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iSeq 100™ Sequencing System

Executive Summary

Illumina sequencing by synthesis (SBS) chemistry has revolutionized biological and medical research. In the twelve years since its commercial launch, it has enabled researchers to make breakthrough discoveries and produce over 5000 original research publications covering whole-genome sequencing, targeted resequencing, *de novo* sequencing, epigenomics, transcriptional analysis, and DNA-protein interactions.[1] These studies were performed at a cost that was orders of magnitude lower than the cost had they been performed using other available technologies.[2]

Illumina SBS chemistry is greatly accelerating medical research and significantly enhancing our knowledge of important disease areas like cancer. Illumina SBS chemistry is used to generate more than 90% of the world's sequencing data.[3] Furthermore, Illumina sequencing systems have been the core next-generation sequencing (NGS) platforms used in the Encyclopedia of DNA Elements (ENCODE),[4] The Cancer Genome Atlas (TCGA),[5] and the 1000 Genomes projects.[6] The first cancer genome was sequenced using Illumina technology.[7] Since this initial cancer genome publication, multiple cancer genomes have been sequenced using Illumina technology, generating additional publications.[8–10] Also, a number of important epigenomic and transcriptomic discoveries have been made and published with this technology.[11,12] Illumina sequencing technology may play a key role in the future of diagnosis and treatment of disease. In addition, Illumina sequencing technology is being used by a new generation of researchers to establish their scientific careers, allowing them to pursue breakthrough science with reasonable budgets and minimal infrastructure.

iSeq 100 System Advantages

The iSeq 100 System delivers speed and simplicity in a standalone benchtop sequencer. With the smallest footprint in the Illumina portfolio, approximately one square foot, the iSeq 100 System fits easily into virtually any laboratory environment. The iSeq 100 System employs Illumina SBS chemistry, the most widely adopted NGS chemistry in the world.[3] This makes the iSeq 100 System an ideal platform for any lab seeking to perform rapid and cost-effective genetic analysis.

- Accessible Illumina Sequencing—With the lowest price of acquisition of any benchtop sequencing system, the iSeq 100 System enables virtually any lab to access the proven power of Illumina NGS technology.
- Broad application flexibility—Demonstrated by customer publications, which use Illumina sequencing systems, including:
  - Targeted resequencing—Amplicon sequencing, hybrid capture, 16S metagenomics, clone checking
  - AmpliSeq for Illumina targeted resequencing—A supported solution that features a growing menu of curated content design, including ready-to-use, community, and custom panels
  - Small whole-genome sequencing—*De novo* sequencing, resequencing, plasmid sequencing
  - Transcriptional analysis—Targeted RNA sequencing, digital gene expression, microRNA profiling

- Broad range of applications at low cost—Results from the ability of the iSeq 100 System to perform 2 × 150 bp cycles combined with up to 4M paired-end reads per flow cell. .
- Fast publication rate—More than 5000 original, peer-reviewed papers published in the last twelve years cite Illumina NGS technology.[1]
- Highly scalable technology and high rate of innovation—Over the course of eight years, Illumina has scaled its sequencing output from approximately 1 Gb on the Genome Analyzer System (introduced in 2005), to up to 15 Gb on the MiSeq™ System, 120 Gb on the NextSeq™ 500 System, 1 Tb on the HiSeq™ 2500 System, and 6 Tb on the NovaSeq™ System.
- High-quality reads—The iSeq 100 System delivers high-quality reads that do not require error correction by comparison to a reference sequence. The iSeq 100 System leverages SBS chemistry, a proprietary reversible terminator–based method that enables the massively parallel sequencing of millions of DNA fragments, to detect single bases as they are incorporated into growing DNA strands. The method virtually eliminates errors and missed calls associated with strings of repeated nucleotides (GC-rich, homopolymers).[13] Greater than 75% of bases are above Q30 for 2 × 150 bp iSeq run (Table 1).
- Increased productivity—Illumina offers scalable sequencing reagents for the iSeq 100 System, enabling users to employ a wide range of read lengths and data outputs depending on the application and sample numbers. Library denaturation and cluster amplification is fully automated on the iSeq 100 System, which reduces hands-on time and provides a complete walk-away solution.
- Ready access to a large user community—The iSeq 100 System harnesses proprietary Illumina SBS chemistry, a well-tested, proven technology that has been widely adopted across genome centers, as well as by individual investigator labs and core labs.

Instrument Detail

The iSeq 100 System is a massively parallel NGS system that sequences up to four million templates simultaneously, producing read lengths of up to 2 × 150 bp from each template, generating up to 1.2 Gb of high-quality data in a single run (Table 1).

Table 1: iSeq 100 System Performance Parameters[a]

Run Configuration	Reads (Passing Filter)/Run	Output	Quality Scores[b]	Run Time[c]
1 × 36 bp	4 M	144 Mb	> 85%	9 hours
1 × 50 bp	4 M	200 Mb	> 85%	9 hours
1 × 75 bp	4 M	300 Mb	> 85%	10 hours
2 × 150 bp	4 M	1.2 Gb	> 75%	17.5 hours

a. Performance parameters may vary based on sample type, sample quality, and clusters passing filter.
b. The percentage of bases > Q30 is averaged over the entire run.
c. Times include cluster generation, sequencing, base calling, and quality scoring.

Justification of Need

Direct access to an NGS system, such as the iSeq 100 System, has led to groundbreaking publications that have already transformed our understanding of genomes, epigenomes, and transcriptomes in unexpected ways.[4–12] The Illumina iSeq 100 System offers many unique attributes that can facilitate these studies in institutions ranging from very small labs to large genome centers. The publication rate for Illumina sequencing technologies has been greater than 600 publications per year for each of the last five years, more than the total number of publications currently available for the closest equivalent sequencing platform, the Ion Torrent system from Thermo Fisher Scientific.[1]

Applications Flexibility of the iSeq 100 System

The iSeq 100 System generates 1.2 Gb of data per run in about 17 hours, offering rapid, multiplexed sequencing for a wide range of applications:

- Resequencing
- AmpliSeq for Illumina Targeted Resequencing
- TruSeq™ Custom Amplicon Assay
- TruSeq Amplicon Cancer Panel
- TruSeq Custom Enrichment
- TruSeq Targeted RNA Expression
- Nextera™ PCR Amplicon
- Nextera Mate-Pair
- Nextera Rapid Capture Exome
- Nextera DNA Flex
- Small Genome *de novo* Sequencing
- Small RNA Sequencing
- 16S Metagenomics
- Transcriptome Sequencing of Small Genomes (< 5 Mb)

With the iSeq 100 System, even the smallest laboratory can perform a wide range of sequencing applications to advance their studies. Due to it applications flexibility, the critical needs of shared research facilities are also effectively addressed by the iSeq 100 System.

Global Support

Illumina offers a large, well-trained support team. The Illumina global support team consists of more than 300 people worldwide, 75% of whom have advanced degrees. It includes engineers, scientists, informatics experts, computer networking specialists, and professional trainers. The Illumina global support team qualifies, maintains, and repairs Illumina systems and will provide training to ensure iSeq 100 System customers are proficient and able to use their systems to perform all of the many supported applications. Assistance with data analysis may also be available.

Criterion	Justification	Illumina iSeq 100 System
Simple operation and high ease of use	Addresses the needs of a broad user base, enabling rapid generation of data without significant learning effort.	The iSeq 100 System has the simplest workflow of any benchtop NGS system. Sequencing libraries can be generated in as little as three hours with the Nextera DNA Flex Library Prep Kit. Denaturation and amplification of libraries is performed in a closed flow cell on board the instrument, obviating the need for additional robotics, clean rooms, or separated facilities. The system does not use emulsion PCR with its inherent protocol complexities, therefore, library preparation and clonal amplification can be performed with relative ease. No wash protocols are required. The simple workflow enables multiple samples from different labs to be run together, further increasing efficiency.
Demonstrated / published applications	The SBS-based iSeq 100 System simplifies the integration of a broad base of applications by the user, leading to more efficient research paths, and rapid publication. Over 5000 original papers have been published using Illumina SBS-based systems covering a diverse range of applications.[1]	Illumina sequencing technology has been validated by abstracts and publications for an extensive range of applications: • Targeted resequencing—Amplicon sequencing, hybrid capture, 16S metagenomics, clone checking • AmpliSeq for Illumina targeted resequencing—Ready-to-use panels offer predesigned content targeting variants most commonly seen in an area of interest, community panels contain content selected and designed with input from leading researchers, and custom content can be easily designed and ordered online through DesignStudio™ software • Small genome sequencing—*De novo* sequencing, resequencing, plasmid sequencing • Transcriptional analysis—Targeted RNA sequencing, digital gene expression, microRNA profiling
Short workflows	Maximize instrument use by allowing multiple users to use the system without constraints due to long instrument run times or long sample prep protocols.	The iSeq 100 System delivers high daily throughput, allowing users to complete projects significantly faster than the closest competitive platform. Runs can be completed in 7–17.5 hours, depending on read length and output. The iSeq 100 System also offers the shortest library prep protocols.
Intraplatform compatibility	System compatibility to support study scale up with larger sample numbers at higher throughput.	The iSeq 100 System is supported by the full suite of Illumina sample prep and target enrichment solutions, offering library compatibility across the Illumina sequencing system portfolio. This allows researchers to scale studies on the higher throughput MiniSeq, MiSeq, or NextSeq Systems.
Low input sample amount	Efficient library construction and amplification approaches.	The iSeq 100 System is supported by efficient, simple-to-use library preparation kits that require as little as 1 ng–1 µg of sample. Library construction methodology of competing platforms and emulsion PCR-based workflows are inefficient and require more input sample material.
Flexible chemistry	Users with diverse needs can optimize attributes of the technology to best fit their	The iSeq 100 System allows for reads lengths ranging from 1 × 36 bp to 2 × 150 bp. Read lengths can be tailored to suit the specific application. The iSeq 100 System also provides a paired-end sequencing capability that can effectively

Criterion	Justification	Illumina iSeq 100 System
	research needs. For example, long sequence read lengths may be more valuable for some applications, while sequencing both ends of short fragments may be critical for others. Flexible read length capabilities increase overall system productivity and address the needs of an experiment.	sequence both ends of short fragments (< 600 bp) as well as long DNA fragments (> 1 kb).
Wide application flexibility	Tailor output to user needs, allowing shared facilities to offer the broadest range of applications, and to optimize their sequencing queue to reduce time from DNA/RNA to answer.	The iSeq 100 System offers a wide range of sequencing outputs and read lengths, allowing users to scale their sequencing for maximum efficiency. **iSeq Reagents** **Read Length** / **Data Output** 1 × 36 bp — 144 Mb 1 × 75 bp — 300 Mb 2 × 150 bp — 1.2 Gb
High-quality results	High sequencing accuracy and the capability to detect genome variations with high sensitivity is critical for successful research.	The combination of short- and long-insert paired-end reads is critical for the detection of all genome variations. The iSeq 100 System generates more than 75% of bases greater than Q30 at 2 × 150 bp read lengths.
Availability of prepackaged kits	Ready-to-use, optimized kits increase productivity by reducing preparation and experimental design time.	The iSeq 100 System is supported by many ready-to-use kits for small-genome sequencing, targeted pulldown, RNA sequencing (standard and directional), and digital gene expression.
Availability of analytical tools	Wide range of analysis capabilities for a diverse application base.	Sequencing data can be run through a wide range of open-source or commercial pipelines developed for Illumina data, or transferred, analyzed, and stored securely in BaseSpace™ Sequence Hub, the Illumina genomics computing environment. BaseSpace Sequence Hub includes data analysis apps developed by Illumina. Thanks to Illumina industry-standard data formats, third-party developers have created a rich ecosystem of commercial and open-source tools for more extensive downstream analysis. The iSeq 100 System also comes equipped with Local Run Manager, an on-instrument analysis tool supporting a wide range of applications.
Low price of acquisition	Enable a wide range of project sizes and sample batches.	Lowest price of acquisition of any benchtop sequencing system on the market.

Learn More

To learn more about the iSeq 100 System, visit www.illumina.com/systems/sequencing-platforms/iseq.html or access the following resources:
Sole Source Document

Contact your local Account Manager

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