# NextSeq<sup>™</sup> 550 System

Tunable sequencing output and array scanning on a single instrument

- Support dynamic capacity needs and faster turnaround times with high- and mid-output options
- Experience easy operation with push-button control, load-and-go reagents, and streamlined informatics
- Harness NGS and array technologies to perform accurate genomics studies for multiple applications

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## Introduction

The NextSeq 550 System combines tried-and-true next-generation sequencing (NGS) and array capabilities with tunable outputs, enabling both small and large labs to scale to meet their needs. As a foundational instrument in the Illumina NGS system portfolio, the NextSeq 550 System is ideal for labs that want to expand beyond their current capacity and new labs interested in harnessing the complementary powers of sequencing and genotyping on a single instrument (Figure 1). Its fast DNA-to-results workflow enables rapid sequencing of exomes, targeted panels, and transcriptomes in a single run, with the flexibility to switch to low- or high-throughput sequencing as needed. Illumina scientists are available at every point along the way with support and guidance, enabling large clinical research labs to scale with confidence and smaller labs to employ both genotyping and sequencing technologies.

## Supporting dynamic capacities

The NextSeg 550 System enables researchers to keep pace with technology, switching quickly from one application to another and configuring output based on sample volume and coverage needs. This robust, scalable system turns a broad range of high-throughput applications into affordable everyday tools.

## Meeting the demands of active labs

The NextSeg 550 System offers various throughput levels, providing users with optimal operational efficiency. The NextSeq 550 System also delivers a one-day turnaround for numerous popular sequencing applications. With this instrument, users can sequence a broad range of samples per run:

- 1-12 exomes
- 1-16 transcriptomes
- 6-96 targeted panels
- 12-40 gene expression profiling samples



Figure 1: NextSeq 550 System—Proven platform combining the power of NGS and array technologies to support accurate genomic studies across a broad range of applications.

#### Enabling clinical research labs

For labs that already have a NextSeq 550 System, purchasing an additional instrument increases lab sequencing capacity, allowing for redundancy and modularity without requiring revalidation. Countless publications and adoption by leading genomics centers have demonstrated that the NextSeq 550 System is a reliable addition to any fleet.

#### Enabling smaller labs

The NextSeq 550 System is easily configured, providing users the scalability to meet demand and turnaround time requirements. Two flow cell configurations (highand mid-output) allow labs to shift easily from high- to low-throughput with each sequencing run (Table 1 and Table 2). The NextSeg 550 System provides integrated support for paired-end sequencing, offering user-defined read lengths up to  $2 \times 150$  bp. The system supports the full suite of Illumina library preparation and target enrichment solutions, offering compatibility across the Illumina sequencing portfolio. This allows labs to scale up studies to the higher throughput NextSeq 2000 and NovaSeq™ 6000 Systems or perform follow-up studies on another platform, such as the MiSeq™ System.

Table 1: NextSeq 550 System sequencing performance

Flow cell configuration	Read length	Output	Data quality
	2 × 150 bp	100-120 Gb	> 75% > Q30
High-output flow cell Up to 400M single reads Up to 800M paired-end reads	2 × 75 bp	50-60 Gb	> 80% > Q30
	1 × 75 bp	25-30 Gb	> 80% > Q30
Mid-output flow cell Up to 130M single reads Up to 260M paired-end reads	2 × 150 bp	32-39 Gb	> 75% > Q30
	2 × 75 bp	16-19 Gb	> 80% > Q30

Install specifications are based on Illumina PhiX control library at supported cluster densities (between 129 and 165 k/mm2 clusters passing filter). Actual performance parameters may  $vary\ based\ on\ sample\ type,\ sample\ quality,\ and\ clusters\ passing\ filter.\ The\ percentage\ of\ bases\ >\ Q30\ is\ averaged\ over\ the\ entire\ run.$ 

Table 2: Ultimate flexibility for multiple applications

Sequencing					
American	High-output flow cell		Mid-output	Mid-output flow cell	
Application	No. of samples	Time	No. of samples	Timeª	Required input <sup>b</sup>
Gene expression profiling > 10M reads 1 × 75 bp	40	11 hours	13	11 hours	25-1000 ng mRNA 10-20 ng total RNA
mRNA-Seq > 25M reads 2 × 75 bp	16	18 hours	5	15 hours	25-1000 ng mRNA
Enrichment panel 12 Mb region > 20× coverage at > 95% targets	36	29 hours	12	26 hours	10-100 ng DNA
Whole-exome sequencing 50× mean coverage	12	18 hours	3	15 hours	50 ng DNA
Small whole-genome sequencing 130 Mb genome > 30× coverage 2 × 150 bp	30	29 hours	10	26 hours	1-300 ng DNA
Array scanning performance					
BeadChip	Scan time per BeadChip		No. of samples		Scan time per sample
Infinium MethylationEPIC	40 minutes		8		5 minutes
Infinium CytoSNP-850K	40 minutes		8		5 minutes
Infinium HumanCytoSNP-12	40 minutes		12		3.3 minutes
Infinium HumanKaryomap-12	40 minutes		12		3.3 minutes

a. Total times include cluster generation, sequencing, and base calling on a NextSeq 550 System.

b. Input amount depends on quality of sample and library preparation solution used. For a complete list of Illumina library prep kits, visit illumina.com/products/by-type/sequencingkits/library-prep-kits.html

## Streamlined NGS workflow

The NextSeq 550 System is part of a fully integrated workflow that spans from library preparation to data analysis, report generation, and data sharing (Figure 2).



Figure 2: Example NextSeq 550 System sequencing workflow— The NextSeq 550 System offers a simple, integrated sequencing workflow from library preparation to data analysis. Workflow times will vary by experiment and assay type. Details shown are for Illumina Stranded mRNA Prep and a 2 × 75 bp read length. Analysis includes differential expression and identification of alternative transcripts.

## Library preparation kits for a range of applications

Illumina offers multiple library preparation kits that are compatible with the NextSeg 550 System. Solutions include targeted panels for investigating specific regions of interest and support for various methods. For a complete list, visit illumina.com.

## Easy system operation

The NextSeq 550 System features an intuitive user interface and load-and-go design that allow users to perform various sequencing applications with minimal training and instrument set-up time.

The NextSeq 550 System offers:

- Ready-to-use reagents
- Radio-frequency identification (RFID)-encoded consumables
- Integrated cluster generation and sequencing capabilities
- Easy-to-follow controls

Prepared libraries are loaded into the NextSeg 550 System where sequencing is automated and fast. Data are generated in as few as 6.5 hours for a 75-cycle sequencing run, and less than 30 hours for a paired-end 150-cycle run.

"The most valuable aspect of the NextSeq 550 System is ease of use. It really has become our workhorse over the last five years."

> —Dr. Angie Fawkes, Genetics Core, Edinburgh Clinical Research Facility

#### Streamlined bioinformatics

For data analysis, integrated system controls perform the initial base calling and quality scoring. There are several options available for secondary analysis, including the DRAGEN™ Bio-IT Platform, a suite of fast, accurate, and intuitive data analysis pipelines available through the cloud-based BaseSpace™ Sequence Hub or on-premise via a local server. A broad ecosystem of commercial and open-source data analysis software tools can also be used.

## Fast analysis with the DRAGEN Bio-IT Platform on BaseSpace Sequence Hub

The DRAGEN (Dynamic Read Analysis for GENomics) Bio-IT Platform offers ultrarapid secondary analysis. This PrecisionFDA award–winning informatics solution\*1 uses optimized, hardware-accelerated algorithms to provide industry-leading accuracy2 for calling multiple variant types, including small variants, copy number variations (CNVs),† and indels. Using DRAGEN best-in-class pipeline algorithms, novice and expert users can overcome bottlenecks in data analysis and reduce reliance on external informatics experts.

<sup>\*</sup> The DRAGEN Bio-IT Platform was awarded Best Performance for difficult-to-map regions and Best Performance for all benchmark regions in the 2020 PrecisionFDA Truth Challenge V2.1

<sup>†</sup> CNVs called in germline whole-exome sequencing studies.

The DRAGEN platform is available in the cloud through BaseSpace Sequence Hub, the easy-to-use Illumina cloud-computing data management platform. In the cloud, users can access a wide selection of bioinformatics tools in a user-friendly format, and share data globally.

BaseSpace Sequence Hub is a security-first platform that includes end-to-end encryption, auditing, and fine-grained access control. It is compliant with Health Insurance Portability and Accountability Act (HIPAA), ISO 27001 (Information Security Management System), and General Data Protection Regulation (GDPR) regulations.

#### Local Run Manager

Alternatively, customers can perform on- or off-instrument analysis using Local Run Manager. Local Run Manager provides an integrated solution for the creation and analysis of sequencing runs with the ability to link modules from run setup through analysis.

## Exceptional accuracy

At the core of the NextSeq 550 System is proven Illumina SBS chemistry, which is used to generate > 90% of the world's sequencing data.<sup>3</sup> This proprietary, reversible terminator-based method enables the parallel sequencing of millions of DNA fragments, detecting single bases as they are incorporated into growing DNA strands. The method virtually eliminates errors and missed calls associated with strings of repeated nucleotides (homopolymers).

Flow cells in the optimized NextSeq v2.5 Reagent Kits maintain starting fluorescent intensity, further improving performance throughout the run.4 In addition, the v2.5 flow cells are more resilient than their predecessors, able to withstand more humidity, heat, and prolonged storage than the v2 flow cells, and can be shipped at room temperature. With NextSeq v2.5 chemistry, the NextSeq 550 System provides exceptional accuracy for a broad range of methods, including small whole genome, targeted panel, exome, and transcriptome data at a wide range of coverage levels.

## One system, two technologies

The NextSeq 550 System integrates sequencing and microarray scanning capabilities into a single platform (Table 2). This reduces the need for multiple instruments, saving money and limited lab space.

## Experimental flexibility with array scanning

Microarray scanning on the NextSeg 550 System provides multifaceted labs with instant access to a powerful technology for further exploration. Array scanning is powered by the bead-based BeadArray<sup>™</sup> technology and proven Infinium<sup>™</sup> chemistry. Trusted Infinium chemistry produces exceptional data quality and call rates, as well as reproducible results.

The ability to scan microarrays provides a cost-effective, orthogonal method for identifying differentially methylated regions using the Infinium MethylationEPIC BeadChip and rapid confirmation of CNVs using the Infinium CytoSNP BeadChip. The NextSeq 550 System simultaneously maximizes the menu of cutting-edge research applications and minimizes instrument costs.

## Analyzing array data

Array data can be analyzed using various Illumina software solutions or third-party apps. For Infinium MethylationEPIC BeadChip data, Illumina offers the GenomeStudio™ Methylation Module, enabling researchers to perform differential methylation analysis for small-scale studies. For largescale studies, there are many freely available analysis packages that work in the software framework R for normalization and differential analysis of methylation data.<sup>5,6</sup>

"To toggle between sequencing and methylation is very straightforward."

> —Dr. Mike Friez, Director of the Diagnostic Laboratories at Greenwood Genetic Center

## World-class service and support

### Increase sample control and reduce downtime

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.

## 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 the applications that Illumina customers perform around the globe. Technical support is available worldwide and in multiple languages via phone 5 days a week or online 24/7.

Each system purchase includes a one-year service warranty. Comprehensive maintenance, repair, and gualification solutions are also available. In addition, Illumina offers onsite training, ongoing support, phone consults, webinars, and courses at various Illumina locations globally.

## Summary

Leveraging proven SBS chemistry, user-friendly operation, and a streamlined DNA-to-results workflow. the NextSeg 550 System is an ideal lab partner. Access industry-leading NGS and array technologies on a single system, achieve the needed throughput and timing with multiple flow cell options, and see what can be accomplished with a NextSeq 550 System today.

#### NextSeq 550 System specifications

Parameter	Specification	
Instrument configuration	RFID tracking for consumables	
Instrument control computer (Internal) <sup>a</sup>	Base Unit: Dual Intel Xeon E5-2618L 2.2 GHz CPU Memory: 96 GB RAM Hard Drive: 750 GB Operating System: Windows 10 enterprise	
Operating environment	Temperature: 19°C to 25°C (22°C ± 3°C) Humidity: Noncondensing 20%–80% relative humidity Altitude: Less than 2000 m (6500 ft) Air Quality: Pollution degree rating of II Ventilation: Up to 2048 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 × 25.0 in × 23.0 in) Weight: 83 kg (183 lb) Crated Weight: 151.5 kg (334 lb)	
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 200 mW	
Product safety and compliance	NRTL certified IEC 61010-1 CE marked FCC/IC approved	

### Learn more

To learn more about the NextSeg 550 System, visit illumina.com/systems/sequencing-platforms/nextseq.html.

To learn more about the DRAGEN Bio-IT Platform, visit illumina.com/DRAGEN.

#### Ordering information

Product	Catalog no.
NextSeq 550 System	SY-415-1002
NextSeq 500/550 Mid-Output v2.5 Kit (150 cycles)	20024904
NextSeq 500/550 Mid-Output v2.5 Kit (300 cycles)	20024905
NextSeq 500/550 High-Output v2.5 Kit (75 cycles)	20024906
NextSeq 500/550 High-Output v2.5 Kit (150 cycles)	20024907
NextSeq 500/550 High-Output v2.5 Kit (300 cycles)	20024908
TG NextSeq 500/550 Mid-Output Kit v2.5 (150 cycles)	20024909
TG NextSeq 500/550 Mid-Output Kit v2.5 (300 cycles)	20024910
TG NextSeq 500/550 High-Output Kit v2.5 (75 cycles)	20024911
TG NextSeq 500/550 High-Output Kit v2.5 (150 cycles)	20024912
TG NextSeq 500/550 High-Output Kit v2.5 (300 cycles)	20024913

TG-labeled consumables have features that help customers reduce the frequency of revalidation. These consumables are available only under supply agreement and  $% \left( 1\right) =\left( 1\right) \left( 1\right)$ require customers to provide a binding forecast. Contact your account manager for more.

## References

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- 2. Illumina. Accuracy Improvements in Germline Small Variant Calling with the DRAGEN Platform. Accessed January 8, 2021.
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- 5. Morris TJ, Butcher LM, Feber A, et al. ChAMP: 450K chip analysis methylation pipeline. Bioinformatics. 2014;30(3):428-430.
- 6. Assenov Y, Muller F, Lutsik P, et al. Comprehensive analysis of DNA methylation with RnBeads. Nat Methods. 2015;11(11):1138-1140.

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