# illumına<sup>®</sup>

## NextSeq<sup>®</sup> Series WGS Solution

An accessible, high-quality whole-genome sequencing solution for any species.

#### Highlights

- High-Quality, High-Coverage Genome Illumina chemistry offers highest read quality and alignability, with best coverage of GC-rich regions
- Unrestricted View of the Genome Now and in the Future Rich data set can be mined repeatedly as new discoveries are made
- Scalable, Affordable WGS Rapid and cost-effective sequencing of complex genomes in your laboratory
- Easy, Accessible Data Analysis and Storage Solutions Rich ecosystem of user-friendly informatics tools to analyze and interpret data
- End-to-End Illumina Support Illumina scientists and engineers are there every step of the way, providing installation, training, applications, and data analysis support

#### Introduction

The NextSeq Series Whole-Genome Sequencing (WGS) Solution enables researchers and clinicians to explore the entire genome of any species cost-effectively for a deeper understanding of biology. It leverages industry-standard Illumina next-generation sequencing (NGS) technology responsible for most global WGS, delivering the best data quality and highest coverage to identify variants in coding and noncoding regions of the genome. High-quality library preparation kits are optimized for low-input, unbiased coverage, and rapid workflow. With push-button sequencing, simple data analysis, and minimal hands-on time, the NextSeq Series WGS Solution enables researchers to interrogate simple prokaryotic and complex eukaryotic genomes quickly and efficiently.



Figure 1: NextSeq Series Sample-to-Answer WGS Sequencing Workflow – The NextSeq Series features a simple workflow that delivers highly accurate sequencing data. Data analysis includes alignment and variant calling.

## A Fast, Efficient WGS Workflow

The NextSeq Series WGS workflow offers a simple, fast, end-to-end solution for characterization of any genome (Figure 1). It begins with streamlined library preparation using high-quality TruSeq<sup>®</sup> library preparation kits. The NextSeq Series offers dual density flow cell configurations and accompanying reagent kits deliver 20–120 Gb, enabling researchers to match sequencer output with sequencing of prokaryotic and eukaryotic genomes. New NextSeq v2 reagent kits are optimized to improve base calling and data quality even further.

Data analysis, from alignment and variant calling to annotation and beyond, can be performed in BaseSpace®, the Illumina genomics computing environment. Utilizing the Illumina industry-leading NGS workflow, the NextSeq Series provides access to the world's largest collection of commercial and open-source data analysis software tools (Figure 2).



Figure 2: NextSeq System – The NextSeq Series of sequencing systems leverages the latest advances in SBS chemistry and the industry's simplest workflow.

The NextSeq Series also offers cross-application flexibility, enabling researchers to transition easily between sequencing projects (Figure 3). The system is fully compatible with the industry's widest range of library preparation kits from Illumina and third parties, enabling an easy transition between Illumina WGS, exome, and RNA-Seq. For example, researchers can follow up WGS with exome sequencing to obtain much deeper coverage and increased ability to call rare variants in coding portions of the genome. They can also pair WGS with RNA-Seq to assess whether the coding and noncoding variants they just identified interfere with transcript expression. A wide range of Illumina targeted resequencing solutions are also available to validate variants discovered from any sequencing application. With the NextSeq 550 System, researchers can perform NGS and array scanning on the same platform for further exploration or confirmation of copy number variants detected through sequencing.

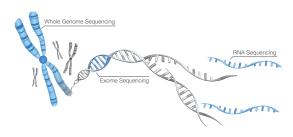


Figure 3: NextSeq Series Sequencing Applications—The flexible NextSeq Series enables researchers to transition seamlessly between applications to advance their research.

### Simple, Rapid Library Preparation

Illumina offers various library preparation kits to accommodate a range of sample types and genome sizes. These kits have been developed and tested for Illumina systems and include everything needed to prepare libraries. TruSeq DNA library preparation kits deliver unsurpassed data quality with fast, streamlined workflows that require minimal hands-on time (Table 1).

#### Table 1: Illumina WGS Library Preparation Kits

Specification	TruSeq DNA PCR-Free	TruSeq Nano DNA
Sample DNA input type	Genomic DNA	Genomic DNA
WGS applications	Human or other large, complex genomes	Human or other large, complex genomes
Input DNA	1–2 µg	100–200 ng
Typical median insert size	350 bp	350 bp
Read lengths supported	All read lengths	All read lengths
DNA libraries	Single, paired-end, and indexed sequencing	Single, paired-end, and indexed sequencing
Assay time	~ 6 hours	~ 6 hours
Hands-on time	~ 4 hours	~ 5 hours

## Fully Characterized, Highly Accurate Genomes of Any Species

The NextSeq Series delivers the power of high-throughput sequencing with the simplicity of a desktop sequencer, enabling researchers to scale their WGS studies from 1 to hundreds of genomes. Its dual sequencing output modes and accompanying reagents deliver 20–120 Gb, enabling researchers to fine-tune their WGS studies to optimize for sample volume and coverage needs. These desktop systems deliver a cost-effective whole human genome in a single run with the high data quality researchers demand. Leveraging Illumina NGS technology, the NextSeq Series delivers industry-leading sequencing accuracy of > 75% of sequenced bases over Q30.\*

The NextSeq Series delivers a rich data set that can be reanalyzed in the future to provide an up-to-date view of the genome as new gene variant discoveries are made. It can successfully sequence even the most difficult regions (eg, GC-rich, homopolymers) delivering the data output and quality necessary to characterize a genome fully. Paired-end sequencing on a NextSeq System provides even greater resolution, enabling the detection of structural variants such as deletions, duplications, and large copy number variants that play a role in disease.

Based on industry-leading Illumina sequencing by synthesis chemistry, the NextSeq Series enables researchers to compare and integrate the data it generates with data from studies performed on other Illumina systems. For example, NextSeq System WGS data can be integrated with data from follow-up studies on the Illumina MiSeq® System or large-scale WGS sequencing studies run on an Illumina HiSeq® System (Table 2).

#### Table 2: Illumina WGS Solutions

Illumina Sequencing System	Read Length	Output	No. of Samples	
NextSeq Series				
Mid-Output Flow Cell (130 M)	2 × 150 bp	40 Gb	Up to 24 small genomes	
High-Output Flow Cell (400 M)	2 × 150 bp	120 Gb	1 x 30× human genome	
HiSeq 2500 System				
Rapid-Run Mode	2 × 100 bp	100–120 Gb	1 x 30× human genome	
High-Output Mode	2 × 125 bp	900 Gb-1 Tb	8 x 30× human genome	

### Simplified Bioinformatics in BaseSpace

Illumina has removed much of the complexity from the typical informatics workflow. Bases generated on a NextSeq System are instantly and securely transferred, stored, and analyzed in BaseSpace (Cloud or Onsite), delivering annotated variants in as little as 12 hours using the Illumina Isaac pipeline<sup>1</sup> or 45 hours using the industry-standard BWA/GATK method. Analytic tools from NextBio<sup>®</sup> (an Illumina company) can be used to annotate and filter variants, as well as integrate and interpret genomic data in the context of other molecular and phenotypic data. Analysis results, including coverage statistics and annotated SNPs and indels, are presented in intuitive, easy-to-interpret reports.

The BaseSpace environment also includes BaseSpace apps, a growing community of software solutions for visualization, analysis, and sharing. Because Illumina NGS technology is the most established and broadly adopted sequencing solution, researchers can also take advantage of the largest collection of WGS analysis software available. Through its intuitive user interface and a rich ecosystem of commercial and third-party tools and apps, BaseSpace enables researchers to transform raw data into biologically meaningful results (Figure 4).

<sup>\*</sup> Q30 = 1 error in 1000 base calls or an accuracy of 99.9%

Flow Cell Configuration	Read Length (bp)	Output (Gb)	Run Time	Data Quality	Required Input
High-Output Flow Cell Up to 400 M single reads Up to 800 M paired-end reads Mid-Output Flow Cell Up to 130 M single reads Up to 260 M paired-end reads	2 × 150	100-120	29 hours	- - > 75% higher than - Q30 at 2 × 150 bp 100 ng–1 μg with T	100 ng–1 µg with TruSeg Library Prep Kits
	2 × 75	50-60	18 hours		
	1 × 75	25–30	11 hours		
	2 × 150	32.5–39	26 hours		
	2 × 75	16.25–19.5	15 hours	_	

#### Table 3: NextSeq Series Performance Parameters

Total times include cluster generation, sequencing, and base calling on a NextSeq System. Install specifications are based on Illumina PhiX control library at supported cluster densities (between 129 and 165 K/mm<sup>2</sup> clusters passing filter). Actual performance parameters may vary based on sample type, sample quality, and clusters passing filter. The percentage of bases > Q30 is averaged over the entire run.

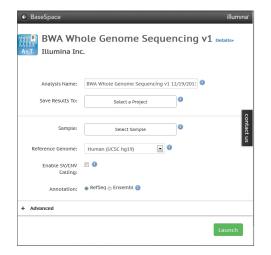


Figure 4: Storage and Analysis of NextSeq Series Data in the BaseSpace Cloud—NextSeq Series data can be securely and seamlessly uploaded to the BaseSpace cloud for fast, cost-effective analysis and storage.

#### Summary

Delivering the highest data quality and accuracy, the NextSeq Series WGS Solution enables researchers to characterize and explore whole genomes. This high-throughput, desktop sequencer can cost-effectively perform WGS of any species, with a streamlined workflow that minimizes hands-on time. Integrated data analysis in BaseSpace, supplemented with a large collection of commercial and open source software tools, enables researchers to mine the rich NextSeq Series WGS data set now and in the future as new discoveries are made.

#### Learn More

Go to www.illumina.com/applications/sequencing/dna\_sequencing/ whole\_genome\_sequencing.html to learn more about the next revolution in WGS.

#### Join the Illumina Community

With a NextSeq System in their laboratory, researchers join a worldwide community of over 60,000 scientists using Illumina technology for their research studies. Illumina schedules community events throughout the year, bringing researchers together to share ideas. User group meetings, scientific symposiums, and blog forums provide venues to discuss new research methods and breakthrough studies.

An integral part of the Illumina community is our dedicated service and support team, consisting of more than 300 people worldwide, 75% of whom have advanced degrees. Illumina technical support begins when the NextSeq System is delivered, with Illumina scientists and engineers assisting with system installation and setup, and the training of laboratory personnel. They are there 24/7 globally to answer questions every step of the way, giving researchers the peace of mind to focus on their next research study.

As researchers' needs change, new systems are brought into the laboratory, or new methods are undertaken, the Illumina support and training teams are there to provide assistance. In addition to on-site support, training courses (via webinar or at an Illumina facility) are available to bring laboratory personnel quickly up to speed.

Table 4: NextSeq Series Specifications	
Instrument Configuration	
RFID tracking for consumables	
Instrument Control Computer (Internal) <sup>a</sup>	
Base Unit: Dual Intel Xeon ES-2448L 1.8 GHz CPU	J
Memory: 96 GB RAM	
Hard Drive: 750 GB	
Operating System: Windows 7 embedded standard	b
Operating Environment	
Temperature: 19°C to 25°C (22°C $\pm$ 3°C)	
Humidity: Noncondensing 20%-80% relative humid	dity
Altitude: Less than 2,000 m (6,500 ft)	
Air Quality: Pollution degree rating of II	
Ventilation: Up to 2,048 BTU/hr @ 600 W	
For Indoor Use Only	
Light Emitting Diode (LED)	
520 nm, 650 nm; Laser diode: 780 nm, Class IIIb	
Dimensions	
W×D×H: 53.3 cm × 63.5 cm × 58.4 cm (21.0 in × 3	25.0 in × 23.0 in)
Weight: 83 kg (183 lbs)	
Crated Weight: 151.5 kg (334 lbs)	
Power Requirements	
100-120 VAC 15 A	
220–240 VAC 10 A	
Radio Frequency Identifier (RFID)	
Frequency: 13.56 MHz	
Power: Supply current 120 mA, RF output power 2	200 mW
Product Safety and Compliance	
NRTL certified IEC 61010-1	
CE marked	
FCC/IC approved	

a. Computer specifications are subject to change.

### **Ordering Information**

System Name	Catalog No.
NextSeq 500 System	SY-415-1001
NextSeq 550 System	SY-415-1002
Output Kit Name	Catalog No.
NextSeq 500 Mid-Output Kit (150 cycles)	FC-102-1001
NextSeq 500 Mid-Output Kit (300 cycles)	FC-404-1003
NextSeq 500 High-Output Kit (75 cycles)	FC-404-1005
NextSeq 500 High-Output Kit (150 cycles)	FC-404-1002
NextSeq 500 High-Output Kit (300 cycles)	FC-404-1004
NextSeq 500 Mid-Output v2 Kit (150 cycles)	FC-404-2001
NextSeq 500 High-Output v2 Kit (150 cycles)	FC-404-2002
NextSeq 500 Mid-Output v2 Kit (300 cycles)	FC-404-2003
NextSeq 500 High-Output v2 Kit (300 cycles)	FC-404-2004
NextSeq 500 High-Output v2 Kit (75 cycles)	FC-404-2005

#### Reference

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Raczy C, Petrovski R, Saunders CT, et al. Isaac: Ultra-fast whole genome secondary analysis on Illumina sequencing platforms. *Bioinformatics*. 2013;29:2041-2043.