

# Non-Invasive Prenatal Testing (NIPT) Factsheet



## Introduction

NIPT, which analyzes cell-free fetal DNA circulating in maternal blood, is a new option in the prenatal screening and testing paradigm for trisomy 21 and a few other fetal chromosomal aneuploidies. (For more information about current screening/testing options, see Background.)

## NIPT Test Characteristics

### Genetic testing using cell-free fetal DNA

DNA from the fetus circulates in maternal blood. Unlike intact fetal cells in maternal blood, which can persist for years after a pregnancy, circulating cell-free fetal DNA (ccfDNA) results from the breakdown of fetal cells (mostly placental) and clears from the maternal system within hours. Fetal DNA detected during a pregnancy, therefore, represents DNA from the current fetus.

Although only about 10-15% of the cell-free DNA circulating in maternal blood is from the fetus, it can be detected and measured. Quantitative differences in chromosome fragments in maternal blood can be used to distinguish fetuses affected with trisomy 21, and a few other fetal aneuploidies, from those that are not affected.

### When NIPT is performed

Testing can be done any time after 10 weeks; typically it is done between 10-22 weeks. Results can take a week or more.

### Maternal indications for NIPT

NIPT technologies have been validated in singleton pregnancies at high risk for trisomy 21 due to:

- advanced maternal age
- an abnormal serum screen
- personal or family history of aneuploidy
- abnormal ultrasound

At least one laboratory will accept samples that do not meet these high risk criteria. Additionally, at least one laboratory offers the test in twin pregnancies, and another for Turner syndrome (monosomy X) when the fetus presents with a cystic hygroma. Contact individual laboratories for additional information.

### Risk

The testing is non-invasive, involving a maternal blood draw, so the pregnancy is not put at risk for miscarriage or other adverse outcomes associated with invasive testing procedures.

## NIPT Detection Rate and Accuracy

At present, NIPT provides information about specific fetal aneuploidies.

All laboratories offering NIPT report on trisomy 21 and trisomy 18. Others may also provide information about trisomy 13, or some sex chromosome abnormalities. NIPT does not typically provide any other genetic information about the genetic constitution of the mother or fetus.

### Detection rate for trisomy 21

At least 99% of all pregnancies with trisomy 21 can be detected using this test. However, up to 1 in 100 pregnancies with trisomy 21 will have a normal result and be missed on screening.

The false-positive rate is approximately 0.2%, meaning 1 in about 500 unaffected

pregnancies are reported as 'positive' or 'consistent with' trisomy 21. For that reason, it is recommended that CVS or amniocentesis be considered after an abnormal result to confirm the presence of a chromosome abnormality.\*

### Detection rate for trisomy 18

The detection rate for trisomy 18 may be similar to that for trisomy 21; approximately 99% of pregnancies with trisomy 18 will be detected by NIPT. About 1 in 100 pregnancies with trisomy 18 will be missed on screening.

The false-positive rate is also similar to trisomy 21. About 1 in 500 pregnancies unaffected with trisomy 18 will have an abnormal, or positive, result, so confirmatory testing is recommended.\*

### Detection of trisomy 13 and sex chromosome abnormalities

There is less confidence in NIPT as a screen for trisomy 13 due to technical issues and the infrequency of the condition. Detection rates between 79-92% have been reported, meaning between 8 to 21 out of 100 pregnancies with affected fetuses will be missed. The false-positive rate may be about 1%, so 1 out of 100 unaffected pregnancies may be positive for trisomy 13, so confirmatory testing is recommended.\*

Some laboratories may report results for sex chromosomes abnormalities. If testing for sex chromosome abnormalities is desired, contact the NIPT laboratory.

*\*(Actual numbers will vary. Check with the specific laboratory)*

## WHAT THIS RESOURCE CAN HELP YOU ACCOMPLISH:

This resource is designed to help you:

- Apply NIPT appropriately in the general prenatal screening/testing paradigm for trisomy 21
- Critically evaluate NIPT tests and laboratories
- Counsel women about their screening/testing options and interpret and counsel about results
- Anticipate future directions of the technology

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## NIPT Reporting

### General

One form of NIPT known as massively parallel shotgun sequencing (MPSS) typically measures the proportion of fragments from targeted maternal and fetal chromosomes in maternal blood and compares the proportion to what is expected for that chromosome. Another methodology measures selected regions of the targeted chromosome and compares observed to expected results. Each laboratory may use proprietary algorithms and specific cut-offs to determine the amount.

Some laboratories report the result as 'positive' or 'negative', or 'consistent with' trisomy 21, 18, or 13. Others may report a quantitative risk for each trisomy. The report should reflect whether a laboratory distinguishes between results close to, or distant from, the cut-off (in either direction). There is typically less confidence in results close to the cut-off, so those results could be more difficult to interpret. Because of this some laboratories may choose to report only those results in which they have near 100% confidence, and those near the cutoff are reported as unclassifiable.

### Implications of a positive result

NIPT is highly sensitive and specific for trisomies 21 and 18; positive results are 'near diagnostic'. However, false positives have been reported so at this time it is recommended that positive results be followed with confirmatory testing by CVS or amniocentesis. Confirmatory testing can also provide important information about the cause of the trisomy; specifically, CVS or amniocentesis will identify cases of Down syndrome that are due to a 21 chromosome translocation as opposed to the more common trisomy 21. This has important recurrence risk implications for the parents and other family members.

Fetal anatomic ultrasound can also be a helpful tool for pregnancies that test positive on NIPT, looking for additional ultrasound findings that support the diagnosis.

### Implications of a negative result

Even though NIPT is highly sensitive and specific, it is important to remember that it is not 100%. There are false-negative results, so a negative result cannot absolutely rule out an affected fetus. A laboratory may provide a risk score, allowing the clinician to quantify risk for trisomy.

### Implications of an 'unreportable' or 'no-call' result

Depending on the laboratory, 0.5-7% of women who undergo NIPT will not get a result, often because there is an insufficient amount of fetal DNA in the sample (low fetal fraction) due to various clinical reasons which may include high maternal weight or early gestational age. Or, as noted above, a laboratory may decline to report results near the cutoff. In any case, the clinician must determine, in conjunction with the NIPT

laboratory and the patient, whether to draw another sample later in the pregnancy, revert to conventional serum or ultrasound screening, move on to invasive testing, or decline any further testing.

## Other NIPT Considerations

### Use of NIPT in other groups of women

NIPT has not been validated in low risk women (although these studies are expected soon), triplet or other higher order multiple pregnancies, or in pregnancies conceived using egg donation.

### Sample processing

Special tubes are required. The specific sample processing procedures will depend on the laboratory. Contact the NIPT laboratory to obtain testing kits and processing instructions.

### Turnaround time

Results may take a week or more. Check with the specific laboratory.

### Cost and insurance coverage

As this is a new test, insurance coverage can be variable. Testing could involve significant out-of-pocket costs in the hundreds of dollars to the patient. Providers and patients should discuss laboratory and coverage policies with the laboratory they are using. Some laboratories may have financial support available for the patient.

### Screening for open neural tube defects (ONTD; spina bifida, anencephaly)

Unlike second trimester maternal serum screening, NIPT does not screen for open neural tube defects (ONTD). Therefore, maternal serum alpha-fetoprotein screening and/or an anatomic fetal ultrasound should still be offered in the second trimester to detect these conditions.

## What to Ask Laboratories Offering NIPT

NIPT technologies are relatively new and the number of laboratories currently offering testing is limited. Additionally, there is variability in the conditions, requirements, and reporting procedures between laboratories.

When selecting a laboratory ask about:

- technology used by the laboratory
- eligibility requirements
- validation data
- how results near the cut-off are reported
- availability of sample reports
- patient information brochures
- financial support for patients

Some laboratories require that the patient and/or provider document informed consent before the test will be resulted. Informed consent is an important step in the genetic testing process. In addition to being required by law in some states and by some laboratories, reviewing informed consent and pre-test counseling information can help prepare the patient when discussing testing results.

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## NIPT Performance Comparison

### Patient selection of NIPT versus other forms of prenatal screening or testing

All forms of prenatal screening and testing for fetal genetic conditions have benefits and limitations with respect to risk to the pregnancy, what can be detected, and accuracy. The selection of which screening or testing paradigm to use should be made in consultation with the patient, and in keeping with her values.

(SEE COMPARISON TABLE FOR SPECIFICS)

### NIPT healthcare costs compared to other screening and testing paradigms

There have been limited cost-benefit analyses done thus far comparing NIPT to other screening or testing alternatives. One study predicted a 1% cost reduction in the detection of aneuploidy using NIPT, as well as 66% reduction in procedure-related miscarriages and a 38% increase in trisomy 21 diagnoses.

## Counseling Women About Screening/Testing Options

Women making decisions about whether to undergo screening or testing for Down syndrome during pregnancy consider many factors in their decision – concern about having a child with an abnormality, concern about putting the pregnancy at risk for miscarriage, what they would do with the results, family values, religious beliefs, family and life situation, and others.

Information used to counsel women about options should be sensitive to all of these issues and present accurate and balanced information about the risks and benefits of each screening or diagnostic testing methodology. Up-to-date, balanced information about what it means to have a child with one of these conditions should be available. Genetic counseling to help patients understand the options, including the option of not having any screening or testing at all, may be helpful to many women.

For women who prefer to start with screening, there is a significant difference in the predictive power of the information obtained from NIPT as compared to other screening approaches, and this impacts pre-test counseling. Serum screening typically allows a screen-positive woman to consider whether she wants the certainty associated with the next step in the paradigm, diagnostic testing. However, the sensitivity and specificity of NIPT is close to diagnostic, and an NIPT-positive result may imply an overwhelming likelihood of trisomy; CVS or amniocentesis are performed for confirmation.

## Future Directions of NIPT

The number of pregnancies for which there is NIPT data is still relatively small. As the population of patients who elect the test grows, experience in the performance of the test will increase, especially in sub-populations of pregnancies. Validation studies are underway in 'low risk' women and

results should be available within a year.

It is expected that labs will continue to explore the number of conditions that can be detected using circulating cfDNA. This could include using other testing technologies to interrogate the fetal DNA, such as chromosome microarray testing to detect microdeletion and duplication syndromes.

Legal issues have been raised regarding patent rights surrounding the technology being used by several laboratories. It is unclear at this time how these issues will be resolved.

## Professional Society Statements

Professional societies are beginning to make statements about the use of NIPT. ACOG (American College of Obstetricians and Gynecologists) recommends offering aneuploidy screening or invasive testing to all women, regardless of age. ACOG as yet has not made any statement about the use of NIPT specifically as an option in the screening/testing paradigm.

### ISPD (International Society of Prenatal Diagnosis)

ISPD recognizes that NIPT can be helpful as a screening test for women who are at high risk for Trisomy 21 with suitable genetic counseling. A positive test should be confirmed through invasive testing.

### NSGC (National Society of Genetic Counselors)

NSGC supports NIPT as an option for patients whose pregnancies are considered to be increased risk for certain chromosome abnormalities. NSGC urges that NIPT only be offered in the context of informed consent, education, and counseling by a qualified provider, such as a certified genetic counselor. Patients whose NIPT results are abnormal, or who have other factors suggestive of a chromosome abnormality, should receive genetic counseling and be given the option of standard confirmatory diagnostic testing.

## FDA

At this time NIPT is offered as a laboratory-developed test (LDT) which is not being reviewed by FDA.

## General Background on Screening for Down Syndrome and Other Fetal Aneuploidies

In the United States, women presenting for prenatal care in the first or second trimester are offered the option of screening or testing for Down syndrome, also called trisomy 21. This screening can also provide information about two other much less common chromosome aneuploidies, trisomy 18 and 13.

Women who are interested in learning if they carry a fetus with trisomy 21 should have the opportunity to examine the benefits, limitations, and risks of the screening and testing options available to them, balancing diagnostic certainty against risk, weighing their desire to know against their desire to avoid risk.

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## Current Screening Technologies: Maternal Serum Screening

Current screening technologies generally involve measuring biochemical markers associated with trisomy 21, as well as trisomy 18 and 13, in maternal serum during the first and/or second trimester. An ultrasound measuring fetal nuchal translucency (NT) is often included in a first trimester screen as an additional marker of trisomy 21. After first or second trimester screening, the woman typically receives a revised risk for trisomy 21 that is calculated based on her maternal age risk, the results of the serum screen, and the NT measurement (if available).

An advantage of screening is that it is non-invasive, thus posing no risk to the fetus. Only women whose revised risk exceeds a laboratory and test-dependent cut-off are candidates for diagnostic testing by chorionic villus sampling or amniocentesis. An inherent limitation of this approach is that screening only detects 85-95% of fetuses with trisomy 21, and falsely designates 3-6% of pregnancies as "positive" when they are, in fact, unaffected with trisomy 21. For every trisomy 21 fetus detected, perhaps 25 will be subjected to the risk of an invasive procedure.

## Current Testing Technologies: Chorionic Villus Sampling (CVS) or Amniocentesis

An alternative to screening is invasive prenatal diagnosis by chorionic villus sampling (CVS) or amniocentesis which directly assesses the chromosome constitution of the fetus through cells from the pregnancy. The advantage is the diagnostic certainty of detecting trisomy 21, 18, and 13. In addition, testing fetal cells and the amniotic fluid may allow for the detection of other chromosome abnormalities, genetic conditions, or open neural tube defects. The downside is that these procedures are associated with a measurable risk of a procedure-related miscarriage or other adverse pregnancy outcome.

## Publication Acknowledgements

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