown effects on human health, and these are referred to as variants of uncertain significance.

Assessment of a diagnostic technology such as maternal plasma DNA sequencing tests typically focuses on three parameters:

1. Analytic validity;
2. Clinical validity (includes calculations of sensitivity and specificity in appropriate populations of patients); and
3. Clinical utility (demonstration that the diagnostic information can be used to improve patient health outcomes).

The focus of this evidence summary below is on the clinical validity and utility of these tests.

The evidence regarding these three questions was addressed in the 2012 and 2014 BlueCross BlueShield Association Technology Evaluation Center (TEC) Assessments.^[6, 7] The initial Assessment, published in 2012, focused on detection of T21/Down syndrome because the majority of published data at the time was concentrated on this trisomy. Additionally, large numbers of cases were included in several publications, and all companies had published data regarding the detection of T21. The subsequent Assessment, published in 2014, reviewed the available data for detection of T18, T13, and sex chromosome aneuploidies (SCAs). The scope of both TEC Assessments was limited to the evaluation of tests that are available in the United States. Additional literature published after publication the TEC Assessments is also addressed in the analysis below.

CLINICAL VALIDITY

Multiple Conditions

Gil (2017) published a systematic review with meta-analysis which evaluated the performance of screening for fetal trisomies 21, 18 and 13 and sex chromosome aneuploidies.^[8] This summary will only focus on the results for sex chromosome aneuploidies. There were 36 total cases of monosomy X and 7,677 unaffected singleton pregnancies. The pooled weighted detection rate and false positive rate were 95.8% (95% confidence interval [CI] 70.3 to 99.5%) and 0.14% (95% CI 0.05 to 0.38%), respectively. Also, there were 17 cases of sex chromosome abnormalities that were not monosomy X and 5,383 unaffected singleton pregnancies. The pooled weighted detection rate and false positive rate were 100% (95% CI 83.6 to 100%) and 0.003% (95% CI 0 to 0.07%), respectively. The authors concluded that the number of cases for sex chromosome aneuploidy was too small to calculate overall screening performance.

Norton (2016) conducted a high-quality systematic review and meta-analysis which evaluated cohort studies comparing sequential screening to cell free DNA detection rates for fetal chromosomal abnormalities.^[9] A total of 452,901 women underwent sequential screening and out of those women, 2575 (0.57%) had a fetal chromosomal abnormality. Of those abnormalities, the detection rate was 81.6% (total of 2,101). Additionally, 19,929 euploid fetuses had positive sequential screening resulting in a detection rate of 4.5%. The authors concluded that cfDNA testing has good performance for fetal sex and the detection rate of sequential screening for all aneuploidies was significantly greater than cfDNA ($p < 0.0001$).

Mackie (2016) conducted a systematic review with meta-analysis evaluating the performance of cell free fetal DNA testing for all conditions (singleton pregnancies only).^[10] A total of 117

studies addressing 18 conditions were included. The meta-analysis showed that for fetal sex (60 studies with 11,179 tests), the sensitivity and specificity were 0.989 (95% CI 0.980 to 0.994) and 0.996 (95% CI 0.989 to 0.998), respectively. For monosomy X (80 studies and 6,712 tests), the sensitivity was 0.929 (95% CI 0.741 to 0.984) and specificity 0.999 (95% CI 0.995 to 0.999). The authors concluded that fetal sex can be considered diagnostic but that testing for aneuploidies should only be considered as screening.

Fetal Sex Chromosome Aneuploidies

A Cochrane review by Badeau (2017) evaluated diagnostic accuracy of NIPT for sex chromosome anomalies.^[11] Twelve studies were identified on the 45,X chromosome with sensitivities of 91.7% to 92.4% and specificities of 99.6% to 99.8%. Reviewers calculated that of 100,000 pregnancies, 1,039 would be affected by 45,X. Of these, 953 tested with massively parallel shotgun sequencing and 960 tested with targeted massively parallel sequencing would be detected and 86 and 79 cases, respectively, would be missed. Of the 98,961 unaffected women, 396 and 198 pregnant women would undergo an unnecessary invasive test. The authors were unable to perform meta-analyses of NIPT for chromosomes 47,XXX, 47,XXY, and 47,XYY due to insufficient evidence.

A systematic review published after the Cochrane review had similar results, showing high sensitivity (94.1%, 95% CI 90.8% to 96.3%) and specificity (94.1%, 95% CI 90.8% to 96.3%), but more false positives (235 per 100,000) than tests for the common trisomies.^[12] Subgroup analyses showed variation in positive predictive value (PPV) by type of sex chromosome aneuploidy, from 32% (95% CI 27.0% to 37.4%) for monosomy X to 70% (95% CI 63.9% to 77.1%) for XYY syndrome, explained by higher sensitivity and specificity for the Y chromosome and high risk of false-positive results for aneuploidies involving the X chromosome only.

Gil (2015) published results from a systematic review and meta-analysis that examined the analysis of cfDNA in maternal blood in screening for fetal aneuploidies between January 2011 and January 2015.^[13] Thirty-seven articles were included in the review; however, just 28 of these studies reported on sex chromosome aneuploidy testing. Sixteen of the 28 studies addressed the detection of monosomy X (Turner syndrome). The authors found, that of the 177 singleton pregnancies with fetal monosomy X, the detection rate varied between 66.7% and 100% and the false-positive rate varied between 0% and 0.52%. The pooled weighted detection rate was 90.3% (95% CI 85.7 to 94.2%), and the false-positive rate was 0.23% (95% CI 0.14 to 0.34%). The remaining 12 studies reported on the performance of sex chromosome abnormalities other than monosomy X (i.e., 47XXX, 47XXY, 47XYY), in a combined total of 56 affected and 6,699 non-sex chromosome aneuploidy singleton pregnancies. The pooled detection rate was 93.0% (95% CI 85.8 to 97.8% and the false-positive rate was 0.14% (95% CI 0.06 to 0.24%). This study has significant methodological limitations, which include but are not limited to, very small sample sizes, high risk of bias in relation to flow and timing (i.e., consecutive cases), testing performed in selected populations, and a lack of clarity about karyotyping, and the studies did not clearly define the patient's risk category.

The 2014 BCBSA TEC Assessment included a meta-analysis of sequencing-based studies published through April 15, 2014 that included a report on sex chromosome anomalies.^[7] The largest number of studies (14 studies, total of 152 cases) published on sex chromosome aneuploidies addressed detection of monosomy X. Pooled sensitivity for detecting monosomy X was 83% (95% CI 74% to 90%) and pooled specificity was 100% (95% CI 100% to 100%).

In addition, 11 studies with a total of 51 cases were identified on the performance of sequencing-based tests in identifying other sex chromosome anomalies. Pooled sensitivity was 89% (95% CI 50% to 98%) and pooled specificity was 100% (100% to 100%). The meta-analysis of studies on sex chromosome aneuploidies did not differentiate between high and low-risk populations.

Microdeletion Syndromes

In a systematic review of NIPT using cfDNA in general risk pregnancies conducted for ACMG, Rose (2022) included 17 studies of screening for copy number variants (microdeletions and microduplications).^[14] Meta-analyses were not conducted due to study heterogeneity. Although screening identified a small number of copy number variants (CNVs), confirmatory testing was frequently unavailable and complete ascertainment of cases was lacking. Sample sizes in each study were relatively small and sensitivities varied greatly. Additionally, it was often difficult to distinguish between low- and high-risk cohort in individual studies. The study authors concluded that the performance of NIPT was significantly poorer when targeting CNVs than the common trisomies and additional outcome studies are needed to understand the unique clinical value of NIPT for CNVs when compared with other approaches.

Zaninović (2022) conducted a systematic review of NIPT for CNVs and microdeletions.^[15] A total of 32 studies were identified with literature searches conducted through February 2022. Of these, 21 studies concerned screening for microdeletion syndromes. Meta-analyses were not conducted due to study heterogeneity. Although a comprehensive quality assessment of studies was not conducted, the study authors described notable limitations of the included studies. Most studies did not define indications for screening, and some included only high-risk pregnancies. Negative predictive values could not be determined because none of the studies performed systematic confirmatory analysis by chromosomal microarray analysis for negative/low-risk cases, mostly relying on clinical follow-up. The study authors concluded that given the limited follow-up and validation data available, NIPT for microdeletions and CNVs should be used with caution.

Familiari (2021) conducted a systematic review of the literature on screening for fetal microdeletions and microduplications using cfDNA.^[16] A total of seven studies met inclusion criteria, representing 210 cases of microdeletions or microduplications. The overall pooled PPV was 44.1% (95% CI 31.49 to 63.07, range 28.9% to 90.6%). Limitations in the individual studies included retrospective design, low number of cases for each condition, lack of a standardized confirmation of the disease, low detail regarding the presence or absence of ultrasound anomalies and sonographic protocol used, different gestational ages at the time of the test, and variation in background risk. The authors noted that confirmatory testing was seldom reported in studies, under the assumption that all anomalies would have been identified in the newborn by physical exam. However, because many newborns with microdeletion and microduplication syndromes will not demonstrate phenotypical anomalies, standard neonatal examination cannot be considered a reliable ascertainment method and the detection rate and negative predictive value could not be determined from this body of evidence.

Additional non-randomized studies from companies offering microdeletion testing have been published evaluating data from clinical samples submitted for screening. Dar (2022) conducted a prospective analysis of 20,887 women who underwent NIPT testing at 21 centers in six countries.^[17] A genetic outcome result was available for 18,289 women (87.6%), and 12 cases

of 22q11.2 deletion syndrome were confirmed in the cohort. Limitations of the study include the low number of overall confirmed cases, wide confidence intervals for sensitivity, positive and false positive values, and varied indications for testing.

Soster (2021) conducted a retrospective analysis of 55,517 samples submitted for genome-wide cfDNA screening at a commercial laboratory between 2015 and 2018.^[18] [\\pdxnas01\DataPdx1\Saturn\Groups\MedPol\1. Policy Work\Genetic Testing\gt44\Policy drafts\2022 01\ blank](#) Diagnostic testing results were available in 42.5% (n=1,142) of screen-positive samples, and 0.82% of screen-negative samples, with overall 2.98% of samples with diagnostic outcomes. Microdeletion syndromes included 1p36 deletion, Wolf–Hirschhorn, Cri-du-chat, Langer–Giedion, Jacobsen, Prader–Willi, Angelman, and DiGeorge syndrome. Test performance characteristics were based on the 1,569 patients who had diagnostic testing performed, and an overall PPV of 72.6% was reported.

Wang (2021) conducted a prospective analysis of 39,002 pregnant women who received NIPT in a single center between 2018 and 2020.^[19] There were 473 (1.21%) pregnancies that tested positive for fetal chromosome abnormalities, of which 95 were microdeletion/microduplication syndrome cases. Limitations of this study include variable types of diagnostic testing and specimen types, a large number of patients who refused to receive a prenatal diagnosis (n=135) and then were lost to follow-up (n=128), and low percentage of overall specimens that had diagnostic testing results available.

Fetal Sex Determination

The current standard of care for fetal sex determination is ultrasound.

Three reviews report on the use of cfDNA for fetal sex determination. Davaney (2011) published results from a systematic review and meta-analysis to determine if noninvasive prenatal determination of fetal sex using cfDNA provides an alternative to invasive techniques for some heritable disorders.^[20] From 57 selected studies, 80 data sets (representing 3524 male-bearing pregnancies and 3,017 female-bearing pregnancies) were analyzed. Authors reported that despite inter-study variability, performance was high using maternal blood. Sensitivity and specificity for detection of Y chromosome sequences was greatest using RT-qPCR after 20 weeks' gestation. Tests using urine and tests performed before seven weeks' gestation were unreliable.

Wright (2012) published results from a review and meta-analysis of the published literature to evaluate the use of cfDNA for prenatal determination of fetal sex.^[21] The authors reviewed 90 studies, incorporating 9,965 pregnancies and 10,587 fetal sex results. Overall mean sensitivity was 96.6% (95% CI 95.2% to 97.7%) and mean specificity was 98.9% (95% CI 98.1% to 99.4%). The authors identified one limitation of their study as the inability to properly evaluate the proportion of inconclusive or uncertain results, which is known to be problematic with this technique and may vary with gestational age. Further, literature-based reviews are at risk of publication bias due to the suppression of unwanted findings. The authors concluded that fetal sex can be determined with a high level of accuracy by analyzing cfDNA.

Colmant (2013) published a review of the published literature evaluating the use of cfDNA and ultrasound for prenatal determination of fetal sex during the first trimester of pregnancy.^[22] The authors identified 16 reports of the determination of fetal sex in maternal blood and 13 reports of the determination by ultrasound. Authors determined a sensitivity and specificity of nearly 100% from eight weeks of gestation for cfDNA and from 13 weeks of gestation for ultrasound

respectively. Authors concluded that fetal sex can be determined with a high level of accuracy by analyzing cfDNA and at an earlier gestation than ultrasound.

Twin Zygosity

Norwitz (2019) conducted a validation study of a single-nucleotide polymorphism-based NIPT in twin pregnancies.^[3] The study included 95 samples with confirmed zygosity: 30 monozygotic and 65 dizygotic. Two of the 95 samples did not receive results due to low fetal fraction. Among the 93 pregnancies that yielded results, monozygotic sensitivity was 100% (29/29) and monozygotic specificity was 100% (64/64). A major limitation of this study was a lack of information on timing of the index test and the use of different methods to confirm zygosity.

Single-Gene Disorders

Vistara™

The performance characteristics of the Vistara™ NIPT were evaluated in a retrospective validation study conducted by Zhang (2019).^[23] Most of the study participants were high risk due to prenatal ultrasound findings or a family history of genetic disease. The validation cohort included 76 cases (3 positive and 73 negative) and the clinical study included 422 samples (32 positive and 390 negative). Pathogenic or likely pathogenic variants were confirmed using a secondary NGS assay. Sanger sequencing was used to confirm positive findings if an invasive specimen (e.g., amniotic fluid) or a postnatal sample was available. Of the 35 positive results, 20 had a confirmed diagnosis. Pregnancy outcome data were obtained for 26 of 35 (74.2%) positive tests and 198 of 463 (42.7%) negative tests from both the validation and clinical studies.

Mohan (2022) reported on the clinical experience with Vistara™ in a series of 2,208 pregnancies.^[2] Of 2,416 initial tests, 132 (5.5%) tests were ineligible and 76 (3.1%) did not pass quality control. Indications for NIPT included family history (6.0%), abnormal ultrasound finding (23.3%), advanced paternal age (41.3%), and unspecified/other/advanced maternal age (29.4%). In cases without abnormal ultrasound findings or family history, the test positive rate was 6 of 52 (0.4%) (6/52). Positive variants were confirmed by a secondary NGS assay using deeper sequencing, and variants of unknown significance were not reported. Confirmatory prenatal or postnatal diagnostic testing was recommended for all screen-positive patients. Overall, the test positive rate was 125 of 2,208 (5.7%), and of these, follow-up information was available for 67 (53.6%), with none classified as false positive. Positive tests in cases without abnormal ultrasound findings or family history were found for 6/52 (0.4%).

Major limitations of these studies include a lack of confirmatory testing and selection bias. Because of missing data, it is not possible to determine accurate estimates of true positive and true negative tests. In addition, a large proportion of participants in both studies had a previous screening with findings suggestive of a potential disorder. It is unclear if single-gene NIPT is intended to be an adjunct to or replacement for other screening tests such as ultrasound. More clarity on the proposed use of the testing would be needed to adequately evaluate performance characteristics.

UNITY™

Westin (2022) published a retrospective clinical validation study of the UNITY™ single-gene NIPT for 77 pregnant women who had previously been identified as beta hemoglobinopathy (*HBB*) carriers.^[24] Single-gene NIPT was performed from October 2018 to December 2019 and

returned a fetal beta hemoglobinopathy genotype prediction for 68 of the 77 pregnancies, with nine undetermined (11.7%). The UNITY screen accurately distinguished heterozygous from homozygous fetuses with 100% sensitivity (95% CI 90.8% to 100%) and 96.5% specificity (95% CI 82.2% to 99.9%) compared to confirmatory newborn chart review or genotyping of umbilical cord blood. The predicted fetal genotype concurred with the newborn genotype in 67 out of 68 pregnancies (98.5%). Using single-gene NIPT data and a priori risk adjustments, residual risk could classify fetuses as 'low risk,' 'decreased risk,' or 'high risk' in 75 of 77 pregnancies with a 2.6% no-call rate. Two fetuses affected with sickle cell disease were correctly classified as high risk (>9 in 10 residual disease risk), and one fetus, which had a previously undetermined homozygosity score, was also affected and has an elevated residual risk score of 1 in 20.

The performance characteristics of the UNITY™ test were evaluated in a clinical validation study conducted by Hoskovec (2023).^[25] [\\slcnas10\datapdx7\groups\1. Policy Work\Genetic Testing\gt44\Policy drafts\2025 01\ blank](#) The study participants comprised a general population not at high risk for cystic fibrosis, hemoglobinopathies, or spinal muscular atrophy, who were screened with UNITY™ from August 2019 to May 2021. All pregnancies were at least 10 weeks gestation, were singleton pregnancies, and were not conceived with a donor egg or gestational carrier. The cohort included 9,151 pregnancies seen by 240 providers. A total of 1,669 (18.2%) were found to be heterozygous carriers for a pathogenic variant of at least one condition (4.47% were heterozygous for a *CFTR* pathogenic variant, 4.64% for an *HBB* variant, 8.65% for *HBA1/HBA2* variant, and 2.26% for *SMN1*) and underwent reflex single-gene NIPT. Newborn outcomes data was available for 201 (12%) pregnancies with an identified positive maternal carrier, and of these, 10 (4.9%) had no call single-gene NIPT results and were excluded from the analysis. Single-gene NIPT identified 14 out of 15 affected fetuses as 'high risk' for one of the screened conditions on the panel, which resulted in a sensitivity of 93.3% (95% CI 68.1% to 99.8%), a PPV of 48.3% (95% CI 36.1% to 60.1%) and NPV of 99.4% (95% CI 96% to 99.9%). Newborn outcomes by proprietary personalized fetal risk score across all screened conditions showed that four out of four (100%) pregnancies with >9 in 10 risk were affected, 8 out of 17 (47%) with risks between one in two and two in three risk were affected, two out of eight (25%) with risks between 1 in 10 and 1 in 100 were affected, and one out of 162 (0.6%) with risks <1 in 100 were affected. The authors also modeled the end-to-end clinical analytics of carrier screening with UNITY™ versus standard NGS carrier screening. The authors reported that in a real-world scenario accounting for the sensitivity of carrier screening and single-gene NIPT, the end-to-end sensitivity of carrier screening with UNITY™ was 90% (95% CI 71.8% to 98.9%), which was higher than that for conventional carrier screening.

Wynn (2023) evaluated the UNITY™ NIPT in a general population of 42,067 pregnant individuals who underwent UNITY™ carrier screening.^[26] A total of 7,538 (17.92%) carriers were identified and underwent reflex single-gene NIPT. Only 3,299 were able to be contacted for follow-up. The outcomes cohort consisted of 528 neonates and fetuses who were able to be assessed for single-gene disorders across 253 centers in the U.S. The authors calculated that in this cohort, the sensitivity of the UNITY™ test was 96.0% (95% CI 79.65% to 99.90%), with a specificity of 95.2% (95% CI 92.98% to 96.92%), PPV of 50.0% (95% CI 35.23% to 64.77%), and an NPV of 99.8% (95% CI 98.84% to 99.99%). Single-gene NIPT identified 9 of 10 pregnancies affected by cystic fibrosis, 11 of 11 affected by *HBB*, four of four affected by spinal muscular atrophy, and none affected by *HBA* as high risk. The authors also modeled the performance characteristics of maternal carrier screening followed by single-gene NIPT with the UNITY™. They found an end-to-end sensitivity of 92.4% with a specificity of 99.9% and

PPV and NPV values of 50.7% and 99.9%, respectively of the full cohort of 42,067 pregnancies; this was higher than conventional carrier screening and would result in a greater number of fetuses being characterized as high risk.

Major limitations included missing data, a lack of consistent confirmatory testing methods, and selection bias. Because of missing data, it is not possible to determine accurate estimates of true positive and true negative tests. Three studies examined testing for single-gene disorders with UNITY™; sensitivity and specificity across these studies was high and few samples resulted in a no-call result. The available studies on clinical validity have limitations, and the added benefit of UNITY™ test compared with current approaches is unclear.

CLINICAL UTILITY

Fetal Sex Chromosome Aneuploidies

The impact of screening for sex chromosome aneuploidies has not been modeled in published studies. Fetal sex chromosome aneuploidies were not included in the decision analysis of the 2014 BCBSA TEC Assessment because the implications of a screen-positive finding and diagnostic confirmation were considered to differ significantly when compared to T13 and T18.^[7] Finally, fetal sex aneuploidies are generally diagnosed postnatally in association with specific health problems, such as delayed puberty, or diminished fertility or infertility. Therefore, the balance of benefits and harms of cfDNA prenatal screen and subsequent diagnosis of sex chromosome fetal aneuploidies, each of which has variable and uncertain prognosis, is unclear.

Microdeletion syndromes

The clinical utility of testing for any specific microdeletion or any panel of microdeletions is uncertain.

There is a potential that prenatal identification of individuals with microdeletion syndromes could improve health outcomes due to the ability to allow for informed reproductive decision making, and/or to initiate earlier treatment; however, data demonstrating improvement are unavailable. Given the variability of expressivity of microdeletion syndromes and the lack of experience with routine genetic screening for microdeletions, clinical decision making based on genetic test results is not well defined. It is not clear what follow-up testing or treatments might be indicated for screen-detected individuals. Routine prenatal screening may identify a small percentage of fetuses with microdeletion variants earlier in pregnancy than would otherwise have occurred (e.g., by ultrasound evaluation and diagnostic testing). At the same time, routine prenatal screening for microdeletions would also result in false-positive tests and a larger number of invasive confirmatory tests. The large number of confirmatory tests could lead to a net harm because of pregnancy loss.

Most treatment decisions would be made after birth, and it is unclear whether testing in utero will lead to earlier detection and treatment of clinical disease after birth. Moreover, clinical decision making when a maternal microdeletion is detected in a pregnant woman without previous knowledge of a genetic variant is unclear.

Single-Gene Disorders

No studies were identified that evaluated whether cfDNA testing for single-gene disorders improves outcomes compared with standard care. There is a potential that prenatal

identification of pregnancies with single-gene disorders could improve health outcomes due to the ability to allow for informed reproductive decision making and/or initiate earlier treatment; however, data demonstrating improvement are unavailable. Given the variability of single-gene disorders identified by this testing and the lack of experience with routine genetic screening for single-gene disorders, clinical decision-making based on this testing is not well defined.

Twin Zygosity

No studies were identified that evaluated whether cfDNA testing for twin zygosity improves outcomes compared with standard care.

PRACTICE GUIDELINE SUMMARY

AMERICAN COLLEGE OF OBSTETRICIANS AND GYNECOLOGISTS AND SOCIETY FOR MATERNAL-FETAL MEDICINE (ACOG/SMFM)

Noninvasive Prenatal Screening for Fetal Aneuploidies

In 2020, ACOG and SMFM released a practice bulletin summary (No. 226) on screening for fetal aneuploidy.^[27] The following recommendations are based on “good and consistent” scientific evidence:

- “Cell-free DNA is the most sensitive and specific screening test for the common fetal aneuploidies. Nevertheless, it has the potential for false-positive and false-negative results. Furthermore, cell-free DNA testing is not equivalent to diagnostic testing.”
- “Patients with a positive screening test result for fetal aneuploidy should undergo genetic counseling and a comprehensive ultrasound evaluation with an opportunity for diagnostic testing to confirm results.”
- “Patients with a negative screening test result should be made aware that this substantially decreases their risk of the targeted aneuploidy but does not ensure that the fetus is unaffected. The potential for a fetus to be affected by genetic disorders that are not evaluated by the screening or diagnostic test should also be reviewed. Even if patients have a negative screening test result, they may choose diagnostic testing later in pregnancy, particularly if additional findings become evident such as fetal anomalies identified on ultrasound examination.”
- “Patients whose cell-free DNA screening test results are not reported by the laboratory or are uninterpretable (a no-call test result) should be informed that test failure is associated with an increased risk of aneuploidy, receive further genetic counseling and be offered comprehensive ultrasound evaluation and diagnostic testing.”

The following recommendations are based on “limited or inconsistent” scientific evidence:

- “The use of cell-free DNA screening as follow-up for patients with a screen positive serum analyte screening test result is an option for patients who want to avoid a diagnostic test. However, patients should be informed that this approach may delay definitive diagnosis and will fail to identify some fetuses with chromosomal abnormalities.” “No method of aneuploidy screening is as accurate in twin gestations as it is in singleton pregnancies; this information should be incorporated into pretest counseling for patients with multiple gestations.”

- “Cell-free DNA screening can be performed in twin pregnancies. Overall, performance of screening for trisomy 21 by cell-free DNA in twin pregnancies is encouraging, but the total number of reported affected cases is small. Given the small number of affected cases it is difficult to determine an accurate detection rate for trisomy 18 and 13.”

The following recommendations are based primarily on based “primarily on consensus and expert opinion:

- “In multifetal gestations, if a fetal demise, vanishing twin, or anomaly is identified in one fetus, there is a significant risk of an inaccurate test result if serum-based aneuploidy screening or cell-free DNA is used. This information should be reviewed with the patient and diagnostic testing should be offered.
- “Patients with unusual or multiple aneuploidies detected by cell-free DNA should be referred for genetic counseling and maternal–fetal medicine consultation.”

Cell-free DNA Screening for Single-Gene Disorders

In a practice advisory on cell-free DNA screening for single-gene disorders published in 2019 and reaffirmed in 2021,^[28] ACOG stated, “Although this technology is available clinically and marketed as a single-gene disorder prenatal screening option for obstetric care providers to consider in their practice, often in presence of advanced paternal age, there has not been sufficient data to provide information regarding accuracy and positive and negative predictive value in the general population. For this reason, single-gene cell-free DNA screening is not currently recommended in pregnancy.”

AMERICAN COLLEGE OF MEDICAL GENETICS AND GENOMICS

In 2023, the American College of Medical Genetics and Genomics (ACMG) published a position statement on noninvasive prenatal screening (NIPS) for fetal aneuploidy for fetal chromosome abnormalities in a general-risk population.^[29] Relevant recommendations are as follows:

- ACMG recommends NPS over traditional screening methods for all pregnant patients with singleton gestation for fetal trisomies 21, 18, and 13 (strong recommendation based on high certainty of evidence)
- ACMG recommends NIPS over traditional methods for trisomy screening in twin gestations (strong recommendation, based on high certainty of evidence)
- ACMG recommends that NIPS be offered to patients with a singleton gestation to screen for fetal SCA (strong recommendation, based on high certainty of evidence)
- ACMG suggests that NIPS for 22q11.2 deletion syndrome be offered to all patients (conditional recommendation, based on moderate certainty of the evidence)
- At this time, there is insufficient evidence to recommend routine screening for CNVs other than 22q11.2 deletions (no recommendation, owing to lack of clinically relevant evidence and validation)

- At this time, there is insufficient evidence to recommend or not recommend NIPS for the identification of RATS [rare autosomal trisomies] (no recommendation, owing to lack of clinically relevant evidence)

SUMMARY

FOR MEMBER CONTRACTS SUBJECT TO WASHINGTON'S STATE BOARD OF HEALTH RULE (WAC 246-680)

For member contracts subject to Washington's State Board of Health Rule, criteria for sex chromosome aneuploidy testing are based on the Rule. Therefore, for member contracts subject to Washington's State Board of Health Rule (WAC 246-680), sex chromosome aneuploidy testing using cell-free DNA may be considered medically necessary.

FOR MEMBER CONTRACTS NOT SUBJECT TO WASHINGTON'S STATE BOARD OF HEALTH RULE (WAC 246-680)

There is not enough research to show an improvement in health outcomes for non-invasive screening using fetal DNA to detect fetal sex chromosome aneuploidies. The current research shows mixed results for detection of abnormalities, including high false-positive rates. Therefore, non-invasive prenatal testing (NIPT) for fetal sex chromosome aneuploidies is considered investigational.

FOR ALL MEMBER CONTRACTS

Testing for Fetal Trisomies 13, 18, and 21

There is enough research to show that non-invasive prenatal testing (NIPT) for fetal trisomies 13, 18, and 21 are important for informing patient management and reproductive decision making. This testing is recommended by evidence-based clinical practice guidelines. Therefore, NIPT testing for trisomies 13, 18, and 21 may be considered medically necessary.

Fetal Sex Determination Testing

Research does not show that the use of nucleic acid sequencing-based testing for fetal sex determination is more beneficial than fetal ultrasound, which is the current clinical standard for determining fetal sex. Therefore, non-invasive prenatal testing (NIPT) for fetal sex determination is considered not medically necessary.

Microdeletion, Single-gene Disorder, and Twin Zygosity Testing

There is not enough research to show an improvement in health outcomes for non-invasive screening using fetal DNA to detect fetal microdeletion syndromes, fetal single-gene disorders, or twin zygosity. In addition, there are no evidence-based practice guidelines that recommend these types of testing. Therefore, non-invasive prenatal testing (NIPT) for fetal microdeletion syndromes, fetal single-gene disorders, or twin zygosity is considered investigational. This includes combination tests such as the Panorama™ and Unity™ tests that include one or more investigational components.

REFERENCES

1. Nicolaides KH, Syngelaki A, Gil M, et al. Validation of targeted sequencing of single-nucleotide polymorphisms for non-invasive prenatal detection of aneuploidy of chromosomes 13, 18, 21, X, and Y. *Prenatal diagnosis*. 2013;33(6):575-9. PMID: 23613152
2. Mohan P, Lemoine J, Trotter C, et al. Clinical experience with non-invasive prenatal screening for single-gene disorders. *Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology*. 2022;59(1):33-39. PMID: 34358384
3. Norwitz ER, McNeill G, Kalyan A, et al. Validation of a Single-Nucleotide Polymorphism-Based Non-Invasive Prenatal Test in Twin Gestations: Determination of Zygosity, Individual Fetal Sex, and Fetal Aneuploidy. *J Clin Med*. 2019;8(7). PMID: 31261782
4. UptoDate (2022).Twin-twin transfusion syndrome: Screening, prevalence, pathophysiology, and diagnosis. [cited 2/26/2025]. 'Available from:' <https://www.uptodate.com/contents/twin-twin-transfusion-syndrome-and-twin-anemia-polycythemia-sequence-screening-prevalence-pathophysiology-and-diagnosis>.
5. den Dunnen JT, Dalgleish R, Maglott DR, et al. HGVS Recommendations for the Description of Sequence Variants: 2016 Update. *Human mutation*. 2016;37(6):564-9. PMID: 26931183
6. TEC Assessment 2012. "Sequencing-based Tests to Determine Fetal Down Syndrome (Trisomy 21) from Maternal Plasma DNA " BlueCross BlueShield Association Technology Evaluation Center, Vol. 27 No. 10.
7. TEC Assessment 2014. "Noninvasive maternal plasma sequencing-based screening for fetal aneuploidies other than trisomy 21." Blue Cross Blue Shield Association Technology Evaluation Center, In Press
8. Gil MM, Accurti V, Santacruz B, et al. Analysis of Cell-Free DNA in Maternal Blood in Screening For Aneuploidies: Updated Meta-Analysis. *Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology*. 2017. PMID: 28397325
9. Norton ME, Baer RJ, Wapner RJ, et al. Cell-free DNA vs sequential screening for the detection of fetal chromosomal abnormalities. *American journal of obstetrics and gynecology*. 2016;214(6):727 e1-6. PMID: 26709085
10. Mackie FL, Hemming K, Allen S, et al. The accuracy of cell-free fetal DNA-based non-invasive prenatal testing in singleton pregnancies: a systematic review and bivariate meta-analysis. *BJOG : an international journal of obstetrics and gynaecology*. 2017;124(1):32-46. PMID: 27245374
11. Badeau M, Lindsay C, Blais J, et al. Genomics-based non-invasive prenatal testing for detection of fetal chromosomal aneuploidy in pregnant women. *The Cochrane database of systematic reviews*. 2017;11:CD011767. PMID: 29125628
12. Bussolaro S, Raymond YC, Acreman ML, et al. The accuracy of prenatal cell-free DNA screening for sex chromosome abnormalities: A systematic review and meta-analysis. *Am J Obstet Gynecol MFM*. 2023;5(3):100844. PMID: 36572107
13. Gil MM, Quezada MS, Revello R, et al. Analysis of cell-free DNA in maternal blood in screening for fetal aneuploidies: updated meta-analysis. *Ultrasound in obstetrics & gynecology : the official journal of the International Society of Ultrasound in Obstetrics and Gynecology*. 2015;45(3):249-66. PMID: 25639627
14. Rose NC, Barrie ES, Malinowski J, et al. Systematic evidence-based review: The application of noninvasive prenatal screening using cell-free DNA in general-risk

- pregnancies. *Genetics in medicine : official journal of the American College of Medical Genetics*. 2022;24(7):1379-91. PMID: 35608568
15. Zaninović L, Bašković M, Ježek D, et al. Validity and Utility of Non-Invasive Prenatal Testing for Copy Number Variations and Microdeletions: A Systematic Review. *J Clin Med*. 2022;11(12). PMID: 35743413
 16. Familiari A, Boito S, Rembouskos G, et al. Cell-free DNA analysis of maternal blood in prenatal screening for chromosomal microdeletions and microduplications: a systematic review. *Prenatal diagnosis*. 2021;41(10):1324-31. PMID: 33710639
 17. Dar P, Jacobsson B, Clifton R, et al. Cell-free DNA screening for prenatal detection of 22q11.2 deletion syndrome. *American journal of obstetrics and gynecology*. 2022;227(1):79.e1-79.e11. PMID: 35033576
 18. Soster E, Boomer T, Hicks S, et al. Three years of clinical experience with a genome-wide cfDNA screening test for aneuploidies and copy-number variants. *Genetics in medicine : official journal of the American College of Medical Genetics*. 2021;23(7):1349-55. PMID: 33731879
 19. Wang C, Tang J, Tong K, et al. Expanding the application of non-invasive prenatal testing in the detection of foetal chromosomal copy number variations. *BMC Med Genomics*. 2021;14(1):292. PMID: 34895207
 20. Devaney SA, Palomaki GE, Scott JA, et al. Noninvasive fetal sex determination using cell-free fetal DNA: a systematic review and meta-analysis. *JAMA*. 2011;306:627-36. PMID: 21828326
 21. Wright CF, Wei Y, Higgins JP, et al. Non-invasive prenatal diagnostic test accuracy for fetal sex using cell-free DNA a review and meta-analysis. *BMC Res Notes*. 2012;5:476. PMID: 22937795
 22. Colmant C, Morin-Surroca M, Fuchs F, et al. Non-invasive prenatal testing for fetal sex determination: is ultrasound still relevant? *European journal of obstetrics, gynecology, and reproductive biology*. 2013;171(2):197-204. PMID: 24094458
 23. Zhang J, Li J, Saucier JB, et al. Non-invasive prenatal sequencing for multiple Mendelian monogenic disorders using circulating cell-free fetal DNA. *Nat Med*. 2019;25(3):439-47. PMID: 30692697
 24. Westin ER, Tsao DS, Atay O, et al. Validation of single-gene noninvasive prenatal testing for sickle cell disease. *Am J Hematol*. 2022;97(7):E270-e73. PMID: 35429177
 25. Hoskovec J, Hardisty EE, Talati AN, et al. Maternal carrier screening with single-gene NIPS provides accurate fetal risk assessments for recessive conditions. *Genetics in medicine : official journal of the American College of Medical Genetics*. 2023;25(2):100334. PMID: 36454238
 26. Wynn J, Hoskovec J, Carter RD, et al. Performance of single-gene noninvasive prenatal testing for autosomal recessive conditions in a general population setting. *Prenatal diagnosis*. 2023;43(10):1344-54. PMID: 37674263
 27. Screening for Fetal Chromosomal Abnormalities: ACOG Practice Bulletin Summary, Number 226. *Obstetrics and gynecology*. 2020;136(4):859-67. PMID: 32976375
 28. American College of Obstetricians and Gynecologists. (2021) Cell-free DNA to Screen for Single-Gene Disorders. [cited 2/26/2025]. 'Available from:' <https://www.acog.org/clinical/clinical-guidance/practice-advisory/articles/2019/02/cell-free-dna-to-screen-for-single-gene-disorders>.
 29. Dungan JS, Klugman S, Darilek S, et al. Noninvasive prenatal screening (NIPS) for fetal chromosome abnormalities in a general-risk population: An evidence-based clinical guideline of the American College of Medical Genetics and Genomics (ACMG).

CODES

NOTE: There are specific CPT codes for trisomy testing and for microdeletion testing. It is inappropriate to use nonspecific molecular pathology CPT codes (i.e., 81400-81408) for trisomy or microdeletion testing.

| Codes | Number | Description |
|-------|--------|---|
| CPT | 0060U | Twin zygosity, genomic targeted sequence analysis of chromosome 2, using circulating cell-free fetal DNA in maternal blood |
| | 0341U | Fetal aneuploidy DNA sequencing comparative analysis, fetal DNA from products of conception, reported as normal (euploidy), monosomy, trisomy, or partial deletion/duplication, mosaicism, and segmental aneuploid |
| | 0449U | Carrier screening for severe inherited conditions (eg, cystic fibrosis, spinal muscular atrophy, beta hemoglobinopathies [including sickle cell disease], alpha thalassemia), regardless of race or self-identified ancestry, genomic sequence analysis panel, must include analysis of 5 genes (<i>CFTR</i> , <i>SMN1</i> , <i>HBB</i> , <i>HBA1</i> , <i>HBA2</i>) |
| | 0489U | Obstetrics (single-gene noninvasive prenatal test), cell free DNA sequence analysis of 1 or more targets (eg, <i>CFTR</i> , <i>SMN1</i> , <i>HBB</i> , <i>HBA1</i> , <i>HBA2</i>) to identify paternally inherited pathogenic variants, and relative mutation-dosage analysis based on molecular counts to determine fetal inheritance of maternal mutation, algorithm reported as a fetal risk score for the condition (eg, cystic fibrosis, spinal muscular atrophy, beta hemoglobinopathies [including sickle cell disease], alpha thalassemia) |
| | 81220 | <i>CFTR</i> (cystic fibrosis transmembrane conductance regulator) (eg, cystic fibrosis) gene analysis; common variants (eg, ACMG/ACOG guidelines) |
| | 81243 | <i>FMR1</i> (fragile X messenger ribonucleoprotein 1) (eg, fragile X syndrome, X-linked intellectual disability [XLID]) gene analysis; evaluation to detect abnormal (eg, expanded) alleles |
| | 81329 | <i>SMN1</i> (survival of motor neuron 1, telomeric) (eg, spinal muscular atrophy) gene analysis; dosage/deletion analysis (eg, carrier testing), includes <i>SMN2</i> (survival of motor neuron 2, centromeric) analysis, if performed |
| | 81363 | <i>HBB</i> (hemoglobin, subunit beta) (eg, sickle cell anemia, beta thalassemia, hemoglobinopathy); duplication/deletion variant(s) |
| | 81364 | <i>HBB</i> (hemoglobin, subunit beta) (eg, sickle cell anemia, beta thalassemia, hemoglobinopathy); full gene sequence |
| | 81408 | Molecular pathology procedure, Level 9 |
| | 81422 | Fetal chromosomal microdeletion(s) genomic sequence analysis (eg, DiGeorge syndrome, Cri-du-chat syndrome), circulating cell-free fetal DNA in maternal blood |
| | 81479 | Unlisted molecular pathology procedure |
| | 81599 | Unlisted multianalyte assay with algorithmic analysis |
| HCPCS | None | |

Date of Origin: January 2013