illumina®

# NovaSeq™ Series of Sequencing Systems

Scalable throughput and flexibility for virtually any genome, sequencing method, and scale of project.

**Highlights**

- **Scalable Platform**
  Match data output, time to results, and cost per sample to study needs
- **Flexible Performance**
  Configure sequencing method, flow cell type, and read length to support the broadest range of applications
- **Streamlined Operation**
  Increase lab efficiency with a simplified workflow and reduced hands-on time

## Introduction

The NovaSeq Series of Systems unleashes a new era in sequencing with groundbreaking innovations that leverage proven Illumina next-generation sequencing (NGS) technology. Consisting of 2 instrument configurations, the NovaSeq 5000 Sequencing System and NovaSeq 6000 Sequencing System, the NovaSeq Series provides users with the throughput, speed, and flexibility to complete projects faster and more economically than ever before (Figure 1). Multiple flow cell types and read length combinations enable effective scaling of throughput to best suit study needs.

Applications requiring large amounts of data, such as human whole-genome sequencing (WGS), ultradeep exome sequencing, and tumor–normal profiling can now be completed in a more cost-effective manner. The same instrument can be used for less data-intensive methods, such as targeted resequencing. Every project, regardless of the size or goal, will benefit from simple load-and-go operation, integrated onboard cluster generation, and seamless integration with data storage and analysis tools that help streamline the overall experimental workflow. Whether running a single NGS system or a large fleet, the NovaSeq Series opens new possibilities across a range of samples types and applications.

## Scalable Platform

Researchers can use the NovaSeq Series to access the highest levels of throughput now possible more economically than before. The NovaSeq 6000 System, available at launch, powers studies with more samples and higher depth of coverage. The NovaSeq 5000 System* provides access to a powerful, high-throughput genomics solution at a lower entry price. Whatever the scale, the NovaSeq Series empowers users to perform studies at a scale that best matches their research objectives.

**Figure 1: The NovaSeq Series of Systems —** Transforming sequencing by combining throughput, flexibility, and ease of use for virtually any method, genome, and scale.

### Match Data Output to Project Needs

The NovaSeq 6000 System offers tunable output up to 6 Tb and 20 billion (B) reads in ~ 2 days, while the NovaSeq 5000 System generates up to 2 Tb and 1.6 B reads of data in ~2.5 days. (Tables 1 and 2, Figure 2). Multiple flow cell types and read length combinations enable further output and run time configurations based on project needs, enabling cost-effective sequencing with rapid results for a wide range of applications.

NovaSeq S1 and S2 flow cells,* available for use on both systems, provide quick and powerful sequencing for most high-throughput applications. With options for fewer reads than a HiSeq® 2500† or HiSeq 4000 flow cell, the NovaSeq S1 flow cell enables existing HiSeq System users to transition easily, without the need for additional samples per run. The NovaSeq S2 flow cell provides a greater number of reads for users looking for additional sequencing output.

Exclusively available on the NovaSeq 6000 System, the NovaSeq S3 and S4 flow cells* provide high throughput, while enabling cost-effective WGS across a range of species and depths of coverage. Using the NovaSeq S3 or S4 flow cells makes performing WGS in-house attractive and affordable for more labs. The NovaSeq 5000 System is fully upgradable to the NovaSeq 6000 System for future extensibility as the lab grows.*

*The NovaSeq 5000 System, NovaSeq 5000 System Upgrade, and NovaSeq Reagent Kits with S1, S3, or S4 flow cells are not currently available for order.

† Comparison reads for the HiSeq 2500 System based on specifications for the v4 (high output) flow cell.

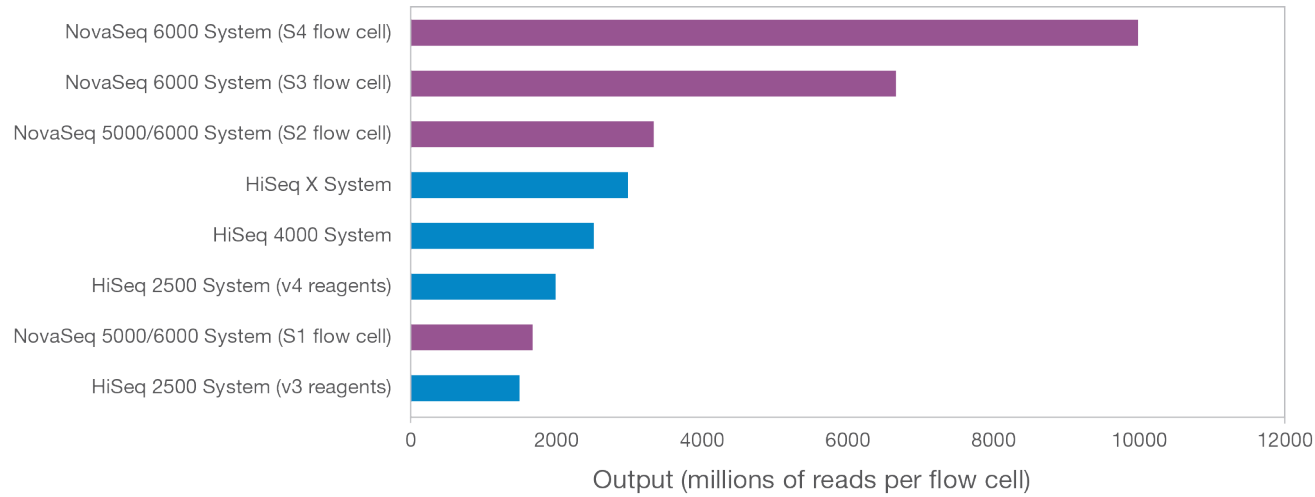


**Figure 2: The NovaSeq Series Offers the Broadest Output Range—** The NovaSeq Series generates from 167 Gb and 1.6 B reads to 3 Tb and 10 B reads of data in single flow cell mode. Note that when run in dual flow cell mode, output can be up to 6 Tb and 20 B reads. The tunable output makes the NovaSeq Series accessible for a wide range of applications. Labs can expand their throughput possibilities with a single instrument.

**Table 1: NovaSeq Series Flow Cell Specifications[a]**

| | NovaSeq 5000[b] and 6000 Systems | | NovaSeq 6000 System | |
|---|---|---|---|---|
| **Flow Cell Type** | S1[b] | S2 | S3[b] | S4[b] |
| **Reads Passing Filter** | up to 1.6 B | 2.8-3.3 B | up to 6.6 B | up to 10 B |
| **Ouput** | | | | |
| 2 × 50 bp | up to 167 Gb | 280-333 Gb | N/A[c] | N/A[c] |
| 2 × 100 bp | up to 333 Gb | 560-667 Gb | N/A[c] | N/A[c] |
| 2 × 150 bp | up to 500 Gb | 850-1000 Gb | up to 2000 Gb | up to 3000 Gb |

a. All output and read number specifications based on a single flow cell. The NovaSeq 5000 and 6000 Systems can run 1 or 2 flow cells simultaneously. Specifications based on Illumina PhiX control library at supported cluster densities.
b. The NovaSeq 5000 System and NovaSeq Reagent Kits with S1, S3, or S4 flow cells have not been released, therefore performance metrics are subject to change.
c. N/A: not applicable.

**Table 2: Performance Specifications of a Dual S2 Flow Cell Run on the NovaSeq 6000 System**

| Read Length | Quality Score (Q30) | Run Time |
|---|---|---|
| 2 × 50 bp | ≥ 85 % | ≤ 19 hr |
| 2 × 100 bp | ≥ 80 % | ≤ 29 hr |
| 2 × 150 bp | ≥ 75 % | ≤ 40 hr |

Quality scores are based on NovaSeq S2 Reagent Kits run on the NovaSeq 6000 System using an Illumina PhiX control library. Performance may vary based on library type and quality, insert size, loading concentration, and other experimental factors. Performance metrics shown here are for the NovaSeq 6000 System only; run times on the NovaSeq 5000 System may be slightly longer.

## Flexible Performance

The NovaSeq 5000 and NovaSeq 6000 Systems offer tremendous flexibility in sequencing options, supporting discovery across an extensive range of applications. Users can mix and match between the 4 flow cell types (S1, S2, S3, or S4*), run 1 or 2 flow cells at a time, and choose between multiple read lengths to easily adjust output for each sequencing run. The systems are compatible with Illumina library preparation kits, supporting a wide range of methods, from expression profiling to WGS and beyond (Table 3). Now users can run what they need, when they need it.

## Streamlined Operation

The NovaSeq 5000 and NovaSeq 6000 Systems increase lab efficiency through innovative user design features, workflow simplification, and reduced hands-on time. Advanced features include (Figure 3):

- Load-and-go reagent cartridges that represent an 80% reduction in consumables compared to the HiSeq Series of Systems
- RFID-encoded consumables enable automated reagent traceability and ensure compatibility of all sequencing reagents and flow cells
- Automated flow cell loading and onboard cluster generation minimize hands-on time and variability due to manual processing

### Load-and-Go Reagents

The NovaSeq Series replaces dozens of tubes and bottles with 1 preconfigured sequencing by synthesis (SBS) reagent cartridge and 1 cluster generation reagent cartridge for each flow cell run. The cartridges require no preparation other than thawing, eliminating the need for user intervention, minimizing opportunities for errors, and drastically reducing run setup time. In all, it takes only minutes to set up a run on the NovaSeq 5000 or NovaSeq 6000 System.

**Table 3: Examples of Compatible Library Prep Kits for Numerous Applications**

| |
|---|
| **Whole-Genome Sequencing** |
| TruSeq® Nano DNA Library Prep Kit[a] |
| TruSeq DNA PCR-Free DNA Library Prep Kit[b] |
| Nextera® Mate Pair Library Prep Kit |
| **Targeted Resequencing** |
| TruSeq Exome Library Prep Kit |
| TruSeq Rapid Exome Library Prep Kit[c] |
| **Transcriptome Sequencing** |
| TruSeq Stranded Total RNA Library Prep Kit[a] |
| TruSeq Stranded mRNA Library Prep Kit[a] |
| TruSeq RNA Access Library Prep Kit[a] |

The library prep kits listed are examples of what is available for use with the NovaSeq Series. For a complete list of Illumina library prep kits, visit www.illumina.com.

a. An Illumina Qualified Method supported by Beckman, Eppendorf, Hamilton, Perkin Elmer, and Tecan liquid-handling systems.
b. An Illumina Qualified Method supported by Beckman, Eppendorf, Perkin Elmer, and Tecan liquid-handling systems.
c. An Illumina Qualified Method supported by the Eppendorf liquid-handling system.

### Onboard Cluster Generation

The NovaSeq 5000 and NovaSeq 6000 Systems feature fully automated onboard cluster generation, significantly reducing hands-on time. Prepared libraries are loaded directly into a sample tube that sits in a preconfigured reagent cartridge, which is loaded directly onto the system for fully automated cluster generation.

### Seamless Integration

The NovaSeq Series is part of a comprehensive sequencing and informatics ecosystem that includes automated workflow management with BaseSpace® Clarity Laboratory Information Management System (LIMS). Data generated on a NovaSeq 5000 or NovaSeq 6000 System can be streamed directly into BaseSpace Sequence Hub for immediate analysis and interpretation.

## Cutting-Edge Technology

The NovaSeq Series represents the most powerful, simple, scalable, and reliable high-throughput Illumina sequencing platforms to date, producing impeccable data quality. At the core of these systems is the proven Illumina SBS chemistry. More than 90% of the world's sequencing data are generated by Illumina SBS chemistry.‡ This proprietary reversible terminator–based method enables the massively parallel sequencing of billions of DNA fragments, detecting single bases as they are incorporated into growing DNA strands. The method significantly reduces errors and missed calls associated with strings of repeated nucleotides (homopolymers).

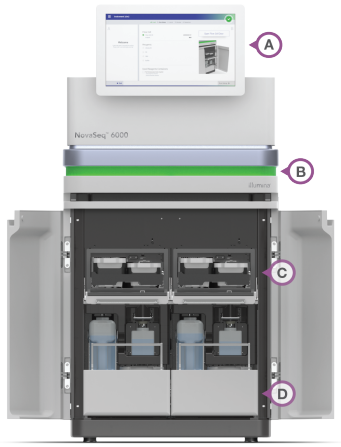


**Figure 3: The NovaSeq Series Provides Straightforward Operation—** Many features of the NovaSeq Series are designed to simplify genomic studies, including A. intuitive touch screen interface, B. lighted LED display indicates flow cell status. C. snap-in cartridges contain ready-to-use reagents, D. waste containers remove easily for disposal.

### Ingenious Design

The NovaSeq Series unites the latest high-performance imaging with the next generation of Illumina patterned flow cell technology to deliver massive increases in throughput.

The superior optics of the NovaSeq Series offer high-resolution, high-speed scanning that represents a significant increase relative to the HiSeq X® System. This quicker scan speed contributes to making the NovaSeq 5000 and NovaSeq 6000 Systems the highest throughput Illumina sequencing platforms yet.

The NovaSeq Series leverages patterned flow cells, first deployed on the HiSeq X System, with an improved, higher density design. Each flow cell contains billions of nanowells at fixed locations, a design that provides even cluster spacing and uniform feature size to deliver extremely high cluster density. The redesigned NovaSeq flow cell design further reduces the spacing between nanowells, significantly increasing cluster density and data output. Combining this increase in cluster density with the proprietary exclusion amplification clustering method maximizes the number of nanowells occupied by DNA clusters originating from a single DNA template. The result is an escalation in data output.

‡ Data calculations on file. Illumina, Inc., 2015

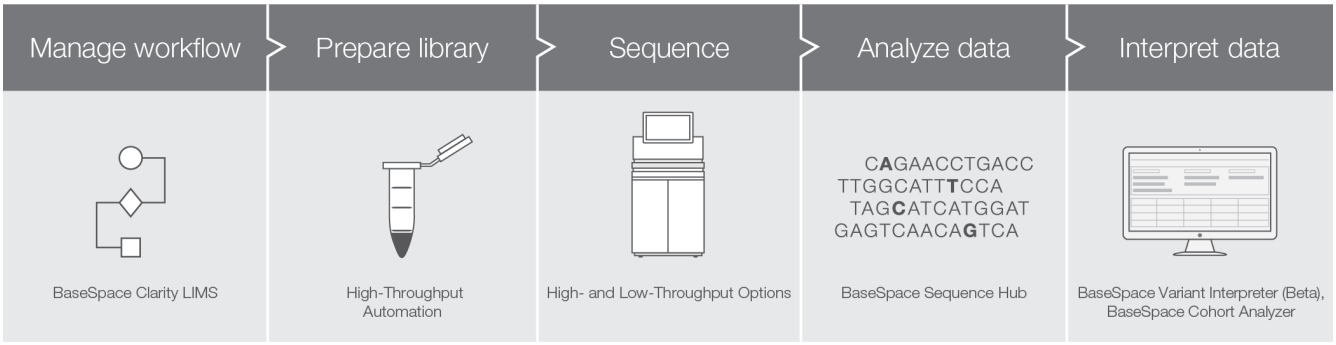


**Figure 4: The NovaSeq Series is Part of a Comprehensive Sequencing Ecosystem —** The NovaSeq Series is compatible with BaseSpace Clarity LIMS, the Illumina library preparation kit portfolio, Illumina Qualified Methods support, and the BaseSpace Sequence Hub, simplifying sequencing operations with a single, unified workflow.

## Comprehensive Sequencing Ecosystem

The NovaSeq Series forms the cornerstone of a comprehensive sequencing ecosystem that encompasses workflow management, manual or automated library preparation, sequencing, data analysis and interpretation, and service and support (Figure 4).

### LIMS: Prepared Sample to Analysis Information Management

The NovaSeq Series is fully compatible, out of the box, with BaseSpace Clarity LIMS. By using a LIMS, labs can improve operational efficiency with comprehensive sample and reagent tracking, automated workflows, and integrated instrument operation. BaseSpace Clarity LIMS provides labs with an intuitive user interface and preconfigured workflows that allows rapid adoption for immediate process tracking and scalability. With BaseSpace Clarity LIMS, sequencing data become easier to manage and assess across experiments. The NovaSeq Series can also be integrated with user-developed and other third-party LIMS.

### Automated Library Preparation

Illumina has partnered with leading automated liquid-handling vendors to develop several "Illumina Qualified" methods supporting the Illumina library portfolio (Table 3). The Illumina Qualified designation means that Illumina analysis has shown that libraries prepared with these methods perform comparably to those prepared manually. Illumina Qualified methods can be rapidly installed for immediate use in any lab, minimizing time-consuming, costly development efforts. Automating library preparation increases uniformity across experiments, minimizes errors, reduces hands-on time, and enables higher throughput, allowing users to harness the unmatched productivity of the NovaSeq Series.

### Data Analysis

Data from the NovaSeq Series can be seamlessly streamed into BaseSpace Sequence Hub, a user-friendly genomics cloud computing platform that offers simplified data management, analytical sequencing tools, and data storage.[§] BaseSpace Sequence Hub is optimized to automate processing of the large volume of data generated. Researchers will find a rich ecosystem of commercial and open-source tools, from Illumina and third-party developers, for data analysis, including alignment and variant detection, annotation, visualization, interpretation, and somatic variant calling.

For users interested in other analysis options, NovaSeq System Software generates base calls and quality scores in real time, saving these as per cycle base call (*.cbcl) files. Using the included, standalone bcl2fastq2 software, these *.cbcl files are translated into FASTQ files for use in downstream analysis.

Whichever approach to data analysis is used, in-house pipelines or BaseSpace Sequence Hub, the NovaSeq Series accommodates a seamless transition from sequencing to data analysis.

## Summary

The NovaSeq Series of Sequencing Systems expands NGS possibilities for all researchers. With unmatched scalable throughput, tremendous flexibility to support a range of applications, and streamlined operation, the NovaSeq 5000 and NovaSeq 6000 Systems are the most powerful high-throughput Illumina sequencing systems to date, perfectly positioned to help users uncover more about the genome than ever before.

## Learn More

To learn more about the NovaSeq Series of Systems, visit www.illumina.com/novaseq

[§]Learn more about the BaseSpace Sequence Hub at www.illumina.com/informatics.

## Ordering Information

| System | Catalog No. |
|---|---|
| NovaSeq 5000 System[a] | 20012849 |
| NovaSeq 6000 System | 20012850 |
| NovaSeq 5000 System Upgrade[a] | 20015868 |
| **Sequencing Reagent Kits** | **Catalog No.** |
| NovaSeq 5000/6000 S2 Reagent Kit (300 cycles) | 20012860 |
| NovaSeq 5000/6000 S2 Reagent Kit (200 cycles) | 20012861 |
| NovaSeq 5000/6000 S2 Reagent Kit (100 cycles) | 20012862 |

a. Currently not available for order.

## NovaSeq Series of Systems Specifications

| Specifications |
|---|
| **Instrument Configuration** |
| Computer and touch screen display |
| Installation setup and accessories |
| Data collection and analysis software |
| **Instrument Control Computer** |
| Base Unit: Portwell WADE-8022 with Intel i7 4700EQ CPU |
| Memory: 2 × 8 GB DDR3L SODIMM |
| Hard Drive: None |
| Solid-State Drive: 256 GB mSATA |
| Operating System: Windows |
| Note: Computer configurations will be upgraded regularly, Contact your local account manager for current configuration. |
| **Operating Environment** |
| Temperature: 19°C to 25°C (22°C ±3°C), < 2°C change per hour |
| Humidity: Noncondensing 20–80% relative humidity |
| Altitude: Below 2000 meters (6500 feet) |
| Ventilation: Maximum of 8530 BTU/h and average 6000 BTU/h |
| For Indoor Use Only. |
| **Laser** |
| 532 nm, 660 nm, 780 nm, 790 nm |
| **Dimensions** |
| W × D × H: 80.0 cm (31.5 in) × 86.4 cm (34 in) × 134.6 cm (53 in) |
| Weight: 450 kg (1000 lb) |
| Crated Weight: 638 kg (1408 lb) |
| **Power Requirements** |
| 208–240 VAC 50/60Hz, 16A, single phase, 2400 W |
| Illumina provides a region-specific uninterruptible power supply for the NovaSeq Series |

## Maximize Performance and Productivity with Illumina Services, Training, and Consulting

Whether immediate help is needed during an instrument run, or in-depth consultations are required for sophisticated workflows, Illumina can help. Illumina service and support teams provide a full suite of expedient, customized solutions from initial trainings to ongoing NGS consultation. Illumina service and support is available for researchers just getting started with NGS, looking to scale up sequencing production, or interested in maximizing performance:

### Getting Started

- Custom design and wet-lab optimization through Concierge Services
- Customer sample testing with Proof of Concept (POC) Services
- Hands-on Library Preparation Training
- Custom Bioinformatics Services

### Scaling Up

- Computation and storage solutions with Genomics IT Services
- High-throughput operations optimization using Illumina High-Throughput Sequencing Consulting

### Sustaining

- Uptime and productivity maximization with tiered Instrument Service Plans

### Compliance

- Quality guidelines compliance with Qualification Services (Installation Qualification (IQ)/Operational Qualification (OQ)/Performance Qualification(PQ))

For more on Illumina support offerings, visit:
www.illumina.com/product-services

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