# MiSeq<sup>™</sup>i100 and MiSeq i100 Plus Sequencing Systems

Simplest, fastest. For every lab.

- Simplified operations and intuitive, powerful onboard data analysis makes any NGS workflow easier
- Fast, flexible sequencing delivers same-day results and helps to scale your studies
- World-class technology and support from a proven genomics leader accelerates your next discovery

# illumina

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# The standard in sequencing simplicity and speed

Next-generation sequencing (NGS) has revolutionized the biological sciences, allowing labs to perform a wide variety of applications and study biological systems at a level never before possible. Compared to conventional technologies, NGS offers increased scale and sensitivity, providing more comprehensive results to help address many complex genomic questions. However, the need for skilled technicians to perform sequencing and data analysis has presented a challenge to labs wanting to transition to NGS.

Illumina is committed to unlocking the power of the genome by providing users with innovative advances in NGS technology and systems, and is leading the charge to improve sequencing capabilities. With the introduction of the benchtop MiSeq<sup>™</sup> System, we made NGS more accessible and easier to use, regardless of expertise level.

With the MiSeq i100 and MiSeq i100 Plus Sequencing Systems, Illumina continues to set the standard for the simplest, fastest benchtop sequencing (Figure 1). Breakthrough advancements in system design, XLEAP-SBS<sup>™</sup> chemistry, and integrated data analysis deliver enhanced usability, high data accuracy, and exceptional speed, generating results up to 4× faster than the MiSeq System. As part of an end-to-end NGS solution, the MiSeq i100 Series provides same-day results for various applications, including transcriptomics, microbial genomics, and targeted gene sequencing studies that impact microbiology, infectious disease, oncology, and more (Table 1). Combined with support from Illumina genomics experts, the MiSeq i100 Series makes NGS easier to adopt for virtually any lab.

# Impossibly simple from setup to analysis

At Illumina, customer experience is at the center of every innovation, making it as easy as possible to prepare libraries, sequence, and analyze data. Every aspect of the MiSeq i100 Series workflow is optimized to minimize the time and resources required to complete projects (Figure 2). The MiSeq i100 and MiSeq i100 Plus Systems offer a simplified workflow with run setup complete in only three steps and under 20 minutes. Load-and-go reagent cartridges and consumables are shipped and stored at



Figure 1: MiSeq i100 and MiSeq i100 Plus Sequencing Systems— Illumina innovation continues to broaden access to NGS with benchtop systems designed for simplicity and speed.

room temperature, so there's no need to wait for reagents to thaw before sequencing. Intuitive informatics minimize touchpoints and the need for specialized bioinformaticians for streamlined analysis, benefiting both new and advanced users.

#### Easy-to-use sequencing reagents

The MiSeq i100 and MiSeq i100 Plus Systems employ integrated cartridges that include sequencing reagents and the flow cell, simplifying library loading and instrument use and improving efficiency throughout the sequencing run. The cartridge design eliminates the need for maintenance washes on the instrument. Additional usability features include:

- Room temperature storage of consumables with no need to wait for reagents to thaw
- Lightweight reagents, buffer cartridges, and waste containers that are easy to handle
- Automated onboard flow cell denaturation, onboard cluster generation, and no post-run washing streamline the sequencing workflow
- Formamide-free reagents that simplify disposal
- Compatible library prep kits from Illumina and thirdparty vendors that require no additional conversion steps and streamline operations



Figure 2: The MiSeq i100 and MiSeq i100 Plus Sequencing Systems offer an intuitive and simplified workflow to ease the transition to NGS.

#### Sample-to-analysis NGS solutions

The MiSeq i100 and MiSeq i100 Plus Systems offer sample-to-analysis NGS workflows for varied methods, including small whole-genome sequencing (WGS) and metagenomics for microbial genomics and targeted gene sequencing for oncology and infectious disease. These workflows include library prep kits, panels, sequencing on the MiSeq i100 Series, and DRAGEN<sup>™</sup> secondary analysis (Table 1). Data analysis summaries are generated in two hours or less for most applications and simplify analysis by eliminating the need to upload data into bioinformatic pipelines. These workflows ease the transition to NGS or from the MiSeq System to the MiSeq i100 Series and provide several benefits for both new and current NGS users:

- Streamline experiment planning and setup with preselected library prep kits and probe panels
- Simplify data analysis with access to preconfigured DRAGEN pipelines either onboard or in the cloud, minimizing the need for bioinformatics expertise
- Increase confidence in your studies by comparing results to publicly available data sets in BaseSpace™ Sequence Hub

## Accurate, comprehensive and efficient analysis with DRAGEN software

Onboard DRAGEN secondary analysis features pipeline algorithms with award-winning accuracy<sup>\*</sup> to help users overcome bottlenecks in data analysis and reduce reliance on informatics experts. DRAGEN software performs a wide variety of genomic analysis solutions, including base call (BCL) file conversion, read alignment, and variant calling. It is included in the instrument cost and does not require the purchase of an additional license.

In addition to onboard pipelines, data from the MiSeq i100 Series can be streamed into BaseSpace Sequence Hub, a user-friendly genomics cloud-computing environment offering simplified run setup, monitoring, and analysis. There, users can access the full suite of DRAGEN pipelines for accurate secondary analysis and visualization of NGS data to generate meaningful biological results. Alternatively, labs interested in scalability and custom solutions can stream data from the MiSeq i100 Series to Illumina Connected Analytics, a flexible cloud bioinformatics platform that supports a broader range of pipelines and highly configurable, scalable analysis.

<sup>\*</sup> Most accurate secondary analysis in all-benchmark regions, as compared against all participating solutions F1 score using PrecisionFDA v2 Truth Challenge benchmark data; Illumina Internal Data on file for DRAGEN v4.2, Non-Ilumina data from Precision FDA v2 Truth Challenge 2020 (also applicable to DRAGEN v3.10, v4.0, and v4.2).<sup>1</sup>

Table 1: Example workflows for	various sequencing	applications or	n the MiSeq i100 Series
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Application	Library preparation	Reagent configuration	Data analysis	Access point
Small whole-genome sequencing (microbe, virus)	Illumina DNA Prep	MiSeq i100 5M, 25M, 50M, or 100M flow cell, 300-cycle or 600-cycle kit	DRAGEN sWGS	Onboard instrument, BaseSpace Sequence Hub, Connected Analytics
Targeted gene sequencing (amplicon- based, enrichment- based)	AmpliSeq for Illumina Custom DNA Panel, Trusight Hereditary Cancer Panel, oncoReveal NGS panel <sup>a</sup> , GenoScreen Deeplex Myc-TB Combo Kit <sup>b</sup>	MiSeq i100 5M, 25M, 50M, or 100M flow cell, 300-cycle kit	DRAGEN Amplicon, DRAGEN Enrichment,	BaseSpace Sequence Hub, Connected Analytics
16S amplicon sequencing	Illumina DNA Prep	MiSeq i100 5M, 25M, 50M, or 100M flow cell, 300-cycle or 600-cycle kit	16S Metagenomics	BaseSpace Sequence Hub
Shotgun metagenomics sequencing	Illumina DNA Prep, Illumina Stranded Total RNA Prep with Ribo-Zero Plus Microbiome	MiSeq i100 5M, 25M, 50M, or 100M flow cell, 300-cycle or 600-cycle kit	DRAGEN Metagenomics Pipeline, Microbiome Metatranscriptomics	BaseSpace Sequence Hub
Library QC	Illumina DNA PCR-Free Prep	MiSeq i100 5M flow cell, 300-cycle kit	Library QC	Onboard instrument, BaseSpace Sequence Hub, Connected Analytics
Transcriptome sequencing (mRNA- Seq, gene expression profiling)	Illumina Stranded mRNA Prep AmpliSeq for Illumina Custom RNA Panel	MiSeq i100 50M or 100M flow cell, 300-cycle kit	DRAGEN RNA	BaseSpace Sequence Hub, Connected Analytics
Pathogen detection and surveillance	Illumina Viral Surveillance Panel, Illumina Respiratory Pathogen ID/AMR Enrichment Panel Kit, Illumina Microbial Amplicon Prep, Illumina Microbial Amplicon Prep—Influenza A/B, Illumina COVIDSeq <sup>™</sup> Assay (96 samples)	MiSeq i100 5M, 25M, 50M, or 100M flow cell, 300-cycle kit	DRAGEN Microbial Enrichment Plus, DRAGEN Microbial Amplicon	Onboard instrument, BaseSpace Sequence Hub

a. oncoReveal NGS panel is a product of Pillar Biosciences Inc.

b. Not available in all countries.

### Faster, more flexible sequencing

The MiSeq i100 and MiSeq i100 Plus Systems are designed to decrease turnaround time four-fold, as compared to the MiSeq System, with sequencing run times as fast as four hours (Figure 3), same day (and same shift) results are possible. The MiSeq i100 Series features index-first sequencing, which enables early demultiplexing of run data, allowing for users to obtain a preview of sample representation prior to the completion of a run, enabling subsequent run planning as needed.



Figure 3: Decrease sequencing run times with the MiSeq i100 Series—Comparison of time to complete a sequencing run for the MiSeq i100 Series, the MiSeq System, the MiniSeq<sup>™</sup> System, and the iSeq<sup>™</sup> 100 System.

#### Wide output range for various study sizes

The MiSeq i100 and MiSeq i100 Plus Systems offer ten different reagent configurations with read lengths up to 2 × 300 bp that support an output range of 5M–100M reads and 1.5 Gb–30Gb (Table 2). The expanded output of the MiSeq i100 Series enables users to increase sample throughput readily and perform deeper sequencing for various applications. With 4× reads output compared to the MiSeq System, the MiSeq i100 Plus System can sequence 1–10 mRNA-Seq samples (based on 10M reads/ sample) for small, pilot studies or up to 100 small whole genome samples (based on 1M reads/sample) for larger studies in under eight hours (Table 1, Table 3).

#### Powered by XLEAP-SBS chemistry

The MiSeq i100 Series is powered by XLEAP-SBS chemistry, our fastest, most robust, and highest-quality sequencing by synthesis (SBS) chemistry to date. Built from the proven foundation of the most widely adopted and used SBS chemistry, XLEAP-SBS chemistry provides significant improvements in stability, speed, and performance across Illumina kits, delivering higher confidence in generated data and expediting project completion. The MiSeq i100 Series have a minimum specification of > 90% of bases above Q30 at 2 × 150 bp (Table 2), resulting in highly accurate (99.9%) data.

#### Groundbreaking sustainability innovations

The MiSeq i100 and MiSeq i100 Plus Systems were purposefully designed to reduce the environmental impact of sequencing. The improved robustness and stability of XLEAP-SBS reagents allows for shipping and storage at room temperature. This key innovation removes the requirement for cold chain logistics and freezer storage, delivering remarkable benefits in terms of sustainability and user experience:

- Reagent kits shipped at room temperature (with no dry ice and no ice packs) for less waste
- Consumables stored at room temperature don't require thawing, expediting sequencing setup time and saving freezer space

- Consumables have recyclable components that disassemble without special tools for simple disposal and a ~85% reduction in packaging waste compared to the MiSeq System
- Recyclable plastics and buffer cartridges reduce plastic
   waste to landfills

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Table 2: MiSeo	1100 Series	performance	parameters

Flow cell type <sup>▷</sup>	5M	25M	50M	100M
Outputª				
1 × 100 bp	—	2.5 Gb	5 Gb	10 Gb
2 × 150 bp	1.5 Gb	7.5 Gb	15 Gb	30 Gb
2 × 300 bp	3 Gb	15 Gb	30 Gb	_
Reads passing filter	per flow ce	a		
Single reads	5M	25M	50M	100M
Paired-end reads	10M	50M	100M	200M
Instrument run time				
1 × 100 bp	—	~ 4 hr	~ 4.5 hr	~ 5 hr
2 × 150 bp	~ 7 hr	~ 7 hr	~ 7.5 hr	~ 8 hr
2 × 300 bp	~ 15 hr	~ 15 hr	~ 15.5 hr	—
Quality scores <sup>d</sup>				
1 × 100 bp	≥ 90% of bases higher than Q30			
2 × 150 bp	≥ 90% of bases higher than Q30			
2 × 300 bp	≥ 85% of bases higher than Q30			

a. Specifications based on Illumina PhiX control library or a TruSeq<sup>™</sup> DNA Library created with Coriell sample NA12878 at supported cluster densities. Performance may vary based on library type and quality, insert size, loading concentration, and other experimental factors. MiSeq i100 Plus System available now. MiSeq i100 System available later in 2025. Performance metrics subject to change.

- b. 5M and 25M flow cells available on the MiSeq i100 System; 5M, 25M, 50M, and 100M flow cells available on the MiSeq i100 Plus System. 5M and 25M flow cells available now; 50M and 100M flow cells available in 2025.
- c. Run times include automated onboard cluster generation, sequencing, and base calling.
- d. A quality score (Q-score) is a prediction of the probability of an error in base calling. The percentage of bases ≥ Q30 is averaged across the entire run.

A un un line a trian un			No. of samples			
Application		Reads per sample	5M	25M	50M	100M
Transcriptomics	3' gene expression	1–5M	1–5	5–25	10-50	25–100
	Targeted RNA panel	1–5M	1–5	5–25	10-50	25–100
	mRNA-Seq	10-25M		1–2	1–5	1–10
	Total RNA-Seq	50M	_	—	1	1–2
Microbial genomics Shotgur	Pathogen detection	1M	1–5	1–25	1–50	1–100
	16S amplicon sequencing	0.1-0.2M	1–50	1–250	1–384	1–384
	Shallow shotgun metagenomics	0.5–10M	1–10	1–12	1–25	1–50
	Shotgun metagenomics	10-25M	—	1–2	1-5	1–10
	Small WGS	1M	1–5	1–25	1–50	1–100
Targeted gene sequencing <sup>a</sup>	Amplicon-based	0.1-50M	1–50	1–250	1–384	1–384
	Enrichment-based	0.1-50M	1–50	1–250	1–384	1–384
	Genome editing	0.1–50M	1–50	1–250	1–384	1–384
	Immune repertoire	2–25M		1–12	1–25	1–50
Quality control	Library QC	> 0.02M <sup>b</sup>		up to 3	84-plex <sup>c</sup>	

Table 3: Estimated sample throughput for key applications on the MiSeq i100 Series<sup>a</sup>

a. Reads per sample and sample throughputs are estimates and highly variable, depending on the panel and desired coverage.

b. Reads per sample is variable, depending on the plexity of the library.

c. Based on available Illumina indexes; additional indexes can be added.

## Trusted technology, trusted partner

Trusted for over a decade, Illumina has shipped over 10,000 MiSeq Systems globally. Cited in over 160,000 peer-reviewed publications, the MiSeq System is the most widely used NGS instrument on the market.<sup>3</sup> Building on our extensive expertise, Illumina has a relentless commitment to innovation and building future NGS capabilities and applications. The MiSeq i100 Series demonstrates our commitment to increasing access to genomics technology by continuing to deliver faster, simpler sequencing.

#### Committed to customer success

Illumina provides a world-class support team composed of experienced scientists who are experts in library prep, sequencing, and analysis. Technical support is available via phone five days a week or access online support 24/7, worldwide and in multiple languages, with rapid response time near most major metropolitan areas. Illumina provides excellent product consistency, supply, and quality enabled by a mature global manufacturing infrastructure. The MiSeq i100 and MiSeq i100 Plus Sequencing Systems can be connected to Illumina Proactive, a secure and remote instrument performance and proactive support service for enhanced and reliable instrument operation. Customers receive access to performance data, real-time updates on run progress, and assisted troubleshooting. Proactive risk detection by the Illumina support team minimizes unplanned downtime and increases sample success.

## Summary

The MiSeq i100 and MiSeq i100 Plus Sequencing Systems provide advancements in system design, sequencing chemistry, and integrated data analysis to deliver operational simplicity, exceptional speed, and proven accuracy for a wide variety of applications, including transcriptomics, microbial genomics, and targeted gene sequencing applications. Combined with the trusted Illumina support team, the transition to NGS is easier than ever before. The MiSeq i100 Series sets the highest standard and delivers the fastest, simplest sequencing for benchtop instruments.

#### Learn more

MiSeq i100 and MiSeq i100 Plus Sequencing Systems

DRAGEN secondary analysis

### References

- Mehio R, Ruehle M, Catreux S, et al. DRAGEN Wins at PrecisionFDA Truth Challenge V2 Showcase Accuracy Gains from Alt-aware Mapping and Graph Reference Genomes. illumina.com/science/genomics-research/articles/dragenwins-precisionfda-challenge-accuracy-gains.html. Accessed January 1, 2024.
- Illumina. DRAGEN secondary analysis data sheet. illumina. com/content/dam/illumina/gcs/assembled-assets/marketingliterature/dragen-bio-it-data-sheet-m-gl-00680/dragen-bioit-data-sheet-m-gl-00680.pdf. Published 2018. Updated 2022. Accessed January 1, 2024.
- 3. Data calculations on file, Illumina, Inc. 2022.

### Ordering information

System	Catalog no.	
MiSeq i100 Plus Sequencing System	20115695	
Sequencing reagent kits <sup>a</sup>	Catalog no.	
MiSeq i100 Series 5M Reagent Kit (300 cycles)	20126565	
MiSeq i100 Series 5M Reagent Kit (600 cycles)	20126566	
MiSeq i100 Series 25M Reagent Kit (100 cycles)	20126567	
MiSeq i100 Series 25M Reagent Kit (300 cycles)	20126568	
MiSeq i100 Series 25M Reagent Kit (600 cycles)	20115696	
<ul> <li>a. 5M and 25M flow cells available now. 50M and 100M flow cells available in 2025.</li> </ul>		

#### MiSeq i100 Series instrument specifications

Parameter	Specification
Instrument configuration	Logic for system control and analytics and full-HD touch screen monitor Installation setup and accessories Data collection and analysis software
Operating environment	Temperature: 15°C–30°C, <2°C change per hour Humidity: 20%–80% relative humidity, non-condensing Altitude: Below 2000 meters (6500 feet) Ventilation: Not applicable For Indoor Use Only
RFID	Operating frequency 13.56 MHz, 200 mW output power
Laser	Light emitting diode (LED) 450 nm, 511 nm
Dimensions	W × D × H: 40.2 cm × 44.8 cm × 47.3 cm Dry weight: 79.4 lb (36.0 kg) Crated weight: 108.1 lb (49 kg)
Power requirements	100–240 VAC 50/60 Hz, 300 W, single phase
Network connection	Up to 2 × 2.5 GBE connections using RJ-45 between the instrument and data management system; connect directly or through network
Bandwidth for network connection	50 Mb/s/instrument for internal network uploads 50 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

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