DATA SHEET

VeriSeq™ NIPT Solution v2

An end-to-end, accessible, whole-genome sequencing assay

- Comprehensive view of fetal chromosomes with a broad test menu validated in a clinical accuracy study of > 2300 samples
- Trusted test performance[1] with high accuracy, fast results, and low failure rates
- Simple, scalable IVD solution that can analyze 24, 48, or 96 samples per run

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For *In Vitro* Diagnostic Use. Not available in all regions and countries.

Introduction

Noninvasive prenatal testing (NIPT) performed with next-generation sequencing (NGS) provides reliable screening results for fetal chromosomal aneuploidies as early as 10 weeks gestation—from a single tube of maternal blood.[2,3] VeriSeq NIPT Solution v2 takes advantage of powerful Illumina NGS technology to bring a whole-genome sequencing (WGS) approach to NIPT, expanding test menu options to include common aneuploidies (trisomies 21, 18, and 13), rare autosomal aneuploidies (RAAs), select sex chromosome aneuploidies (SCAs), and partial deletions and duplications, referred to as copy number variations (CNVs), that are ≥ 7 Mb.

By combining a broad test menu, accurate results, and low failure rates, the VeriSeq NIPT Solution v2 offers comprehensive screening of fetal chromosomes, enabling informed, timely pregnancy management decisions.[1] Providing reagents, instruments, software, installation, and training, the VeriSeq NIPT Solution v2 is an automated, reliable solution for in-house NIPT (Figure 1 and Table 1).

Full view of fetal chromosomes

Many in-lab NIPT solutions focus on screening for trisomies in chromosomes 21, 18, and 13, but these conditions represent only a portion of anomalies that can occur. These tests will miss CNVs ≥ 7 Mb that can be associated with fetal anomalies and developmental delay and have a screen positive rate of 0.12% with NIPT.[4] These tests will also miss pregnancies that screen positive for RAAs, which may be associated with adverse outcomes, including miscarriage, intrauterine growth restriction (IUGR), uniparental disomy (UPD), spontaneous preterm labor, and fetal anomalies, among others.[5] The combined screen positive rate for RAAs is 0.34%,[5] compared to 0.30% for trisomy 21.[6,7]

Table 1: VeriSeq NIPT Solution v2, at a glance

Parameter	Description
Method	Whole-genome sequencing
Library prep	PCR-free
Chemistry	Paired-end sequencing
No. of samples	24, 48, or 96 per batch
Time to report	~26 hours
No. of operators	1
Specimen	7–10 mL of a single tube of maternal blood
Analysis offered	Aneuploidy status of all autosomes and sex chromosomes; CNVs ≥ 7 Mb

Trusted test performance

Based on accuracy of results, time to answer, and failure rates, the VeriSeq NIPT Solution v2 demonstrates excellent performance.

High accuracy

VeriSeq NIPT Solution v2 is validated to ensure clinical accuracy and reliability. Samples from affected pregnancies were eligible for testing if clinical outcomes were available and met sample inclusion criteria. The cohort comprised gestational ages of at least 10 weeks, samples with low fetal fractions, and twin pregnancies. The study screened over 2300 maternal samples with

Figure 1: Full IVD NIPT workflow—The VeriSeq NIPT Solution v2 provides everything needed for NIPT using NGS, including reagents for DNA extraction, library prep, and sequencing; instrumentation to automate library prep and sequencing with workflow manager software; an onsite server for secure data storage and analysis; and data analysis software that generates qualitative result reports.

Table 2: Clinical performance of VeriSeq NIPT Solution v2[1]

	Trisomy 21[c]	Trisomy 18	Trisomy 13	RAA[d]	CNVs ≥ 7 Mb	Any anomaly[e]
Sensitivity[a]	> 99.9% (130/130)	> 99.9% (41/41)	> 99.9% (26/26)	96.4% (27/28)	74.1% (20/27)	95.5% (318/333)
2-sided 95% CI[b]	97.1%, 100%	91.4%, 100%	87.1%, 100%	82.3%, 99.4%	55.3%, 86.8%	92.7%, 97.3%
Specificity	99.90% (1982/1984)	99.90% (1995/1997)	99.90% (2000/2002)	99.80% (2001/2005)	99.80% (2000/2004)	99.34% (1954/1967)
2-sided 95% CI[b]	99.63%, 99.97%	99.64%, 99.97%	99.64%, 99.97%	99.49%, 99.92%	99.49%, 99.92%	98.87%, 99.61%

a. Basic screen performance is reported for T21, T18, and T13 and excludes 16 samples with known mosaics and 49 samples affected with anomalies for the genome-wide screen only; genome-wide screen performance is reported for RAAs and partial duplications and deletions.
b. CI based on Wilson's score method.
c. Seven twin pregnancies reported correctly as T21 are not shown.
d. RAA excludes chromosomes 21, 18, and 13.
e. Any anomaly includes samples from SCA basic and genome-wide screens.

Table 3: Concordance of VeriSeq NIPT Solution v2 fetal sex classification results with clinical reference[1]

VeriSeq NIPT Solution v2 results	Newborn physical outcome		Cytogenetic results					
	Female	Male	XX	XY	XO	XXX	XXY	XYY
Percent concordant	100%	100%	100%	100%	90.5%	100%	100%	91.7%

known outcomes for trisomy 21, trisomy 18, trisomy 13, RAAs, CNVs ≥ 7 Mb, and SCAs. Results using the VeriSeq NIPT Solution v2 were compared to clinical reference truth. VeriSeq NIPT Solution v2 demonstrated high sensitivity and specificity for common trisomies, RAAs, CNVs ≥ 7 Mb, high concordance of fetal sex classification with clinical outcome, and a low first-pass sample failure rate of 1.2% (Table 2 and Table 3).[1]

Fast results

The VeriSeq NIPT Solution v2 offers a fast three-step workflow that generates accurate results in just over one day (Table 4). Following the simple, automated workflow, one operator can analyze 24–96 samples in less than 8 hours with minimal hands-on time. Targeted sequencing and array-based methods tend to have longer laboratory protocols, requiring more hands-on time.

Table 4: VeriSeq NIPT complete in just over one day

Step	Hands-on time	Total time
Sample prep and library prep	~2 hours	~8 hours
Sequencing	~15 min	~14 hours
Data analysis and report generation	N/A	~4 hours
Total time	~2.25 hours	~26 hours

Actual times depend on individual lab practices and may vary; N/A, not applicable.

Low test failure rates

Test failures, where no call for disomy or aneuploidy can be made, are an important factor in the reliability and clinical utility of NIPT. Failure rates can vary significantly based on the test used. Tests that use a targeted approach or single nucleotide polymorphism (SNP) method demonstrate higher rates of primary test failure compared to NGS.[8] The VeriSeq NIPT Solution v2 uses WGS to provide ample data across all chromosomes, without impacting accuracy or increasing failure or false positive rates. In the clinical validation study, the first pass failure rate was 1.2%.[1] In lab practice, the initial blood draw will provide sufficient plasma to repeat the VeriSeq NIPT workflow if needed.[9] Repeated testing has demonstrated a reduction in the primary failure rate from 2% to 1.3% for the same sample.[9]

Simple, scalable IVD solution

The integrated VeriSeq NIPT Solution v2 provides everything needed to run the assay. The automated workflow easily scales to analyze 24, 48, or 96 samples per run to allow for efficiency and flexibility in managing

sample volumes. Either basic or genome-wide screening can be selected for each sample.

Automated workflow

The fully automated VeriSeq NIPT assay provides a simple workflow that minimizes hands-on time and potential for error. The protocol requires 7–10 mL of maternal peripheral whole blood collected in the recommended Streck Blood Collection Tube (BCT). Optimized VeriSeq NIPT sample prep kits contain reagents and labels for preparing sequencing libraries from cell-free DNA (cfDNA). Plasma isolation, cfDNA extraction, and PCR-free library preparation, including quantification plate creation, library quantification, and library pooling, are automated on the VeriSeq NIPT Microlab STAR, a Hamilton Microlab STAR system custom configured specifically for use in the VeriSeq NIPT workflow. The user-friendly VeriSeq NIPT Workflow Manager controls all aspects of sample preparation, including sample tracking.

Sequencing

A maternal blood sample contains cfDNA fragments of different lengths; longer lengths tend to be maternal while shorter lengths tend to originate from the fetus (Figure 2).[10] The VeriSeq NIPT Solution v2 quickly and efficiently identifies the lengths of all cfDNA fragments within a single sample and focuses analysis on shorter cfDNA using paired-end sequencing performed on the Illumina NextSeq™ 550Dx System, which delivers the power of high-throughput NGS[11] with the affordability of a benchtop system (Table 5).

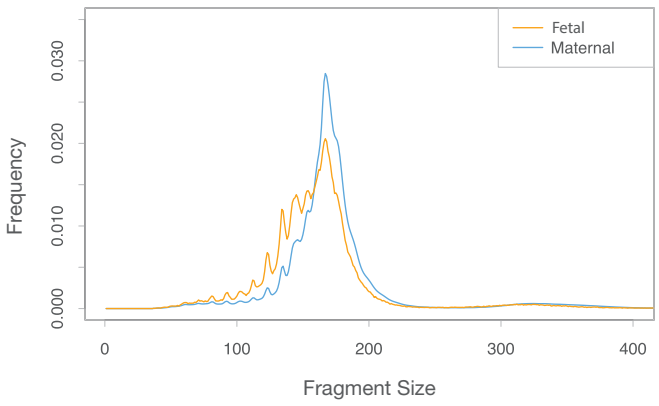

Figure 2: Size comparison of maternal and fetal cfDNA fragments—Paired-end sequencing differentiates cfDNA fragments based on size. Longer fragments tend to be maternal in origin while shorter fragments tend to be fetal.

Table 5: NGS instrument performance requirements

Parameter	Specification
Read length	2 × 36 bp
Sequencing file type	.BCL file
Sequencing output	400M reads
Run time	~14 hours
Multiplexing	24 or 48 samples per run

Onsite analysis

Data analysis is performed on a dedicated VeriSeq v2 Onsite Server with the IVD VeriSeq NIPT Assay Software v2. The server automatically processes sequencing data. Multiple sample batches can be queued for analysis on a single server. There is no need to send out data for analysis, saving time and protecting sample identity.

VeriSeq NIPT Assay Software v2

VeriSeq NIPT Assay Software v2 filters and aligns the reads to a reference genome. An advanced algorithm determines the read density per chromosome (segment) and aids in the detection and differentiation of aneuploidy and CNVs. The software also generates and reports a fetal fraction estimate for each sample. Fetal fraction data are combined with coverage and other statistical inputs generated during sequencing to assess aneuploidy status.

To ensure low test failure rates, VeriSeq NIPT Assay Software v2 includes the individualized fetal aneuploidy confidence test (iFACT) sample quality scoring metric. iFACT indicates whether the assay has generated sufficient sequencing coverage, given the fetal fraction estimate for each sample, to enable an aneuploidy or partial duplication and deletion call, even for samples with low fetal fraction.[12] This dynamic cutoff enables VeriSeq NIPT Assay Software v2 to report on low fetal fraction samples, resulting in low test failures.[1]

Report generation

After data analysis, the VeriSeq NIPT Assay Software generates an “Aneuploidy Detected” or “No Aneuploidy Detected” result for the chromosomes tested in each sample. If a CNV is detected, the exact coordinates in the genome are displayed in the report. Data are provided as a ".CSV" file that can be integrated with an existing LIMS, enabling the creation of a custom clinical report.

Fully supported implementation

For smooth laboratory integration, the VeriSeq NIPT Solution v2 includes complete system installation by a skilled Illumina Field Service Engineer and hands-on training. Illumina scientists lead laboratory personnel step by step through sample extraction, library preparation, sequencing, and analysis (Table 6). When all systems are up and running, continued support is provided by the Illumina Technical Support team.

Table 6: VeriSeq NIPT Solution v2 training

Topic	Details
Introduction to the VeriSeq NIPT Solution v2	Seminar overview of workflow and analysis • Ancillary equipment guide • Consumables guide • Blood draw protocol • Plasma isolation protocol
Instrument operation training	Onsite training • Requires installed instrument
Site inspection	Onsite confirmation • Ancillary equipment installation • Needed reagents • Connectivity of system components
Onsite training	Assay performed by Illumina scientist • Pretested plasma samples with known performance characteristics (provided by Illumina) • Walkthrough of assay workflow from plasma isolation to instrument operation and data analysis • Data analysis training
Onsite competency testing	Assay performed by customer • Pretested plasma samples with known performance characteristics (provided by Illumina)

Summary

VeriSeq NIPT Solution v2 revolutionizes the accessibility, reliability, and power of NIPT. Now laboratories can harness NGS for fast, reliable, highly accurate NIPT results with low failure rates.

Learn more

Illumina VeriSeq NIPT Solution v2

Ordering information

Product	Catalog no.
VeriSeq NIPT Sample Prep Kit (24 samples)	20025895
VeriSeq NIPT Sample Prep Kit (48 samples)	15066801
VeriSeq NIPT Sample Prep Kit (96 samples)	15066802
VeriSeq NIPT Assay Software v2	20047024
VeriSeq Onsite Server v2	20047000 20101927
Streck cell-free DNA BCT (CE)	15073345
NextSeq 550Dx Instrument	20005715
NextSeq 550Dx High Output Reagent Kit v2.5, 75 cycles	20028870

Intended use statement

The VeriSeq NIPT Solution v2 is an *in vitro* diagnostic test intended for use as a screening test for the detection of genome-wide fetal genetic anomalies from maternal peripheral whole blood specimens in pregnant women of at least 10 weeks gestation. VeriSeq NIPT Solution v2 uses WGS to detect partial duplications and deletions for all autosomes and aneuploidy status for all chromosomes. The test offers an option to request the reporting of SCA. This product must not be used as the sole basis for diagnosis or other pregnancy management decisions.

The VeriSeq NIPT Solution v2 includes: the VeriSeq NIPT Workflow Manager v2 for the VeriSeq NIPT Microlab STAR, the VeriSeq NIPT Sample Prep Kits, and the VeriSeq Onsite Server v2 with the VeriSeq NIPT Assay Software v2. The VeriSeq NIPT Solution v2 is intended to be used with a next-generation sequencer.

References

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