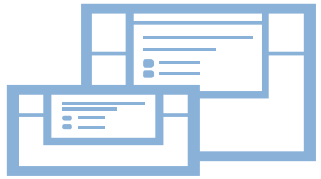
Expanding Sequencing Capabilities with the Illumina NextSeq2000

Ryan Hegarty | Sales Specialist, Sequencing

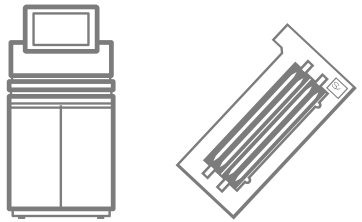
November 12, 2020

Improvements and New Releases Across the Illumina Portfolio

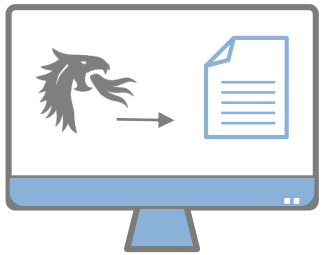
Illumina DNA PCR-Free and RNA Preparation



NextSeq™ 2000



DRAGEN and Illumina Analytics Platform



PREP
from sample to sequencer

SEQUENCE
from library to raw data (FASTQ)

ANALYZE & REPORT
from FASTQ to report

illumina®

Introducing the new Illumina RNA Library Prep Suite

Built for quality, scalability and flexibility

Preliminary Specifications

16 & 96
Sample kit sizes

384
Unique Dual Indexes[3]

	1 **Illumina Stranded Total RNA** Ligation with Ribo-Zero Plus	2 **Illumina Stranded mRNA** Ligation	3 **Illumina RNA Prep with Enrichment** (L) Tagmentation
Detection	Coding & Non-coding regions	Coding transcriptome w/ Poly A tail	Targeted coding region[4]
FFPE Compatibility	✓	-	✓
Input	1-1,000 ng[1] 10ng for optimal quality & FFPE	25-1,000 ng	10ng non-FFPE 20ng FFPE
Total Time (hours)[2]	7	6.5	< 9
Hands-on time (hours)[2]	< 3	< 3	< 2
Automation Friendly	✓	✓	✓
	• Includes Ribo-Zero Plus for multi-species rRNA depletion • Includes cDNA synthesis reagents	• Includes Illumina Poly A capture kit • Includes cDNA synthesis reagents	• Illumina Tested with Illumina Exome & Illumina Respiratory Viral Panel

1. Minimum input for high-quality RNA shown, 10ng minimum recommended for optimal quality and FFPE for Total RNA workflow
2. Hands-on and total time based on manual processing of up to 24 samples for Total & mRNA workflows and 1 sample on Enrichment workflow
3. Up to 192 UDIs available at launch, up to 384 available later in 2020
4. Note new Illumina RNA Prep with Enrichment does not provide strand information (is non-stranded)

illumina®

Ribo-Zero Plus Enzymatic Depletion Methodology

Robust method allows for increased flexibility for mix sample labs

Total RNA

1 Hybridize probes

2 Deplete rRNA

3 Remove probes

RNA of interest Ribo-Zero Probes Abundant rRNA

One Tube to deplete multiple species with Ribo-Zero Plus

Provides seamless study flexibility for mixed sample labs

- Cytoplasmic and mitochondrial rRNA – human/mouse/rat
- Globin transcripts
- Bacterial rRNA (Gram+, Gram-)

Depletion target	rRNAs targeted
Human Cytoplasmic rRNAs	28S, 18S, 5.8S, 5S
Human Mitochondrial rRNAs	12S, 16S
Human Beta Globin transcripts	HBA1, HBA2, HBB, HBG1, HBG2
Mouse and Rat rRNA	16S, 28S
Gram(-) Bacterial rRNAs	*E.coli*: 5S, 16S, 23S
Gram(+) Bacterial rRNAs	*B. subtilis*: 5S, 16S, 23S

illumina®

NEW Illumina RNA Prep with Enrichment workflow

Legacy TruSeq RNA Exome

Start with Total RNA[1]

Quantitation and Qualification
Fragment RNA
cDNA Synthesis
A-Tailing & Ligation
PCR
Quant & Norm
Enrichment 1
Capture 1
Washes & Elute
Enrichment 2
Capture 2
Washes & Elute
PCR

Quant & Norm
Sequence

NEW Illumina RNA Prep with Enrichment

Start with Total RNA[1]

Quantitation and Qualification
cDNA Synthesis

Tagmentation
PCR
Quant & Norm
Enrichment 1
Capture 1
Washes & Elute
PCR

Quant & Norm
Sequence

- No Fragmentation
- Tagmentation replaces A-Tailing & Ligation
- Single Hybridization

>50% Faster[2]
>80% Less Hands-on[2]

1 Before starting the workflow, quantify the total RNA using standard methods and assess quality using a fragment analysis method
2 Compared to TruSeq RNA Exome based on 2+ days total time and 10+ hours hands-on time vs. <9 hours total time and <2 hours hands-on time for New Illumina RNA prep with Enrichment

illumina®

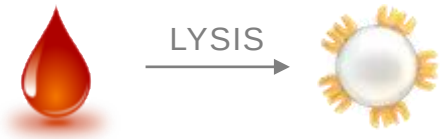
Illumina DNA PCR-Free Library Prep

Optional front-end extraction to direct input into PCR-free workflow

Direct sample input

30ul saliva

10ul blood

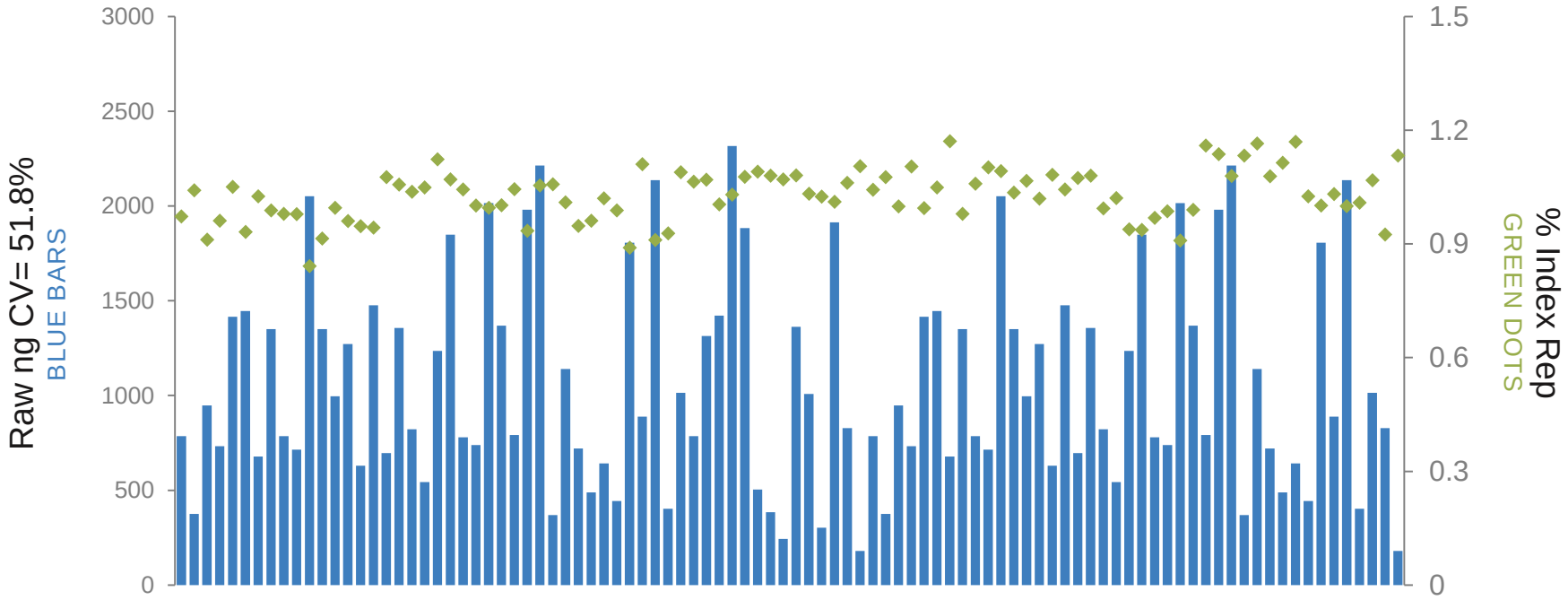


½ loop from culture

*Internal data on file

Sample input versus index representation after data analysis

Index rep CV = 6.5%



illumina

NextSeq 2000

Deeper, larger studies, on a benchtop platform.

NextSeq 1000/2000
P2 Flow Cell

120 Gb

Max output

NextSeq 2000
P3 Flow Cell

330 Gb

Max output

RELEASED TODAY

illumina

Current NextSeq 2000 Customers

Access new features through simple customer installable upgrades

New P2 & P3 consumables

NextSeq™ 2000 system
(ICS v1.1.0)

ICS v1.2.0

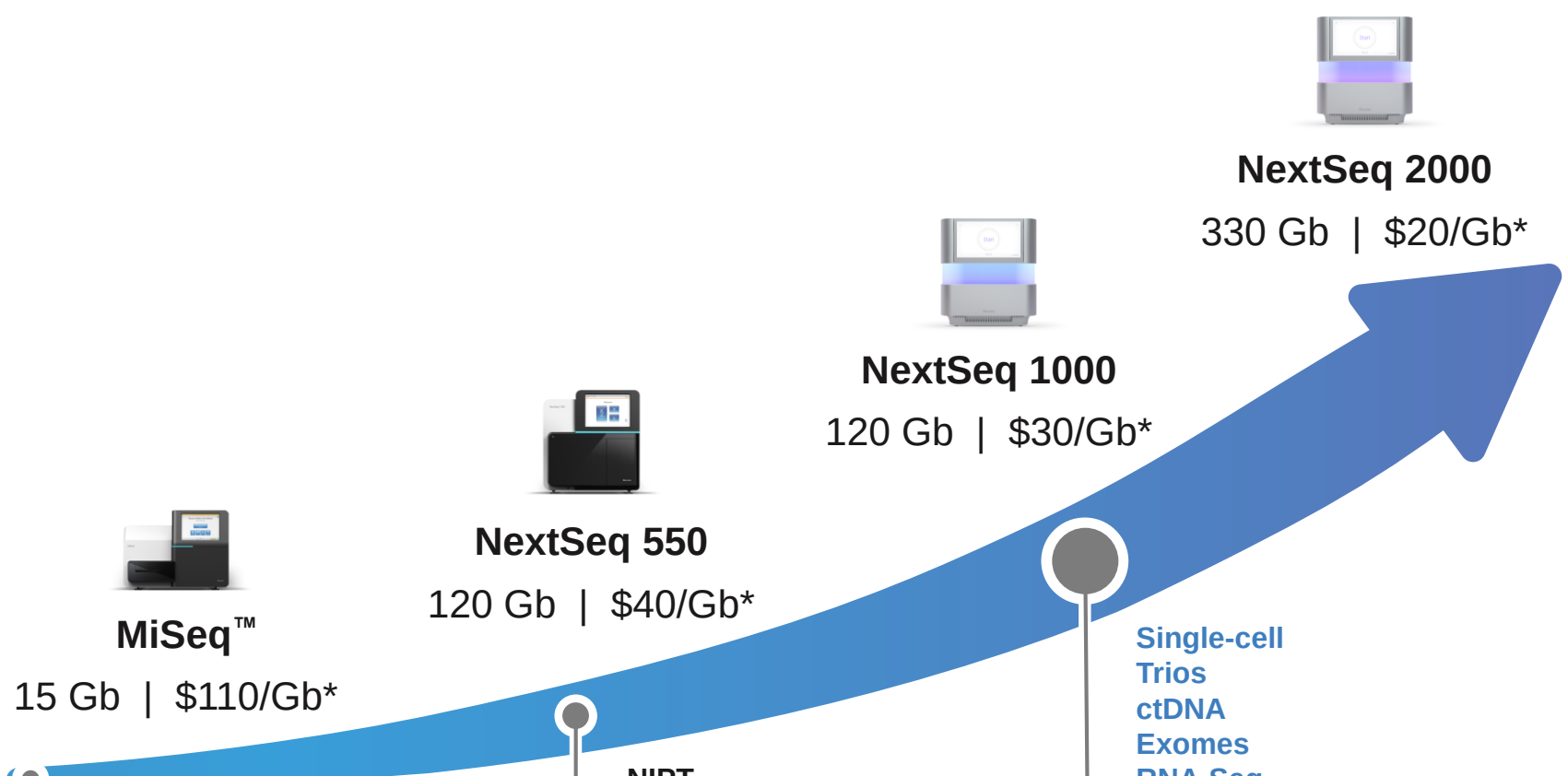
DRAGEN v3.7.4

Custom primers & recipes

New DRAGEN analysis pipelines

On board data compression

Nextseq 1000 And Nextseq 2000 Provide Sequencing Power for High-throughput Applications



MiSeq™
15 Gb | $110/Gb*

NextSeq 550
120 Gb | $40/Gb*

NextSeq 1000
120 Gb | $30/Gb*

NextSeq 2000
330 Gb | $20/Gb*

Panels
Small genomes

NIPT
Oncology Panels
Enrichment
Panels
Small genomes

Single-cell
Trios
ctDNA
Exomes
RNA-Seq
NIPT
Oncology Panels
Enrichment
Small genomes

More Samples

New Applications

Greater Depth

illumina®

* Estimated prices provided for comparison

Addition of P3 50 Cycle Kit

Kit	List	$/G (list)	$/M read
P2 100 cycles (40G)	$1,420	$35.50	$3.55
P2 200 cycles (80G)	$2,670	$33.38	$6.68
P2 300 cycles (120G)	$3,540	$29.50	$8.85
P3 50 cycles (55G)	$2,250	$40.91	$2.05
P3 100 cycles (110G)	$3,250	$29.55	$2.95
P3 200 cycles (220G)	$4,500	$20.45	$4.09
P3 300 cycles (330G)	$6,000	$18.18	$5.45

Applications

Infectious disease:
Small genome characterization (Currently not validated for COVIDSeq)

Proteomics:
Antibody-linked oligo tags BioLegend Cite Seq.

Spatial transcriptomics:
NanoString's GeoMx (27bp)

Small RNA analysis:
50 bp reads will sequence most small RNAs

Other counting applications

NextSeq 2000 is the First Systems to Integrate DRAGEN Bio-IT Platform On-Board

DRAGEN Bio-IT platform:

- Fast
- Accurate
- Cost efficient
- Industry standard pipelines
- Great for both novice and expert users.

Pipelines available on-board at launch:

- Dragen Enrichment pipeline
- Dragen RNA pipeline
- Dragen Germline
- Generate FASTQ
- *Additional pipelines available in BSSH*

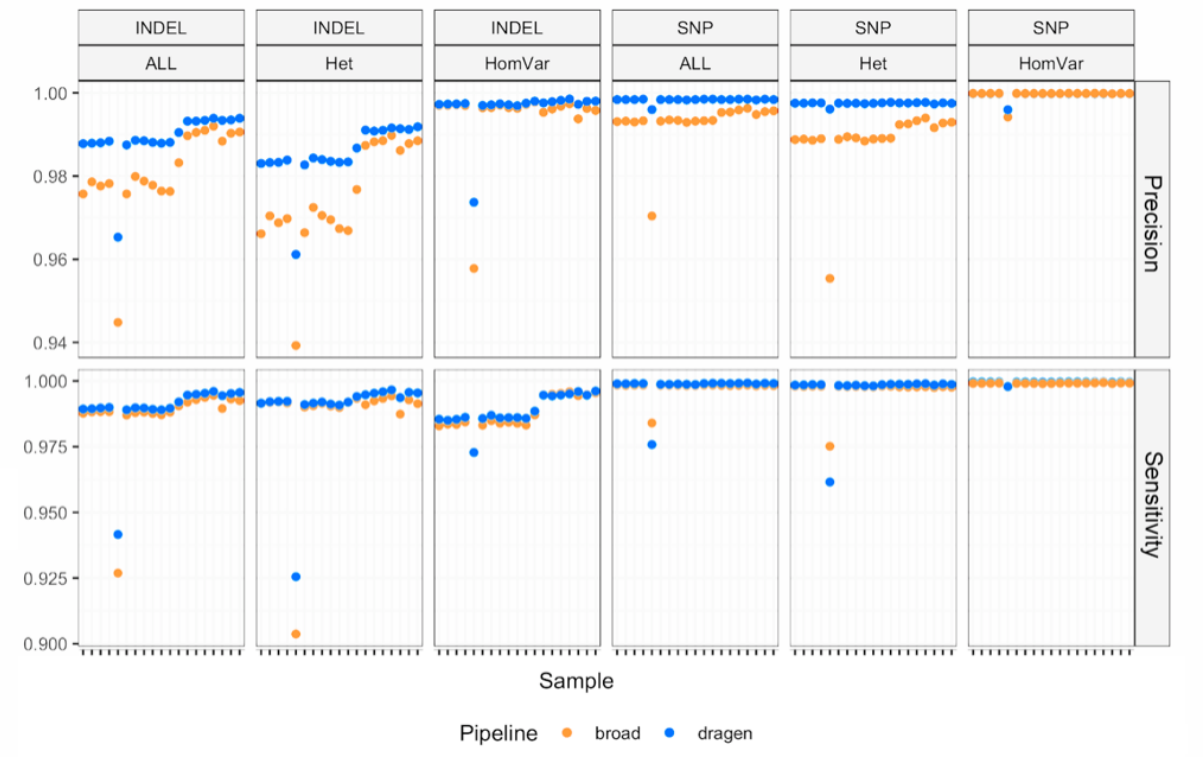
DRAGEN™ wins PrecisionFDA Truth Challenge V2 for Difficult-to-Map regions and All Benchmark Regions on Illumina sequencing data

illumina®

illumina®

The GATK team's evaluation confirmed DRAGEN™ accuracy gains

”..we wanted to do our own evaluation … and long story short, we saw the same overall improvements in sensitivity and specificity, which you can see for yourself in the figure below.”



“finally got a taste of that famous acceleration -- yeah it is *fast*, no kidding”

THE GATK TEAM

Content published on Feb. 19, 2020 DRAGEN demonstrated gains in indel and heterozygous SNP calling precision & Increase in indel sensitivity
https://gatk.broadinstitute.org/hc/en-us/articles/360039984151-DRAGEN-GATK-Update-Let-s-get-more-specific

illumina®

DRAGEN Single Cell RNA on NextSeq2000

Outputs functional starting point for downstream single cell analysis: Cell x Gene Expression Matrix

FEATURES

Ultra-rapid
Accurate analysis in < 2 hours for a full NextSeq 2000 P3 flow cell

Widely compatible
Supports a range of input library prep types for compatibility with downstream analysis tools

Set up and walk away
Goes from run set up to quantified expression per cell with a single touch point

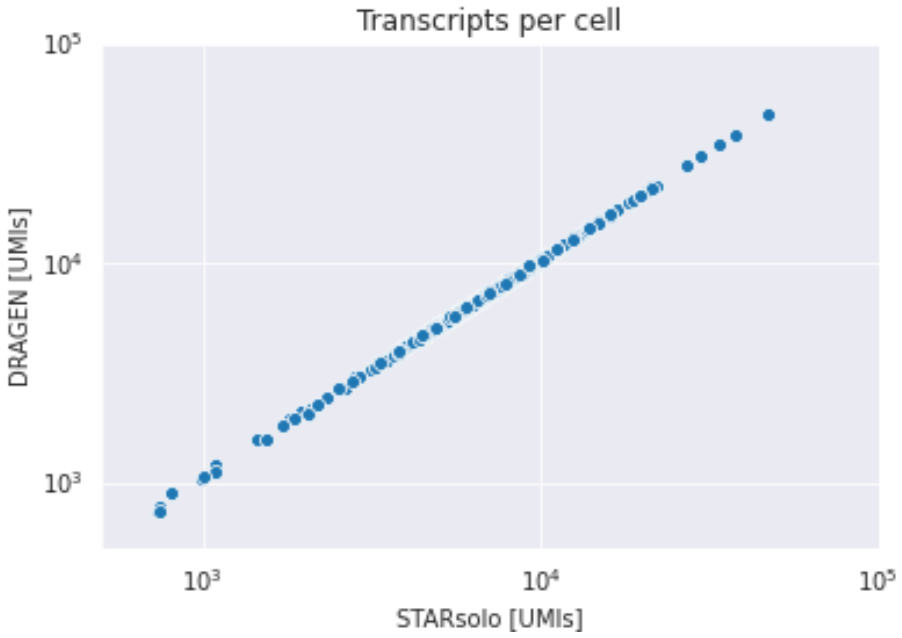
BENEFITS

Accelerate your research
On-board QC of single cell expression libraries and single cell analysis pipeline executed in a fraction of the time – without the need for additional compute hardware or sacrificing accuracy

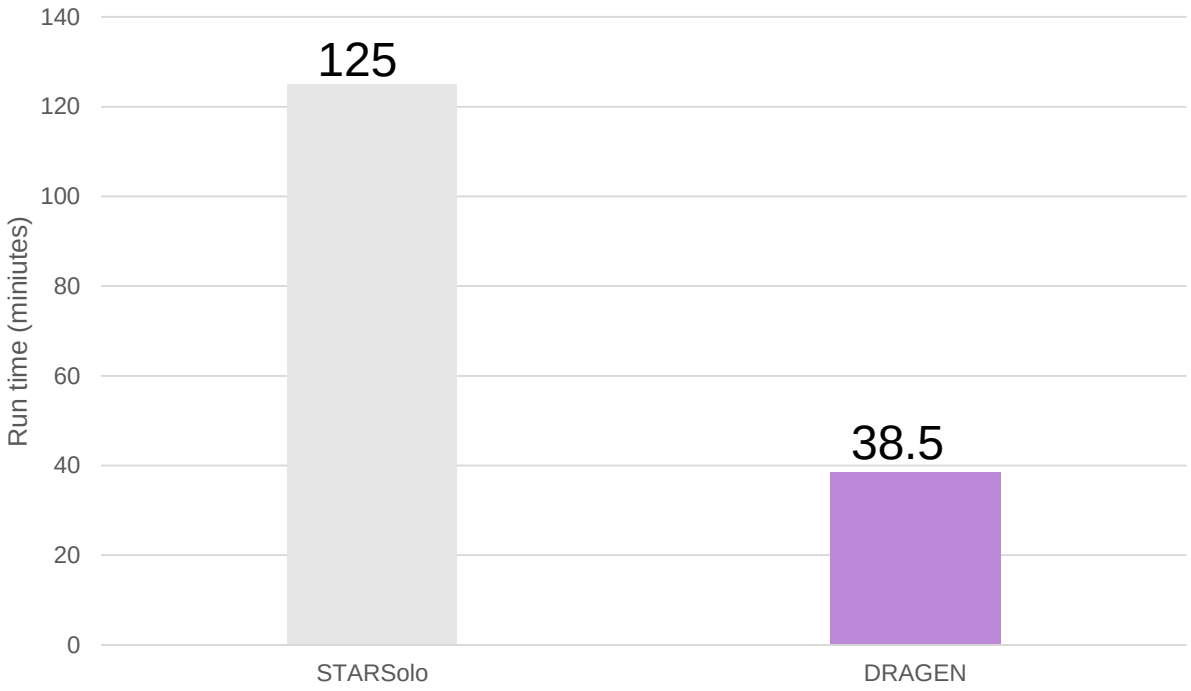
Choose your tools
Cell x gene matrix in an open data format supports a variety of library prep and downstream single cell analysis tools

Streamline your work
Eliminate the set up & execution of a separate pipeline post-sequencing

Consistent with Established Tools, Yet Much Faster

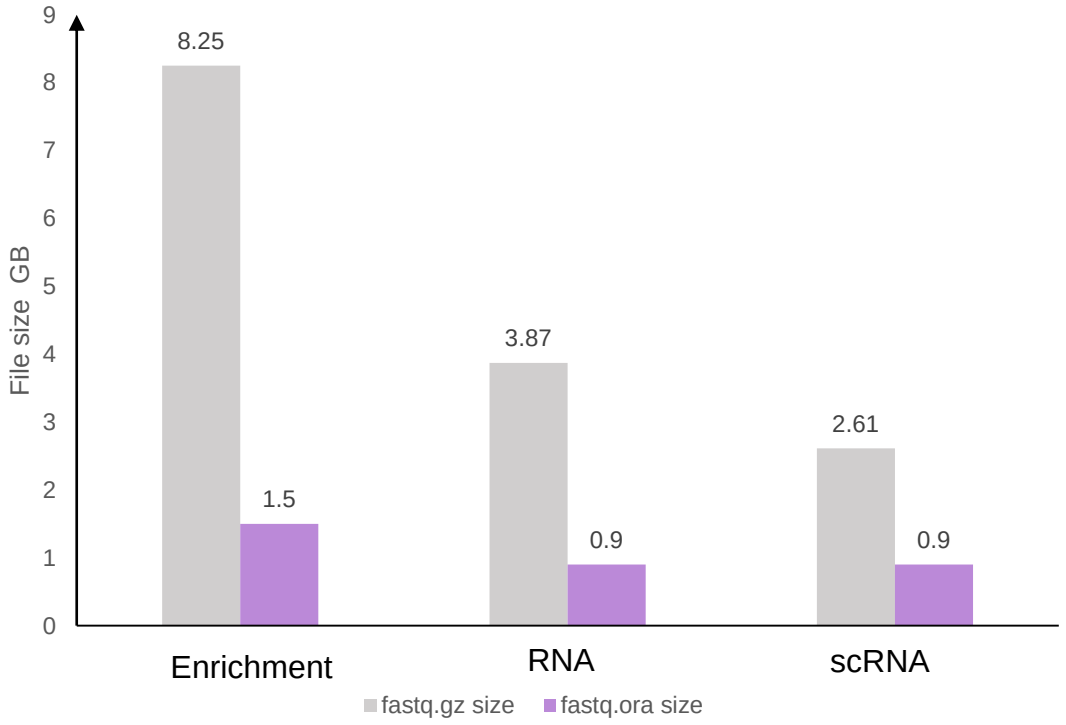


Correlation of per-cell gene expression results with STARsolo: r > 0.985
857 / 858 cells overlap



8018 cell sample with 1.4 billion reads

New: DRAGEN Compression Reduces Data Storage Costs



Save ~ $3,000 - $10,000 on FASTQ storage costs*

*Compared to storing fastq.gz. Assumptions: 36 to 125 runs per year, 300 GB fastq.gz file sizes, compression ratio of 5, files stored 1 year in hot storage + 2 years in cold storage on AWS.

Reduce FASTQ file transfer times from 25 minutes to 5 minutes**

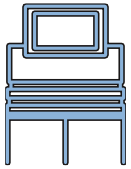
**Compared to transferring fastq.gz . Assumptions: 200 MB/s file transfer speeds and 300 GB fastq.gz file

Software license included in NextSeq 1000/2000 purchase

Illumina Analysis Platform (IAP) is a Comprehensive Platform to Drive Insights

Flexible, interoperable components supporting life sciences research

Sequence	Analyze	Aggregate	Explore
• Out-of-the-box instrument integration • Data management & permissions	• DRAGEN pipelines • Bring-your-own tools • Customize analytical pipelines • Business analytics	Aggregate NGS and metadata to derive cohort-level insights	Dynamic data analysis with Jupyter notebooks and R Studio



BlueFlow
NGS analysis + data portal

BlueBase
Secure data warehouse

BlueBench*
Data science + AI Workspace

Illumina Analytics Platform

Data Production

Data Trending & Mining

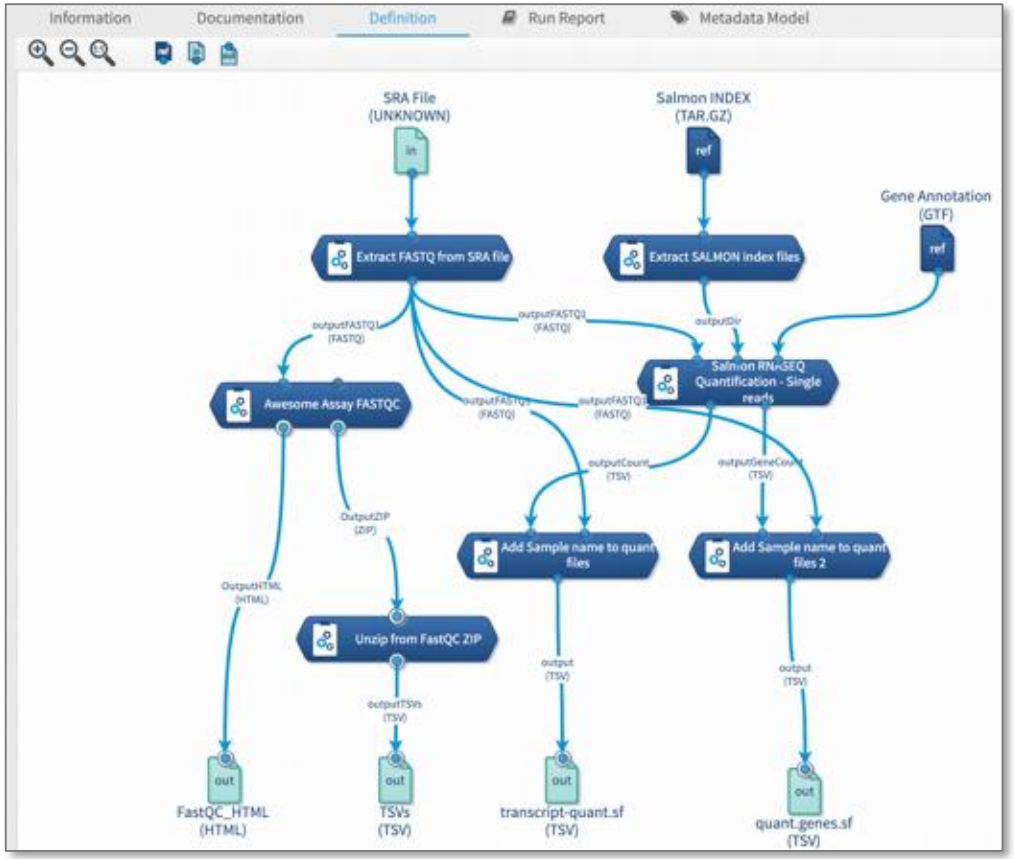
Data Science

illumina®

Interactive Portals & Data Science Workbench

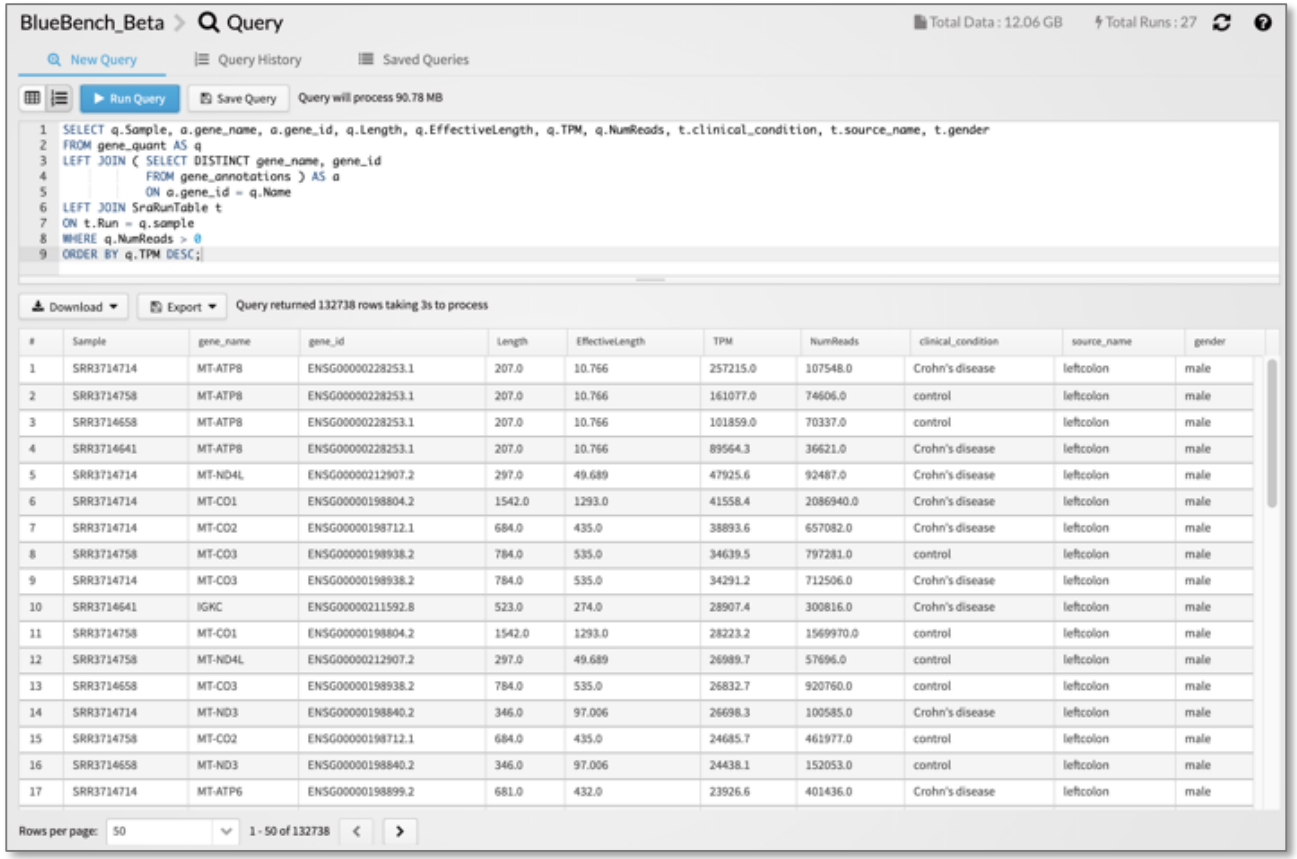
Analyze

Drag & Drop Pipeline Creator



Aggregate

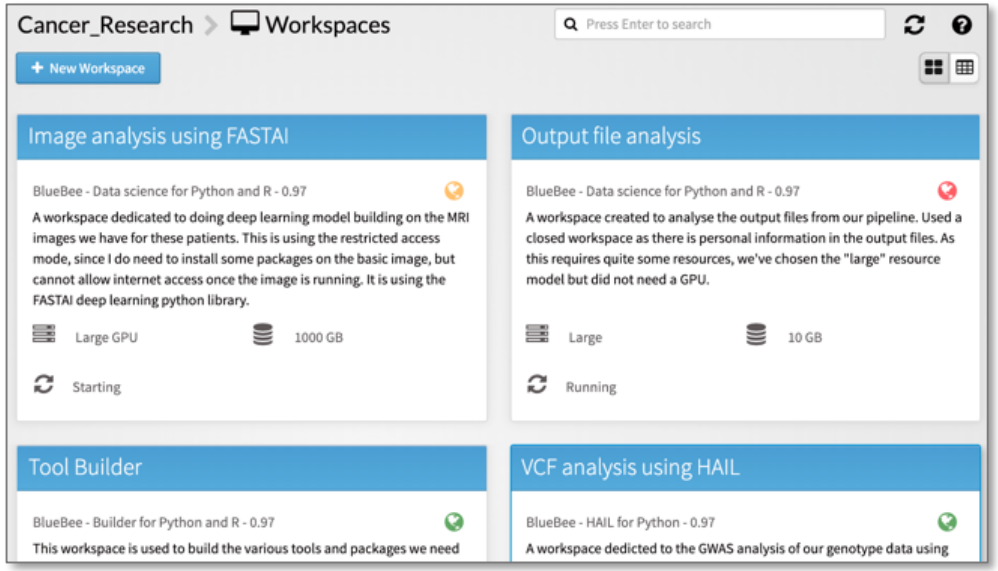
SQL Query of Data Warehouse

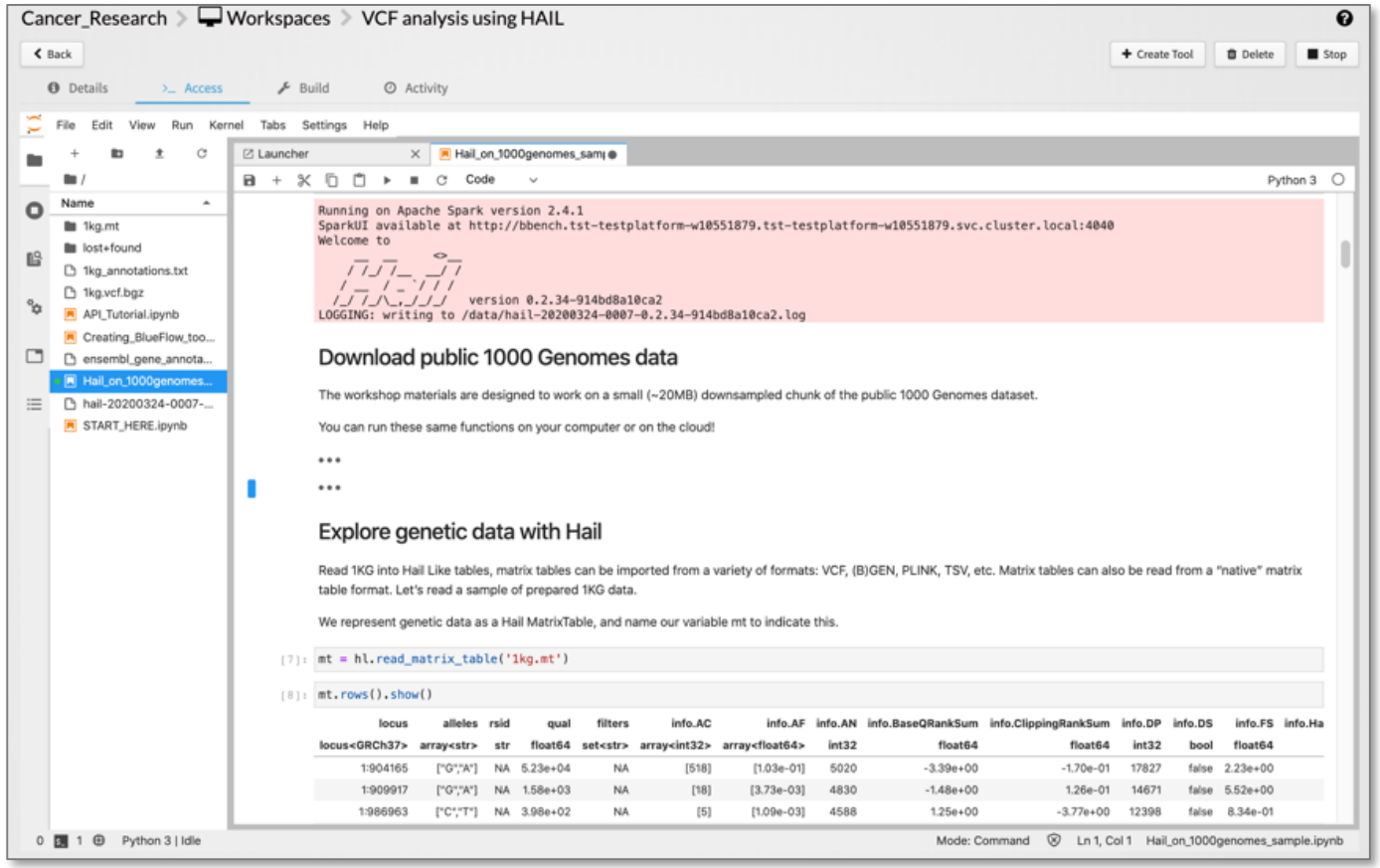


Interactive Portals & Data Science Workbench

Explore

Data Science Workbench with Notebooks





illumina®

Thank You!

Frankie Gwynne

Account Manager

fgwynne@illumina.com

Paige Burres

Inside Sales

pburres@illumina.com

Ryan Hegarty

Sequencing Specialist

rhegarty@illumina.com