NextSeq™ 1000 & NextSeq 2000 Sequencing Systems

Explore current and emerging applications with higher efficiency and fewer restraints

- Flexibility and scalability to expand the breadth of applications performed on a benchtop system
- Operational simplicity with load-and-go cartridges and integrated onboard informatics
- Improved run economics with high output to support larger studies and data-intensive methods

For Research Use Only. Not for use in diagnostic procedures.
Introduction

Innovations in next-generation sequencing (NGS) are helping the genomics community ask, and answer, increasingly complex scientific questions. Across the spectrum of oncology, microbiome research, single-cell studies, and other emerging applications, researchers need sequencing power to fuel larger studies of more samples at greater sequencing depth and at a lower cost.

To support this expansion in research, Illumina is committed to enabling exploration of the genome, transcriptome, and epigenome by providing users with innovative advances in technology and systems. Over the last 20 years, Illumina has led the charge to improve sequencing capabilities across the entire workflow, making it easier to use while driving down costs for both low-throughput and high-throughput users.

This tradition continues with the NextSeq 1000 and NextSeq 2000 Sequencing Systems (Figure 1, Table 1). These game-changing platforms offer breakthrough system design, chemistry innovations, compatibility with an expansive list of library preparation options, and onboard integrated informatics for rapid secondary analysis. A range of flow cell types and kits support a variety of customer batching and throughput needs, from bulk RNA sequencing (RNA-Seq) to 16S sequencing and shotgun metagenomics.

The result: Illumina NextSeq 1000 and NextSeq 2000 Sequencing Systems are flexible, scalable platforms with a breadth of flow cell options that will support the research of today and tomorrow.

Flexibility to do more, scalability to grow more

The NextSeq 1000 and NextSeq 2000 Sequencing Systems leverage the latest advances in optics, instrument design, and reagent chemistry to miniaturize the volume of the sequencing reaction while increasing output and reducing the cost per run. Now, users can obtain the throughput, data quality, and cost required to meet their needs, from smaller batch sizes and lower throughputs, to higher throughput, high-intensity applications, all on a benchtop sequencing system.

Technology innovations enabling enhanced performance

The NextSeq 1000 and NextSeq 2000 Sequencing Systems use patterned flow cells similar to those that power the NovaSeq™ 6000 System. The result is a highly flexible, robust, and scalable benchtop system that offers high-cluster-density flow cells to drive down the cost per gigabase (Gb) of the sequencing run.

To take full advantage of these higher density flow cells, the NextSeq 1000 and NextSeq 2000 Sequencing Systems feature a novel super-resolution optics system that yields highly accurate imaging data with greater resolution and higher sensitivity than traditional benchtop systems. This miniaturization provides scalability for a variety of output quantities while maintaining the same high standards of data quality enjoyed by NextSeq 550 and MiSeq™ System users.
Building on decades of expertise, the NextSeq 1000 and NextSeq 2000 Systems feature industry-leading sequencing by synthesis (SBS) chemistry that is optimized to increase cluster brightness, reduce channel cross talk, and improve signal-to-noise ratio. The combination of this advancement with formulation improvements that reduce the actual reaction size enables NextSeq 1000/2000 reagent users to realize high-quality data and an overall reduction in reagent volume and waste, minimizing physical storage requirements. Additionally, the improved robustness and stability enables ambient shipment of the flow cell.

Table 1: Performance parameters for the NextSeq 1000 and NextSeq 2000 Sequencing Systems

<table>
<thead>
<tr>
<th>Read length</th>
<th>NextSeq 1000/2000 P1 Reagents</th>
<th>NextSeq 1000/2000 P2 Reagents</th>
<th>NextSeq 2000 P3 Reagents</th>
</tr>
</thead>
<tbody>
<tr>
<td>Output per flow cell(^a)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reads CPF</td>
<td>100M</td>
<td>400M (300M for 2 × 300 bp)</td>
<td>1.2B</td>
</tr>
<tr>
<td>1 × 50 bp</td>
<td>–</td>
<td>–</td>
<td>60 Gb</td>
</tr>
<tr>
<td>2 × 50 bp</td>
<td>10 Gb</td>
<td>40 Gb</td>
<td>120 Gb</td>
</tr>
<tr>
<td>2 × 100 bp</td>
<td>–</td>
<td>80 Gb</td>
<td>240 Gb</td>
</tr>
<tr>
<td>2 × 150 bp</td>
<td>30 Gb</td>
<td>120 Gb</td>
<td>360 Gb</td>
</tr>
<tr>
<td>2 × 300 bp</td>
<td>60 Gb</td>
<td>180 Gb (300M reads CPF)</td>
<td>–</td>
</tr>
<tr>
<td>Quality scores(^b)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1 × 50 bp</td>
<td>≥ 90% of bases higher than Q30</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2 × 50 bp</td>
<td>≥ 90% of bases higher than Q30</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2 × 100 bp</td>
<td>≥ 85% of bases higher than Q30</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2 × 150 bp</td>
<td>≥ 85% of bases higher than Q30</td>
<td></td>
<td></td>
</tr>
<tr>
<td>2 × 300 bp</td>
<td>≥ 80% of bases higher than Q30</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Run time</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>1 × 50 bp</td>
<td>–</td>
<td>–</td>
<td>~11 hr</td>
</tr>
<tr>
<td>2 × 50 bp</td>
<td>~10 hr</td>
<td>~13 hr</td>
<td>~19 hr</td>
</tr>
<tr>
<td>2 × 100 bp</td>
<td>–</td>
<td>~21 hr</td>
<td>~33 hr</td>
</tr>
<tr>
<td>2 × 150 bp</td>
<td>~19 hr</td>
<td>~29 hr</td>
<td>~48 hr</td>
</tr>
<tr>
<td>2 × 300 bp</td>
<td>~34 hr</td>
<td>~44 hr</td>
<td>–</td>
</tr>
</tbody>
</table>

\(^a\) Output specifications based on a single flow cell using Illumina PhiX control library at supported cluster densities; CPF, clusters passing filter.

\(^b\) Quality scores are based on an Illumina PhiX control library; performance may vary based on library type and quality, insert size, loading concentration, and other experimental factors.
Pioneer breakthrough studies with access to broader capabilities

With fast, accurate results, extensibility from 10 Gb up to 360 Gb, and flexible informatics options, the NextSeq 1000 and NextSeq 2000 Sequencing Systems are ideal for a wide range of applications (Table 2) across metagenomics, spatial transcriptomics, oncology, genetic disease, and more. Add in unwavering expert support and labs are set for today's workload and future emerging applications.

A powerful, simplified workflow driven by an integrated system and advanced informatics

At Illumina, customer experience is at the center of every innovation, making it as easy as possible to prepare samples, sequence, and analyze data. The NextSeq 1000 and NextSeq 2000 Sequencing Systems offer a simplified workflow that combines load-and-go ease and included advanced informatics (Figure 2 and Figure 3) to benefit both new and advanced users.

Easy-to-use cartridge-based platform

The NextSeq 1000 and NextSeq 2000 Sequencing Systems take advantage of an integrated cartridge that includes reagents, fluidics, and the waste holder, simplifying library loading and instrument use. Simply thaw the reagent cartridge, insert the flow cell into the cartridge, load the library into the cartridge, and insert the assembled cartridge into the instrument. Denaturation and dilution steps occur onboard automatically.

In addition to ease of use, the fully integrated cartridge design improves efficiency throughout the sequencing run. By miniaturizing many of the sequencing reactions, the unique design:

- Lowers operating costs
- Improves recyclability
- Minimizes waste volume

Because the reagents never leave the cartridge, the dry instrument design does not require washing, enabling streamlined instrument maintenance and optimizing instrument efficiency.

<table>
<thead>
<tr>
<th>Manage</th>
<th>Prepare</th>
<th>Sequence</th>
<th>Analyze</th>
<th>Monitor</th>
</tr>
</thead>
<tbody>
<tr>
<td>Track samples and manage runs easily and efficiently with cloud-based and manual options</td>
<td>Prepare libraries with a wide ecosystem of compatible library preparation kits</td>
<td>Load prepared libraries into the sequencing cartridge and insert into the NextSeq 1000 or NextSeq 2000 System</td>
<td>Analyze using onboard DRAGEN secondary analysis or stream to the cloud for additional analysis flexibility</td>
<td>Increase lab efficiency with Illumina Proactive instrument performance service</td>
</tr>
</tbody>
</table>

Figure 2: Intuitive library-to-analysis workflow—The NextSeq 1000 and NextSeq 2000 Sequencing Systems provide a comprehensive workflow that includes user-friendly run setup, a wide ecosystem of compatible library prep kits, load-and-go operation, and integrated, onboard secondary analysis.
Table 2: Some of the broader applications available on the NextSeq 1000 and NextSeq 2000 Sequencing Systems

<table>
<thead>
<tr>
<th>Application&lt;sup&gt;a&lt;/sup&gt;</th>
<th>NextSeq 1000/2000 P1 Reagents</th>
<th>NextSeq 1000/2000 P2 Reagents</th>
<th>NextSeq 2000 P3 Reagents</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No. samples</td>
<td>Time</td>
<td>No. samples</td>
</tr>
<tr>
<td>Small whole-genome sequencing (300 cycles) 130 Mb genome; &gt; 30× coverage</td>
<td>7</td>
<td>~19 hr</td>
<td>30</td>
</tr>
<tr>
<td>Whole-exome sequencing (200 cycles) 50× mean targeted coverage; 90% targeted 20× coverage</td>
<td>4&lt;sup&gt;b&lt;/sup&gt;</td>
<td>~19 hr</td>
<td>16</td>
</tr>
<tr>
<td>Total RNA-Seq (200 cycles) 50M read pairs per sample</td>
<td>2&lt;sup&gt;bc&lt;/sup&gt;</td>
<td>~19 hr</td>
<td>16</td>
</tr>
<tr>
<td>mRNA-Seq (200 cycles) 25M read pairs per sample</td>
<td>4&lt;sup&gt;bc&lt;/sup&gt;</td>
<td>~19 hr</td>
<td>32</td>
</tr>
<tr>
<td>Single-cell RNA-Seq (100 cycles)&lt;sup&gt;a&lt;/sup&gt; 5K cells, 20K reads/cell</td>
<td>1&lt;sup&gt;a&lt;/sup&gt;</td>
<td>~10 hr</td>
<td>4</td>
</tr>
<tr>
<td>miRNA-Seq or small RNA analysis (50 cycles) 1M reads/sample</td>
<td>9&lt;sup&gt;e&lt;/sup&gt;</td>
<td>~10 hr</td>
<td>36&lt;sup&gt;f&lt;/sup&gt;</td>
</tr>
<tr>
<td>16S RNA sequencing (600 cycles)</td>
<td>384&lt;sup&gt;g&lt;/sup&gt;</td>
<td>~34 hr</td>
<td>384&lt;sup&gt;g&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

<sup>a</sup> Recommended sequencing depth will largely depend on sample type and experimental objective and will need to be optimized for each study.

<sup>b</sup> Use P1 300-cycle kit.

<sup>c</sup> Recommended read lengths are 2 × 75 bp for Illumina Stranded Total RNA Prep and Illumina Stranded mRNA Prep and 2 × 100 bp for Illumina RNA Prep with Enrichment.

<sup>d</sup> P1 reagents are a good option for single-cell quality control experiments.

<sup>e</sup> Use P1 100-cycle kit.

<sup>f</sup> Use P2 100-cycle kit.

<sup>g</sup> A maximum of 384 unique dual indexes is available.

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**Figure 3: Flexible informatics suite**—The NextSeq 1000 and NextSeq 2000 Systems feature local and cloud-based options for run setup, run management, and data analysis, enabling users to run their sequencing their way.
Simplified analysis, flexible options

The NextSeq 1000 and NextSeq 2000 Systems offer access to onboard, local, and cloud-based analysis software, giving users the flexibility to analyze data in a manner that meets their needs.

Runs can be set up locally or in the cloud. For local setup, users can create their own sample sheet or take advantage of a convenient, preset Illumina template. Cloud-based setup uses the Run Planner app in BaseSpace™ Sequence Hub. Once the run setup information is ready, it is imported into the NextSeq 1000 and NextSeq 2000 Systems. Users then select and start the run of interest. Optimized instrument software provides a cleaner interface with easy-to-read screens, easier-to-understand run metrics, and improved visualization of instrument and run status compared to earlier benchtop systems. The NextSeq 1000 and NextSeq 2000 Systems output industry-standard file formats used by various laboratory information management systems (LIMS) for secure, automated sample tracking and information management. Secondary analysis can be configured as part of run setup, reducing the number of user touchpoints required.

Accurate and efficient analysis with onboard DRAGEN secondary analysis

Onboard DRAGEN (Dynamic Read Analysis for GENomics) secondary analysis offers an accurate, efficient solution for variant calling. The DRAGEN platform uses optimized, hardware-accelerated algorithms for a wide variety of genomic analysis solutions, including base call (BCL) file conversion, compression, mapping, alignment, sorting, duplicate marking, and variant calling. New pipelines will be made available for new and emerging applications. The onboard solution provides access to select DRAGEN informatics pipelines (Table 3), enabling users to generate results in as little as two hours. DRAGEN informatics use best-in-class pipeline algorithms to help novice and expert users overcome bottlenecks in data analysis and reduce reliance on external informatics experts. Users spend less time and effort running production-level pipelines and can focus more on results. Onboard DRAGEN analysis is included in the instrument cost and does not require the purchase of an additional license.

### Table 3: Push-button DRAGEN informatics pipelines integrated into the NextSeq 1000 and NextSeq 2000 Systems

<table>
<thead>
<tr>
<th>Pipeline</th>
<th>Applications</th>
<th>Key functionality</th>
</tr>
</thead>
</table>
| DRAGEN Enrichment | • Whole-exome sequencing  
• Targeted resequencing | • Alignment  
• Small variant calling  
• Germline and somatic (tumor only) modes  
• Structural variant (SV) calling  
• Copy number variant (CNV) calling  
• Custom manifest files |
| DRAGEN RNA | • Whole-transcriptome gene expression  
• Gene fusion detection | • Alignment  
• Fusion detection  
• Gene expression  
• Differential expression |
| DRAGEN Single-Cell RNA | • Single-cell whole-transcriptome sequencing | • Cell barcodes and error correction  
• Alignment  
• Gene expression  
• Cell filtering  
• Basic reporting and visualization |
| DRAGEN ORA Compression | • Compression of FASTQ files | • Lossless compression  
• Up to 5x reduction in file size |
| DRAGEN Germine | • Whole-genome sequencing | • Alignment  
• Small variant calling  
• SV/CNV<sup>c</sup> calling  
• Repeat expansion<sup>c</sup>  
• Regions of homozygosity<sup>c</sup>  
• CYP2D6 genotyping<sup>c</sup> |
| DRAGEN Amplicon<sup>d</sup> | • DNA amplicon panels  
• Targeted resequencing | • Alignment  
• Small variant calling  
• Germline and somatic (tumor only) modes |

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<sup>a</sup> Additional DRAGEN informatics pipelines are available in the cloud; visit illumina.com/DRAGEN for a complete list.

<sup>b</sup> ORA, original read archive; DRAGEN ORA Compression can be enabled with any of the DRAGEN pipelines.

<sup>c</sup> Feature available for human genomes only.

<sup>d</sup> Supported for DNA samples only; available beginning with DRAGEN v3.8.
An ecosystem of apps in BaseSpace Sequence Hub

Users who prefer to use a cloud-based analysis solution can do so with BaseSpace Sequence Hub, a genomics cloud-computing platform that brings simplified data management and analytical sequencing tools directly to researchers in a user-friendly format. In the cloud, users can access a wide selection of bioinformatics tools, and share data globally. Data generated using the NextSeq 1000 and NextSeq 2000 Systems is compatible with industry-standard formats for convenient import into the solution of choice.

World-class service for more sample control and reduced downtime

To help you maximize investment, support peak performance, and minimize interruptions, Illumina provides a world-class support team comprised of experienced scientists who are experts in library prep, sequencing, and analysis. This dedicated team includes highly qualified field service engineers (FSE), technical applications scientists (TAS), field application scientists (FAS), system support engineers, bioinformaticians, and IT network experts, all deeply familiar with NGS and the applications that Illumina customers perform around the globe. Technical support is available via phone five days a week or access online support 24/7, worldwide and in multiple languages. Either way, support teams are available when you need them.

Modular construction for simpler, faster support

The NextSeq 1000 and NextSeq 2000 Systems are built in a modular fashion, simplifying service and support. Onboard sensors monitor system performance and alert users to possible issues. Troubleshooting and repairs are easier for the service engineer to perform, ultimately saving time and reducing frustration.

Each system purchase includes a one-year service warranty. Comprehensive maintenance, repair, and qualification solutions are also available. In addition, Illumina offers on-site training, ongoing support, phone consultations, webinars, and courses at various Illumina locations globally. We’re here with all the resources that you need to accelerate progress.

Illumina Proactive is a secure and remote instrument performance support service designed to detect risk failure preemptively, troubleshoot runs more efficiently, and prevent in-run failures. The service helps minimize unplanned downtime and avoid unnecessary sample loss by anticipating repairs and alerting Illumina field personnel to schedule maintenance visits.

Scale for the future while driving efficiencies today

With over 17,000 active systems, Illumina is setting the standard for NGS solutions. Joining this community provides access to a large ecosystem of applications, protocols, and informatics that have been built in collaboration with thousands of researchers and industry thought leaders across the globe.

Illumina has a proven track record of producing genomics solutions that empower researchers to perform studies at the throughput, scale, and price meeting their research objectives. The NextSeq 2000 System provides a wider range of throughput options to meet the needs of new and emerging applications while achieving better run economics for current applications. The NextSeq 1000 System has lower throughput relative to the NextSeq 2000 System and is available at a lower system price. To ensure flexible future scalability, customers who purchase a NextSeq 1000 System can easily upgrade to the NextSeq 2000 System.

Summary

The NextSeq 1000 and NextSeq 2000 Sequencing Systems revolutionize what can be accomplished with a benchtop sequencing system. With high flexibility and scalability to support a wide range of applications, small to large labs will have unprecedented capabilities to seek, and discover, more.
Learn more

NextSeq 1000 and NextSeq 2000 Sequencing Systems, illumina.com/NextSeq2000

Ordering information

<table>
<thead>
<tr>
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<th>Catalog no.</th>
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<tr>
<td>NextSeq 1000 Sequencing System</td>
<td>20038898</td>
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<td>NextSeq 1000 to NextSeq 2000 upgrade</td>
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<td>NextSeq 1000/2000 P1 Reagents (100 cycles)</td>
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<tr>
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</tr>
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NextSeq 1000 and NextSeq 2000 System specifications

Specifications

Instrument configuration
Self-contained, dry instrument with integrated DRAGEN field-programmable gate array (FPGA) secondary analysis

Instrument control computer
Base Unit: 2U Microserver located inside the instrument
Memory: 288 GB
Hard Drive: 3.8 TB SSD
Operating System: Linux CentOS 7.6

Operating environment
Temperature: 15°C–30°C
Humidity: 20%–80% relative humidity, non-condensing
Altitude: 0–2000 meters
For Indoor Use Only

Laser
Wavelengths: 449 nm, 523 nm, 820 nm
Safety: Class 1 Laser Product

Dimensions
W × D × H: 55 cm × 65 cm × 60 cm
Weight: 141 kg

Crated Dimensions
Crated W × D × H: 92 cm × 120 cm × 118 cm
Crated Weight: 232 kg

Power requirements
Instrument Input Voltage: 100 VAC to 240 VAC
Instrument Input Frequency: 50/60 Hz

Bandwidth for network connection
200 MB/s/instrument for internal network uploads
200 MB/s/instrument for BaseSpace Sequence Hub uploads
5 MB/s/instrument for Instrument Operational Data uploads

Product safety and compliance
NRTL certified IEC
61010-1 CE marked
FCC/IC approved