Noninvasive Prenatal Screening for Fetal Aneuploidies and Microdeletions, and Twin Zygosity Using Cell-Free Fetal DNA

Policy # 00345
Original Effective Date: 12/20/2013
Current Effective Date: 05/10/2021

Applies to all products administered or underwritten by Blue Cross and Blue Shield of Louisiana and its subsidiary, HMO Louisiana, Inc.(collectively referred to as the "Company"), unless otherwise provided in the applicable contract. Medical technology is constantly evolving, and we reserve the right to review and update Medical Policy periodically.

Note: Genetic Testing for Developmental Delay/Intellectual Disability, Autism Spectrum Disorder, and Congenital Anomalies is addressed separately in medical policy 00536.

When Services Are Eligible for Coverage

Coverage for eligible medical treatments or procedures, drugs, devices or biological products may be provided only if:

- *Benefits are available in the member's contract/certificate, and*
- *Medical necessity criteria and guidelines are met.*

Based on review of available data, the Company may consider nucleic acid sequencing-based testing of maternal plasma to screen for trisomy 21, 18, and 13 in women with singleton pregnancies to be **eligible for coverage.****

When Services Are Considered Investigational

Coverage is not available for investigational medical treatments or procedures, drugs, devices or biological products.

Based on review of available data, the Company considers nucleic acid sequencing-based testing of maternal plasma for fetal sex chromosome aneuploidies to be **investigational.***

Based on review of available data, the Company considers nucleic acid sequencing-based testing of maternal plasma for trisomy 21 in women with twin or multiple pregnancies to be **investigational.***
Based on review of available data, the Company considers nucleic acid sequencing-based testing of maternal plasma for trisomy 13 and/or 18, other than in the situations specified above to be **investigational.***

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Based on review of available data, the Company considers nucleic acid sequencing-based testing of maternal plasma for microdeletions to be **investigational.***

Based on review of available data, the Company considers nucleic acid sequencing-based testing of maternal plasma for twin zygosity to be **investigational.***

Based on review of available data, the Company considers Vanadis NIPT of maternal plasma to screen for trisomy 21, 18 and 13 in all situations to be **investigational.***

Policy Guidelines

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 Committee Opinion No. 640, the American College of Obstetricians and Gynecologists (2015) 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 (ie, "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 the risk of neural tube defects. Patients should continue to be offered ultrasound or maternal serum α-fetoprotein screening.

Genetic Counseling
Experts recommend formal genetic counseling for patients who are at risk for inherited disorders and who wish to undergo genetic testing. Interpreting the results of genetic tests and understanding

risk factors can be difficult for some patients; genetic counseling helps individuals understand the impact of genetic testing, including the possible effects the test results could have on the individual or their family members. It should be noted that genetic counseling may alter the utilization of genetic testing substantially and may reduce inappropriate testing; further, genetic counseling should be performed by an individual with experience and expertise in genetic medicine and genetic testing methods.

Background/Overview

Fetal Aneuploidy

Fetal chromosomal abnormalities occur in approximately 1 in 160 live births. Most fetal chromosomal abnormalities are aneuploidies, defined as an abnormal number of chromosomes. The trisomy syndromes are aneuploidies involving 3 copies of 1 chromosome. The most important risk factor for trisomy syndromes is maternal age. The approximate risk of a trisomy 21 (T21; Down syndrome)-affected birth is 1 in 1100 at age 25 to 29. The risk of a fetus with T21 (at 16 weeks of gestation) is about 1 in 250 at age 35 and 1 in 75 at age 40.

T21 is the most common chromosomal aneuploidy and provides the impetus for current maternal serum screening programs. Other trisomy syndromes include T18 (Edwards syndrome) and T13 (Patau syndrome), which are the next most common forms of fetal aneuploidy, although the percentage of cases surviving to birth is low and survival beyond birth is limited. Detection of T18 and T13 early in pregnancy can facilitate preparation for fetal loss or early intervention.

Fetal Aneuploidy Screening

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 chorionic villous sampling (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.

Commercial, noninvasive, sequencing-based testing of maternal serum for fetal trisomy syndromes is now available. The testing technology involves the detection of cell-free fetal DNA fragments present in the plasma of pregnant women. As early as 8 to 10 weeks of gestation, these fetal DNA fragments comprise 6% to 10% or more of the total cell-free fetal DNA in a maternal plasma sample. The tests are unable to provide a result if the fetal fraction is too low (ie, <4%). The fetal fraction can be affected by maternal and fetal characteristics. For example, the fetal fraction was found to be lower at higher maternal weights and higher with increasing fetal crown-rump length.

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. Dizygotic or "fraternal" twins occur from ovulation and fertilization of 2 oocytes, which results in dichorionic (DC) placentation and 2 separate placentas. In contrast to DC twins, MC twin pregnancies share their blood supply. Monochorionic (MC) 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 DC twins. Up to 15% of MC twin pregnancies are affected by twin to twin transfusion syndrome (TTTS), a condition characterized by relative hypovolemia of 1 twin and hypervolemia of the other. According to estimates from live births, TTTS occurs in up to 15% of MC 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. Noninvasive prenatal testing (NIPT) using cell-free fetal DNA to determine zygosity in twin pregnancies could potentially inform decisions about early surveillance for TTTS and other MC twin-related abnormalties. In particular, determining zygosity with NIPT could potentially assist in the assessment of chorionicity when ultrasound findings are not clear.

Cell-Free Fetal DNA Analysis Methods

Sequencing-based tests use 1 of 2 general approaches to analyzing cell-free fetal DNA. The first category of tests uses quantitative or counting methods. The most widely used technique to date uses massively parallel sequencing (MPS; also known as next-generation sequencing). DNA fragments

are amplified by polymerase chain reaction; during the sequencing process, the amplified fragments are spatially segregated and sequenced simultaneously in a massively parallel fashion. Sequenced fragments can be mapped to the reference human genome to obtain numbers of fragment counts per chromosome. The sequencing-derived percent of fragments from the chromosome of interest reflects the chromosomal representation of the maternal and fetal DNA fragments in the original maternal plasma sample. Another technique is direct DNA analysis, which analyzes specific cell-free fetal DNA fragments across samples and requires approximately a tenth the number of cell-free DNA fragments as MPS. The digital analysis of selected regions (DANSR™) is an assay that uses direct DNA analysis.

The second general approach is single nucleotide variant-based methods. They use targeted amplification and analysis of approximately 20000 single nucleotide variants on selected chromosomes (eg, 21, 18, 13) in a single reaction. A statistical algorithm is used to determine the number of each type of chromosome. At least some of the commercially available cell-free fetal DNA prenatal tests also test for other abnormalities including sex chromosome abnormalities and selected microdeletions.

A newer approach to cell free DNA testing called the Vanadis NIPT does not involve amplification or sequencing. The assay uses maternal serum and applies a series of enzymes to create labelled rolling circle replication products (RCPs) from chromosomal cell-free DNA targets, which are then converted to fluorescent DNA molecules and labeled with chromosome-specific fluorophores. The labeled fluorescent DNA molecules are deposited to a microfilter plate and counted with an automated imaging device. The ratio between the number of each chromosome-specific fluorescent DNA molecules is transferred for risk calculation to proprietary software to calculate the likelihood of a trisomy. Currently, Vanadis NIPT provides results for trisomy 21, trisomy 18 and trisomy 13; although, additional aneuploidies and microdeletions might be added in the future.

Copy Number Variants and Clinical Disorders
Microdeletions (also known as submicroscopic deletions) are chromosomal deletions that are too small to be detected by microscopy or conventional cytogenetic methods. They can be as small as 1 and 3 megabases long. Along with microduplications, microdeletions are collectively known as copy number variants. Copy number variants can lead to disease when the change in copy number of a dose-sensitive gene or genes disrupts the ability of the gene(s) to function and affects the amount of

protein produced. A number of genomic disorders associated with microdeletion have been identified, which may be associated with serious clinical features, such as cardiac anomalies, immune deficiency, palatal defects, and developmental delay as in DiGeorge syndrome. Some of the syndromes (eg, DiGeorge) have complete penetrance yet marked variability in clinical expressivity. A contributing factor is that the breakpoints of the microdeletions may vary, and there may be a correlation between the number of haploinsufficient genes and phenotypic severity.

A proportion of microdeletions are inherited and some are de novo. Accurate estimates of the prevalence of microdeletion syndromes during pregnancy or at birth are not available. The risk of a fetus with a microdeletion syndrome is independent of maternal age. There are few population-based data and most studies published to date have based estimates on phenotypic presentation. The 22q11.2 (DiGeorge) microdeletion is the most common associated with a clinical syndrome. Table 1 provides prevalence estimates for the most common microdeletion syndromes. These numbers likely underestimate the prevalence of these syndromes in the prenatal population because the population of variant carriers includes phenotypically normal or very mildly affected individuals.

Table 1. Recurrent Microdeletion Syndromes

Syndrome	Location	Estimated Prevalence
DiGeorge	22q11.2	1/2000
1p36 deletion	1p36-	1/5000
Prader-Willi and Angelman	Del 15q11.2	1/20,000
Wolf-Hirschhorn	4p-	1/50,000 to 1/20,000
Cri du chat	5p-	1/50,000
Miller-Dieker	Del 17p13.3	1 /100,000

Adapted from Chitty et al (2018).

Routine prenatal screening for microdeletion syndromes is not recommended by national organizations. Current practice is to offer invasive prenatal diagnostic testing in select cases to women when a prenatal ultrasound indicates anomalies (eg, heart defects, cleft palate) that could be

associated with a particular microdeletion syndrome. Samples are analyzed using fluorescence in situ hybridization, chromosomal microarray analysis, or karyotyping. Additionally, families at risk (eg, those known to have the deletion or with a previously affected child) generally receive genetic counseling and those who conceive naturally may choose prenatal diagnostic testing. Most affected individuals, though, are identified postnatally based on clinical presentation and may be confirmed by genetic testing. Using 22q11.2 deletion syndrome as an example, although clinical characteristics vary, palatal abnormalities (eg, cleft palate) occur in approximately 69% of individuals, congenital heart disease in 74%, and characteristic facial features are present in a majority of individuals of northern European heritage.

FDA or Other Governmental Regulatory Approval

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 must meet the general regulatory standards of the Clinical Laboratory Improvement Act. Laboratories that offer laboratory-developed tests must be licensed by the Clinical Laboratory Improvement Act for high-complexity testing. To date, the U.S. Food and Drug Administration has chosen not to require any regulatory review of noninvasive prenatal screening tests using cell-free fetal DNA.

Commercially available tests include but are not limited to the following:

- Myriad Prequel(TM) Prenatal Screen (Myriad Women's Health, Counsyl) utilizes whole genome sequencing for detecting aneuploidy including T21, T18, T13
- VisibiliT (Sequenom Laboratories, now LabCorp) tests for T21 and T18, and tests for sex.
- MateriT21 PLUS (Sequenom Laboratories, now LabCorp) core test includes T21, T18, T13, and fetal sex aneuploidies. The enhanced sequencing series includes testing for T16, T22, and 7 microdeletions: 22q deletion syndrome (DiGeorge syndrome), 5p (cri du chat syndrome), 15q (Prader-Willi and Angelman syndromes), 1p36 deletion syndrome, 4p (Wolf-Hirschhorn syndrome), 8q (Langer-Giedion syndrome), and 11q (Jacobsen syndrome). The test uses MPS and reports results as positive or negative. The enhanced sequencing series is offered on an opt-out basis.
- Harmony (Ariosa Diagnostics, now Roche) tests for T21, T18, and T13. The test uses directed DNA analysis and results are reported as a risk score.

- Panorama (Natera) is a prenatal test for detecting T21, T18, and T13, as well as select sex chromosome abnormalities. It uses single nucleotide variant technology; results are reported as a risk score. An extended panel tests for 5 microdeletions: 22q deletion syndrome (DiGeorge syndrome), 5p (cri du chat syndrome), 15q11-13 (Prader-Willi and Angelman syndromes), and 1p36 deletion syndrome. Screening for 22q11.2 will be included in the panel unless the opt-out option is selected; screening for the remaining 4 microdeletions is offered on an opt-in basis.
- Verifi®‡ (Verinata Health, now Illumina) is a prenatal test for T21, T18, and T13. The test uses MPS and calculates a normalized chromosomal value, reporting results as 1 of 3 categories: no aneuploidy detected, aneuploidy detected, or aneuploidy suspected.
- InformaSeq (Integrated Genetics, now LabCorp) is a prenatal test for detecting T21, T18, and T13, with optional testing for select sex chromosome abnormalities. It uses the Illumina platform and reports results in a similar manner.
- QNatal Advanced (Quest Diagnostics) tests for T21, T18, and T13.
- Vanadis NIPT Solution (PerkinElmer) tests for T21, T18, and T13.
- Veracity (NIPD Genetics) tests for T21, T18, and T13, sex chromosome aneuploidies, and microdeletions.

Rationale/Source

Description

National guidelines recommend that all pregnant women be offered screening for fetal chromosomal abnormalities, most of which are aneuploidies, an abnormal number of chromosomes. Trisomy syndromes are aneuploidies involving 3 copies of 1 chromosome. Trisomies 21, 18, and 13 are the most common forms of fetal aneuploidy that survive to birth. There are numerous limitations to standard screening for these disorders using the maternal serum and fetal ultrasound. Noninvasive prenatal screening analyzing cell-free fetal DNA in maternal serum is a potential complement or alternative to conventional serum screening. Noninvasive prenatal screening using cell-free fetal DNA has also been proposed to screen for microdeletions. Prenatal testing for twin zygosity using cell-free fetal DNA has been proposed to inform decisions about early surveillance for twin twin transfusion syndrome and other monochorionic twin-related abnormalties.

Summary of Evidence

For individuals who have a singleton pregnancy who receive NIPS for T21, T18, and T13 using cell-free fetal DNA, the evidence includes observational studies and systematic reviews. Relevant outcomes are test accuracy and validity, morbid events, and resource utilization. Published studies on available tests and meta-analyses of these studies have consistently demonstrated very high sensitivity and specificity for detecting Down syndrome (T21) in singleton pregnancies. Most studies included only women at high-risk of T21 but several studies have reported similar levels of diagnostic accuracy in average-risk women. Compared with standard serum screening, both the sensitivity and specificity of cell-free fetal DNA screening are considerably higher. As a result, screening with cell-free fetal DNA for T21 will result in fewer missed cases of Down syndrome, fewer invasive procedures, and fewer cases of pregnancy loss following invasive procedures. Screening for T18 and T13 along with T21 may allow for preparation for fetal demise or termination of the pregnancy prior to fetal loss. The evidence is sufficient to determine that the technology results in a meaningful improvement in the net health outcome.

For individuals who have a singleton pregnancy who receive NIPS for sex chromosome aneuploidies using cell-free fetal DNA, the evidence includes observational studies, mainly in high-risk pregnancies, and systematic reviews. Relevant outcomes are test accuracy and validity, morbid events, and resource utilization. Meta-analyses of available data have suggested high sensitivities and specificities but the small number of cases makes definitive conclusions difficult. In addition, the clinical utility of identifying sex chromosome aneuploidies during pregnancy is uncertain. The evidence is insufficient to determine the effects of the technology on health outcomes.

For individuals who have a twin or multiple pregnancy who receive NIPS for aneuploidies using cell-free fetal DNA, the evidence includes observational studies and systematic reviews. Relevant outcomes are test accuracy and validity, morbid events, and resource utilization. The total number of cases of aneuploidy identified in these studies is small and is insufficient to draw conclusions about clinical validity. There is a lack of direct evidence of clinical utility, and a chain of evidence cannot be conducted due to the paucity of evidence on clinical validity. The evidence is insufficient to determine the effects of the technology on health outcomes.

For individuals with pregnancy(ies) who receive NIPS for microdeletions using cell-free fetal DNA, the evidence includes several observational studies. Relevant outcomes are test accuracy and

validity, morbid events, and resource utilization. The available studies on clinical validity have limitations (eg, missing data on confirmatory testing, false-negatives), and the added benefit of NIPS compared with current approaches is unclear. Moreover, the clinical utility of NIPS for microdeletions remains unclear and has not been evaluated in published studies. The evidence is insufficient to determine the effects of the technology on health outcomes.

For individuals who have twin pregnancy who receive noninvasive prenatal testing (NIPT) for twin zygosity using cell-free fetal DNA, the evidence includes an observational study. Relevant outcomes are test accuracy and validity, morbid events, and resource utilization. Sensitivity and specificity were high (100%) in 1 validation study conducted in 95 twin gestations. This evidence is too limited to draw conclusions about performance characteristics and would need to be confirmed in additional, well-conducted studies. Moreover, the clinical utility of NIPT for twin zygosity compared to standard methods such as ultrasound is unclear and has not been evaluated in published studies. The evidence is insufficient to determine the effects of the technology on health outcomes.

For individuals who have a singleton pregnancy who receive NIPS for T21, T18, and T13 usingVanadis NIPT, the evidence includes 2 industry sponsored studies. Relevant outcomes are test accuracy and validity, morbid events, and resource utilization. The available studies on clinical validity have limitations, and the added benefit of Vanadis NIPT compared with current approaches is unclear. Moreover, the clinical utility of Vanadis NIPT remains unclear and has not been evaluated in published studies. The evidence is insufficient to determine the effects of the technology on health outcomes.

Supplemental Information

Clinical Input From Physician Specialty Societies and Academic Medical Centers

While the various physician specialty societies and academic medical centers may collaborate with and make recommendations during this process, through the provision of appropriate reviewers, input received does not represent an endorsement or position statement by the physician specialty societies or academic medical centers, unless otherwise noted.

In response to requests, input was received from 3 physician specialty societies and 4 academic medical centers while this policy was under review in 2013. There was a consensus that sequencing-

based tests to determine trisomy 21 (T21) from maternal plasma cell-free fetal DNA may be considered medically necessary in women with high-risk singleton pregnancies undergoing screening for T21. The input was mixed on whether sequencing-based tests to determine T21 from maternal plasma DNA may be considered medically necessary in women with average-risk singleton pregnancies. An American College of Obstetricians and Gynecologists genetics committee opinion, included as part of the specialty society's input, did not then recommend the new tests for women with singleton pregnancies who were not at high-risk of aneuploidy. There was a consensus that sequencing-based tests to determine T21 from maternal plasma DNA are investigational for women with multiple pregnancies. Regarding an appropriate protocol for using sequencing-based testing, there was a consensus that testing should not be used as a single screening test without confirmation of results by karyotyping. There was mixed input on the use of the test as a replacement for standard screening tests with karyotyping confirmation and on use as a secondary screen in women with screen positive on standard screening tests with karyotyping confirmation. Among the 5 reviewers who responded to the questions (which did not include the American College of Obstetricians and Gynecologist), there was a consensus that the modeling approach is sufficient to determine the clinical utility of the new tests and near-consensus there is no need for clinical trials comparing a screening protocol using the new tests to a screening protocol using standard serum screening before initiation of clinical use of the tests.

Practice Guidelines and Position Statements

American College of Obstetricians and Gynecologists and Society for Maternal-Fetal Medicine
In 2016, the American College of Obstetricians and Gynecologists and the Society for Maternal-Fetal Medicine released a joint practice bulletin summary (No. 163) on the screening for fetal aneuploidy. The following recommendations on cell-free DNA were based on "good and consistent" scientific evidence:

- "Women who have a negative screening test result should not be offered additional screening tests for aneuploidy because this will increase their potential for a false-positive test result."
- "Because cell-free DNA is a screening test with the potential for false-positive and false-negative results, such testing should not be used as a substitute for diagnostic testing."
- "All women with a positive cell-free DNA test result should have a diagnostic procedure before any irreversible action, such as pregnancy termination, is taken."

- "Women whose cell-free DNA screening test results are not reported, are indeterminate, or are uninterpretable (a no-call test result) should receive further genetic counseling and be offered comprehensive ultrasound evaluation and diagnostic testing because of an increased risk of aneuploidy."

The following recommendations were based on "limited or inconsistent" scientific evidence:

- "Cell-free DNA screening tests for microdeletions have not been validated clinically and are not recommended at this time."
- "No method of aneuploidy screening is as accurate in twin gestations as it is in singleton pregnancies. Because data generally are unavailable for higher-order multifetal gestations, analyte screening for fetal aneuploidy should be limited to singleton and twin pregnancies."

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

- "Some women who receive a positive test result from traditional screening may prefer to have cell-free DNA screening rather than undergo definitive testing."
- "This approach may delay definitive diagnosis and management and may fail to identify some fetuses with aneuploidy."
- "Parallel or simultaneous testing with multiple screening methodologies for aneuploidy is not cost-effective and should not be performed."

American College of Medical Genetics and Genomics

In 2016, the American College of Medical Genetics and Genomics published a position statement on noninvasive prenatal screening (NIPS) for fetal aneuploidy. The relevant recommendations are as follows:

- "Informing all pregnant women that NIPS is the most sensitive screening option for traditionally screened aneuploidies (i.e., Patau, Edwards, and Down syndromes)."
- "Referring patients to a trained genetics professional when an increased risk of aneuploidy is reported after NIPS."
- "Offering diagnostic testing when a positive screening test result is reported after NIPS."
- "Providing accurate, balanced, up-to-date information, at an appropriate literacy level when a fetus is diagnosed with a chromosomal or genomic variation in an effort to educate

prospective parents about the condition of concern. These materials should reflect the medical and psychosocial implications of the diagnosis."

The American College of Medical Genetics and Genomics did not recommend "NIPS to screen for autosomal aneuploidies other than those involving chromosomes 13, 18, and 21."

International Society for Prenatal Diagnosis

In 2015, the International Society for Prenatal Diagnosis published a position statement on the prenatal diagnosis of chromosomal abnormalities, updating its 2013 statement. (Note that a number of the authors of the 2015 report had financial links to industry.) The following summarizes the Society's recommendations:

"I. High sensitivities and specificities are potentially achievable with cfDNA [cell-free DNA] screening for some fetal aneuploidies, notably trisomy 21.

II. Definitive diagnosis of Down syndrome and other fetal chromosome abnormalities can only be achieved through testing on cells obtained by amniocentesis or CVS [chorionic villous sampling].

III. The use of maternal age alone to assess fetal Down syndrome risk in pregnant women is not recommended.

IV. A combination of ultrasound NT [nuchal translucency] measurement and maternal serum markers in the first trimester should be available to women who want an early risk assessment and for whom cfDNA screening cannot be provided.

V. A 4 marker serum test should be available to women who first attend for their prenatal care after 13 weeks 6 days of pregnancy and where cfDNA screening cannot be provided.

VI. Protocols that combine first trimester and second trimester conventional markers are valid.

VII. Second trimester ultrasound can be a useful adjunct to conventional aneuploidy screening protocols.

VIII. When cfDNA screening is extended to microdeletion and microduplication syndromes or rare trisomies the testing should be limited to clinically significant disorders or well defined severe conditions. There should be defined estimates for the detection rates, false-positive rates, and information about the clinical significance of a positive test for each disorder being screened."

U.S. Preventive Services Task Force Recommendations
The U.S. Preventive Services Task Force does not currently address screening for Down syndrome. This syndrome was addressed in the 1990s; it is no longer listed on the Task Force website.

Medicare National Coverage
There is no national coverage determination. In the absence of a national coverage determination, coverage decisions are left to the discretion of local Medicare carriers.

Ongoing and Unpublished Clinical Trials
Some currently ongoing and unpublished trials that might influence this evidence review are listed in Table 2.

Table 2. Summary of Key Trials

NCT No.	Trial Name	Planned Enrollment	Completion Date
Ongoing			
NCT03375359	First Trimester Screening for Trisomy 21, 18, 13 and 22q11.2 Deletion Syndrome - ReFaPo02	1000	Dec 2020
NCT01545674[a]	Prenatal Non-invasive Aneuploidy Test Utilizing SNPs Trial (PreNATUS)	1000	Dec 2020
NCT02381457[a]	SNP-based Microdeletion and Aneuploidy RegisTry (SMART)	20960	Mar 2020
NCT04437992[a]	Feasibility Study of a New Screening Program for Major Aneuploidies (T21, T18, T13) in the Emilia-Romagna Region (SAPERER)	7000	Apr 2021
NCT03559374[a]	Study of Vanadis NIPT for Non-Invasive Prental Screening of Trisomies (T21, T18, andT13)	1200	Aug 2020
Unpublished			

NCT03200041[a]	Clinical Evaluation of the IONA Test for Non-invasive Pre Natal Screening in Twin Pregnancies	1000	Jul 2019
NCT01472523[a]	A Safer Pre-Natal Diagnosis Using Free DNA in Maternal Blood	1632	Jul 2019

NCT: national clinical trial.
[a]Denotes industry-sponsored or cosponsored trial.

References

1. Blue Cross and Blue Shield Association, Medical Policy Reference Manual, "Noninvasive Prenatal Screening for Fetal Aneuploidies and Microdeletions Using Cell-Free Fetal DNA", 4.01.21, September 2020.
2. Hook EB, Cross PK, Schreinemachers DM. Chromosomal abnormality rates at amniocentesis and in live-born infants. JAMA. Apr 15 1983; 249(15): 2034-8. PMID 6220164
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. Jun 28 2019; 8(7). PMID 31261782
4. UptoDate (2020).Twin-twin transfusion syndrome and twin anemia polycythemia sequence: Screening, prevalence, pathophysiology, and diagnosis. https://www.uptodate.com/contents/twin-twin-transfusion-syndrome-and-twin-anemia-polycythemia-sequence-screening-prevalence-pathophysiology-and-diagnosis?search=twin%20twin%20transfusion%20syndrome&source=search_result&selectedTitle=1~35&usage_type=default&display_rank=1.
5. Chitty LS, Hudgins L, Norton ME. Current controversies in prenatal diagnosis 2: Cell-free DNA prenatal screening should be used to identify all chromosome abnormalities. Prenat Diagn. Feb 2018; 38(3): 160-165. PMID 29417608
6. Blue Cross Blue Shield Association Technology Evaluation Center (TEC). Noninvasive prenatal cell-free fetal DNA-based screening for aneuploidies other than trisomy 21. TEC Assessment Program. 2014;Volume 29:Tab 7.

7. Blue Cross Blue Shield Association Technology Evaluation Center (TEC). Sequencing-based tests to determine fetal down syndrome (trisomy 21) from maternal plasma DNA. TEC Assessment Program. 2013;Volume 27:Tab 10.
8. Badeau M, Lindsay C, Blais J, et al. Genomics-based non-invasive prenatal testing for detection of fetal chromosomal aneuploidy in pregnant women. Cochrane Database Syst Rev. Nov 10 2017; 11: CD011767. PMID 29125628
9. Ohno M, Caughey A. The role of noninvasive prenatal testing as a diagnostic versus a screening tool--a cost-effectiveness analysis. Prenat Diagn. Jul 2013; 33(7): 630-5. PMID 23674316
10. Liao H, Liu S, Wang H. Performance of non-invasive prenatal screening for fetal aneuploidy in twin pregnancies: a meta-analysis. Prenat Diagn. Sep 2017; 37(9): 874-882. PMID 28728213
11. Gil MM, Galeva S, Jani J, et al. Screening for trisomies by cfDNA testing of maternal blood in twin pregnancy: update of The Fetal Medicine Foundation results and meta-analysis. Ultrasound Obstet Gynecol. Jun 2019; 53(6): 734-742. PMID 31165549
12. Dyr B, Boomer T, Almasri EA, et al. A new era in aneuploidy screening: cfDNA testing in 30,000 multifetal gestations: Experience at one clinical laboratory. PLoS ONE. 2019; 14(8): e0220979. PMID 31393959
13. Kypri E, Ioannides M, Touvana E, et al. Non-invasive prenatal testing of fetal chromosomal aneuploidies: validation and clinical performance of the veracity test. Mol Cytogenet. 2019; 12: 34. PMID 31338126
14. Motevasselian M, Saleh Gargari S, Younesi S, et al. Non-invasive prenatal test to screen common trisomies in twin pregnancies. Mol Cytogenet. 2020; 13: 5. PMID 32042312
15. Gross SJ, Stosic M, McDonald-McGinn DM, et al. Clinical experience with single-nucleotide polymorphism-based non-invasive prenatal screening for 22q11.2 deletion syndrome. Ultrasound Obstet Gynecol. Feb 2016; 47(2): 177-83. PMID 26396068
16. Helgeson J, Wardrop J, Boomer T, et al. Clinical outcome of subchromosomal events detected by whole-genome noninvasive prenatal testing. Prenat Diagn. Oct 2015; 35(10): 999-1004. PMID 26088833
17. Petersen AK, Cheung SW, Smith JL, et al. Positive predictive value estimates for cell-free noninvasive prenatal screening from data of a large referral genetic diagnostic laboratory. Am J Obstet Gynecol. Dec 2017; 217(6): 691.e1-691.e6. PMID 29032050
18. Dahl F, Ericsson O, Karlberg O, et al. Imaging single DNA molecules for high precision NIPT. Sci Rep. 2018;8(1):4549. Published 2018 Mar 14. doi:10.1038/s41598-018-22606-0

19. Ericsson O, Ahola T, Dahl F, et al. Clinical validation of a novel automated cell-free DNA screening assay for trisomies 21, 13, and 18 in maternal plasma. Prenat Diagn. 2019;39(11):1011-1015. doi:10.1002/pd.5528
20. Practice Bulletin No. 163 Summary: Screening for Fetal Aneuploidy. Obstet Gynecol. May 2016; 127(5): 979-81. PMID 27101120
21. Gregg AR, Skotko BG, Benkendorf JL, et al. Noninvasive prenatal screening for fetal aneuploidy, 2016 update: a position statement of the American College of Medical Genetics and Genomics. Genet Med. Oct 2016; 18(10): 1056-65. PMID 27467454
22. Benn P, Borrell A, Chiu RW, et al. Position statement from the Chromosome Abnormality Screening Committee on behalf of the Board of the International Society for Prenatal Diagnosis. Prenat Diagn. Aug 2015; 35(8): 725-34. PMID 25970088
23. Benn P, Borell A, Chiu R, et al. Position statement from the Aneuploidy Screening Committee on behalf of the Board of the International Society for Prenatal Diagnosis. Prenat Diagn. Jul 2013; 33(7): 622-9. PMID 23616385

Policy History

Original Effective Date: 12/20/2013
Current Effective Date: 05/10/2021

02/20/2013 Medical Policy Implementation Committee approval. New policy.
02/06/2014 Medical Policy Committee review
02/19/2014 Medical Policy Implementation Committee approval. Coverage eligibility unchanged.
01/01/2015 Coding Update
02/05/2015 Medical Policy Committee review
02/18/2015 Medical Policy Implementation Committee approval. Title changed from "Sequencing-based Tests to Determine Trisomy 21 from Maternal Plasma DNA" to "Noninvasive Prenatal Testing for Fetal Aneuploidies Using Cell-Free Fetal DNA". Removed the statement from the coverage section that stated to deny as investigational if criteria are not met for clarification. Statement added that concurrent nucleic acid sequencing-based testing of maternal plasma for trisomy 13 and/or 18 may be considered medically necessary in women who are eligible for and are undergoing nucleic acid sequencing-based testing of maternal plasma for

	trisomy 21. In addition, 2 investigational statements were added, 1 for nucleic acid sequencing-based testing of maternal plasma for trisomy 13 and/or 18, other than in the situations specified in the medically necessary statement and the other for fetal sex chromosome aneuploidies.
08/03/2015	Coding update: ICD10 Diagnosis code section added; ICD9 Procedure code section removed.
02/04/2016	Medical Policy Committee review
02/17/2016	Medical Policy Implementation Committee approval. Title change. Testing for microdeletions added to the policy.
01/01/2017	Coding update: Removing ICD-9 Diagnosis Codes
02/02/2017	Medical Policy Committee review
02/15/2017	Medical Policy Implementation Committee approval. Coverage eligibility unchanged.
10/01/2017	Coding update
02/01/2018	Medical Policy Committee review
02/21/2018	Medical Policy Implementation Committee approval. Coverage eligibility unchanged. Policy Guidelines section added to the policy.
03/07/2019	Medical Policy Committee review
03/20/2019	Medical Policy Implementation Committee approval. Added a statement for when nucleic acid sequencing–based testing of maternal plasma for trisomy 13 and/or 18 is investigational.
09/09/2019	Coding update
03/05/2020	Medical Policy Committee review
03/11/2020	Medical Policy Implementation Committee approval. Coverage eligibility unchanged.
06/10/2020	Coding update
11/05/2020	Medical Policy Committee review
11/11/2020	Medical Policy Implementation Committee approval. Title changed from "Noninvasive Prenatal Screening for Fetal Aneuploidies and Microdeletions Using Cell-Free Fetal DNA" to "Noninvasive Prenatal Screening for Fetal Aneuploidies and Microdeletions, and Twin Zygosity Using Cell-Free Fetal NA". Added an investigational statement for noninvasive prenatal testing for twin zygosity. Added investigational statement for noninvasive prenatal testing using Vanadis NIPT.

Noninvasive Prenatal Screening for Fetal Aneuploidies and Microdeletions, and Twin Zygosity Using Cell-Free Fetal DNA

Policy # 00345
Original Effective Date: 12/20/2013
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04/01/2021 Medical Policy Committee review
04/14/2021 Medical Policy Implementation Committee approval. Revised the eligible for coverage statement to cover nucleic acid sequencing-based testing of maternal plasma to screen for trisomy 21, 18, and 13 in women with singleton pregnancies. Removed the Not Medically Necessary section. All investigational statements remain unchanged.
09/30/2021 Coding update
Next Scheduled Review Date: 04/2022

Coding

The five character codes included in the Blue Cross Blue Shield of Louisiana Medical Policy Coverage Guidelines are obtained from Current Procedural Terminology (CPT®)‡, copyright 2020 by the American Medical Association (AMA). CPT is developed by the AMA as a listing of descriptive terms and five character identifying codes and modifiers for reporting medical services and procedures performed by physician.

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Noninvasive Prenatal Screening for Fetal Aneuploidies and Microdeletions, and Twin Zygosity Using Cell-Free Fetal DNA

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Codes used to identify services associated with this policy may include (but may not be limited to) the following:

Code Type	Code
CPT	0060U, 81420, 81422, 81479, 81507, 81599 Code deleted eff 10/1/2021: 0168U
HCPCS	No codes
ICD-10 Diagnosis	O09.511, O09.512, O09.513, O09.519, Z31.5, Z36.0-Z36.9

*Investigational – A medical treatment, procedure, drug, device, or biological product is Investigational if the effectiveness has not been clearly tested and it has not been incorporated into standard medical practice. Any determination we make that a medical treatment, procedure, drug, device, or biological product is Investigational will be based on a consideration of the following:

A. Whether the medical treatment, procedure, drug, device, or biological product can be lawfully marketed without approval of the U.S. Food and Drug Administration (FDA) and whether such approval has been granted at the time the medical treatment, procedure, drug, device, or biological product is sought to be furnished; or
B. Whether the medical treatment, procedure, drug, device, or biological product requires further studies or clinical trials to determine its maximum tolerated dose, toxicity, safety, effectiveness, or effectiveness as compared with the standard means of treatment or diagnosis, must improve health outcomes, according to the consensus of opinion among experts as shown by reliable evidence, including:
    1. Consultation with the Blue Cross and Blue Shield Association technology assessment program (TEC) or other nonaffiliated technology evaluation center(s);
    2. Credible scientific evidence published in peer-reviewed medical literature generally recognized by the relevant medical community; or
    3. Reference to federal regulations.

**Medically Necessary (or "Medical Necessity") - Health care services, treatment, procedures, equipment, drugs, devices, items or supplies that a Provider, exercising prudent clinical judgment, would provide to a patient for the purpose of preventing, evaluating, diagnosing or treating an illness, injury, disease or its symptoms, and that are:

A. In accordance with nationally accepted standards of medical practice;
B. Clinically appropriate, in terms of type, frequency, extent, level of care, site and duration, and considered effective for the patient's illness, injury or disease; and
C. Not primarily for the personal comfort or convenience of the patient, physician or other health care provider, and not more costly than an alternative service or sequence of services at least as likely to produce equivalent therapeutic or diagnostic results as to the diagnosis or treatment of that patient's illness, injury or disease.

For these purposes, "nationally accepted standards of medical practice" means standards that are based on credible scientific evidence published in peer-reviewed medical literature generally recognized by the relevant medical community, Physician Specialty Society recommendations and the views of Physicians practicing in relevant clinical areas and any other relevant factors.

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NOTICE: If the Patient's health insurance contract contains language that differs from the BCBSLA Medical Policy definition noted above, the definition in the health insurance contract will be relied upon for specific coverage determinations.

NOTICE: Medical Policies are scientific based opinions, provided solely for coverage and informational purposes. Medical Policies should not be construed to suggest that the Company recommends, advocates, requires, encourages, or discourages any particular treatment, procedure, or service, or any particular course of treatment, procedure, or service.