



# Sequencing Technologies

Michael Quail  
[mql@sanger.ac.uk](mailto:mql@sanger.ac.uk)





# Short Read Sequencers



Omniome



Element



Ultima UG100



Illumina  
Nextseq

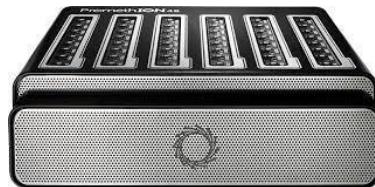


Singular  
S4

# Long Read Sequencers



PacBio Revio



Oxford Nanopore P48



Oxford Nanopore gridION



PacBio Vega

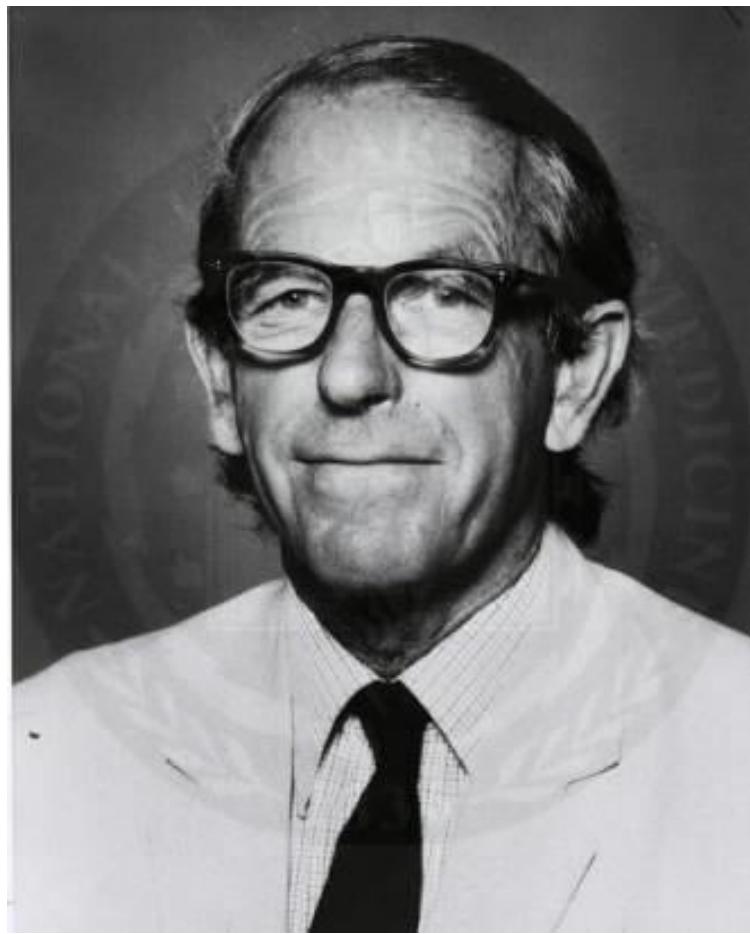


Oxford Nanopore minION

Generate much longer reads that enable more of the genome to be covered  
Generally more expensive than short read, with higher input requirements,  
Single molecule so yields dependent on DNA quality

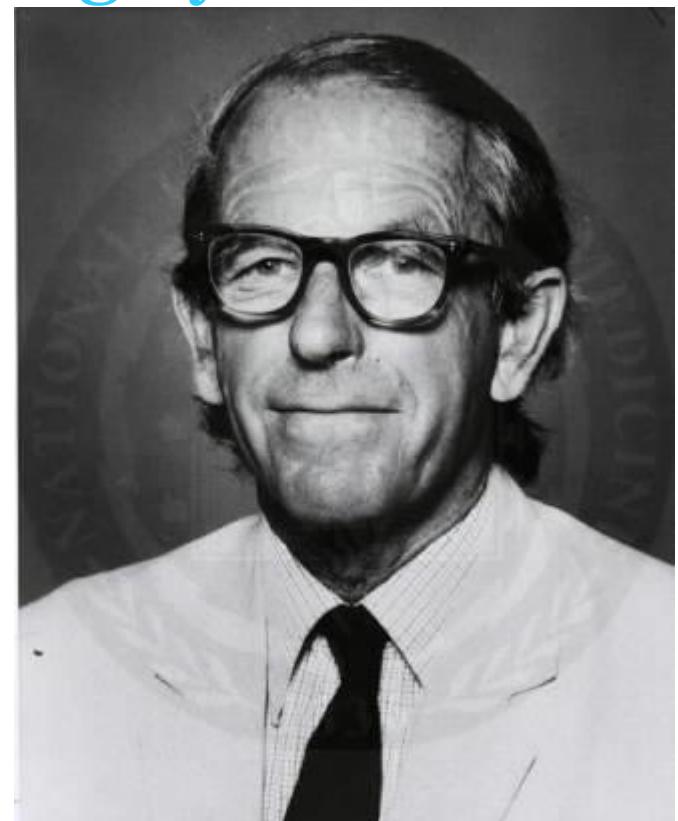


# Sanger Sequencing

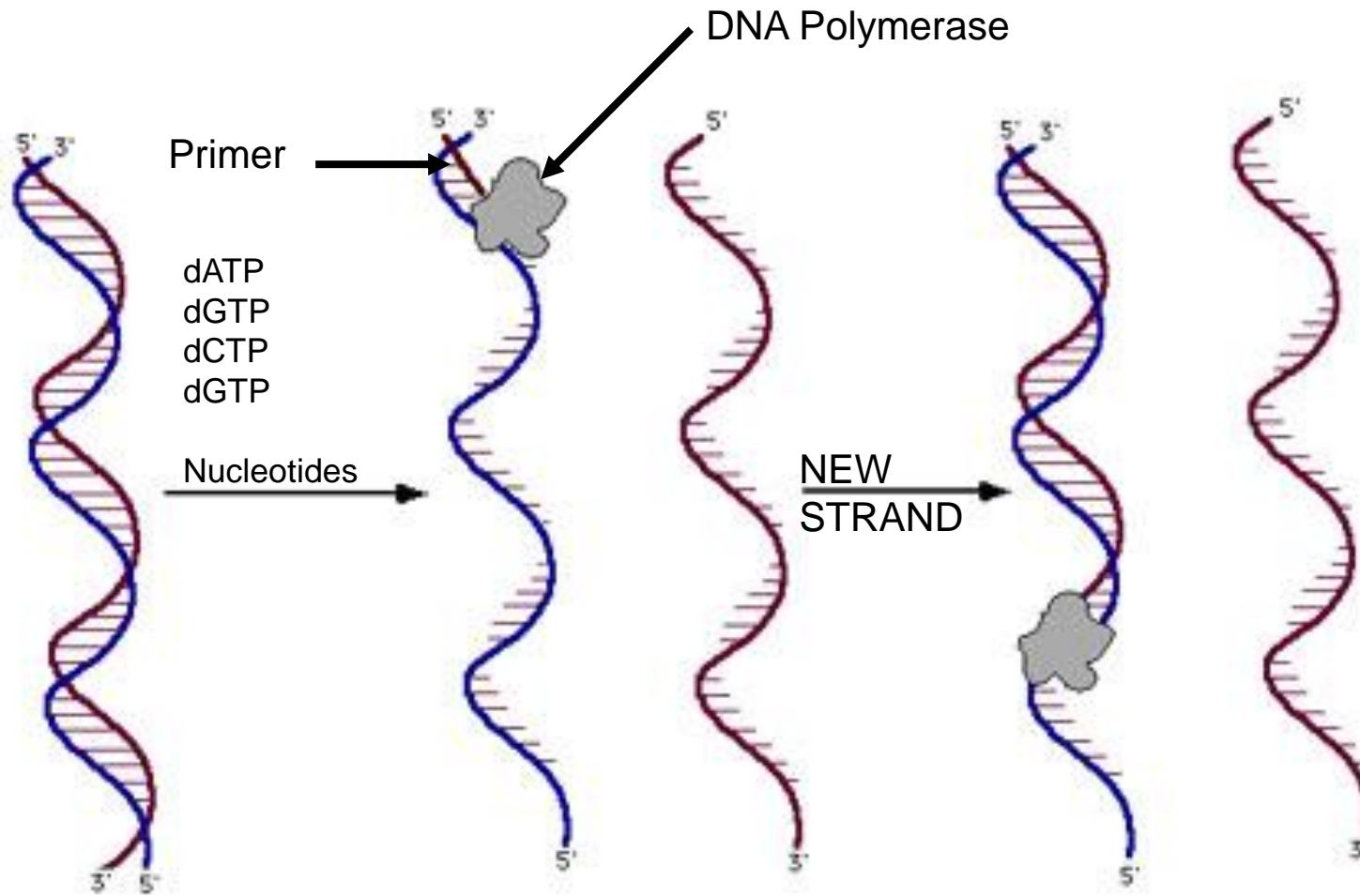


# Frederick Sanger

- » Discovered DNA sequencing by chain termination method
- » Nobel Prize 1 (1958)
  - » Complete amino acid sequence of insulin
- » Nobel Prize 2 (1980)
  - » For DNA sequencing



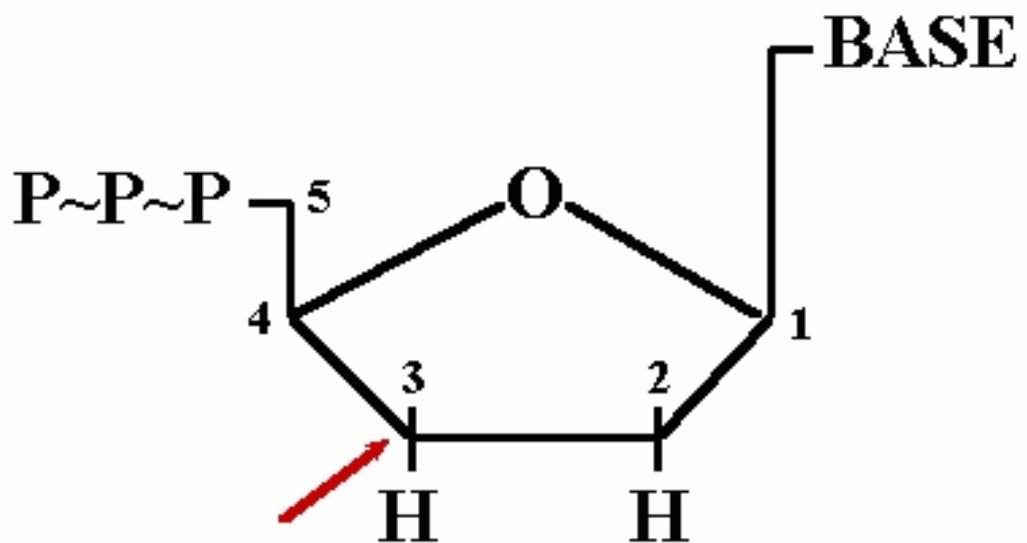
# Primer Extension



Template DNA

# Dideoxy Nucleotides

- Lack an -OH group at the 3-carbon position
- Cannot add another nucleoside at that position
- Prevent further DNA synthesis



# All Possible Terminations

DNA Polymerase reads the template strand and synthesizes a new second strand to match:

5' - TACGCGGTAAACGGTATGTTGACCGTTAGCTACCGAT  
3' - ATGCGCCATTGCCATACARGCTGGCATAATCGATGGCTAGAGATCCAA - 5'



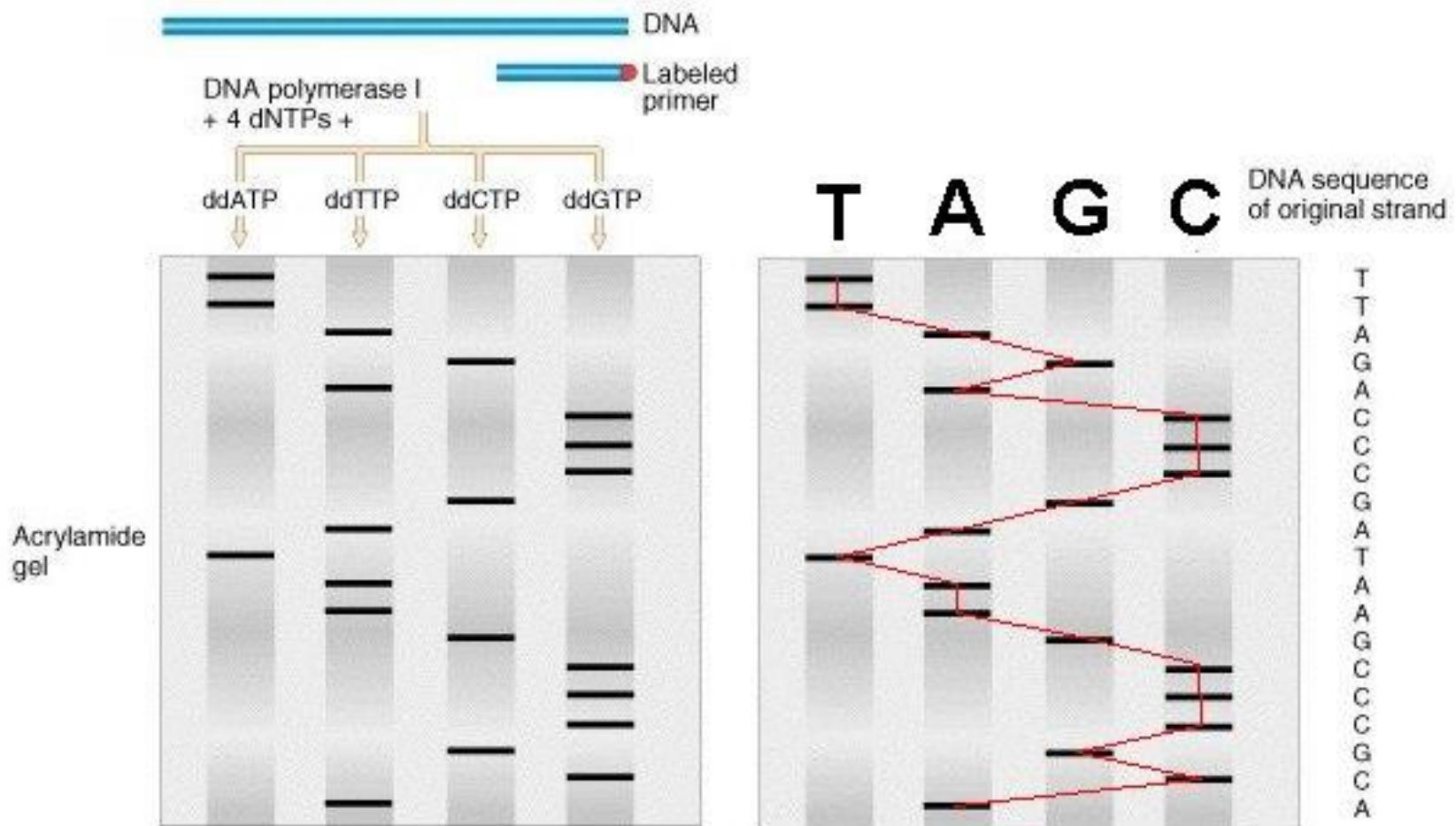
IF 5% of the T nucleotides are actually dideoxy T, then each strand will terminate when it gets a ddT on its growing end:

5' - TACGCGGTAAACGGTATGTTGACCGTTAGCTACCGAT•  
5' - TACGCGGTAAACGGTATGTTGACCGTTAGCT•  
5' - TACGCGGTAAACGGTATGTTGACCGTT•  
5' - TACGCGGTAAACGGTATGTTGACCGT•  
5' - TACGCGGTAAACGGTATGTTGACCGT•  
5' - TACGCGGTAAACGGTATGTT•  
5' - TACGCGGTAAACGGTATGT•  
5' - TACGCGGTAAACGGTAT•  
5' - TACGCGGTAAACGGT•  
5' - TACGCGGTAAACGGT•

# Original Sanger Sequencing

- » 4 sequencing reactions performed for each template, each with different terminator
- » Radioactive primer or nucleotide used
- » Sequencing reactions run on <1mm polyacrylamide gel cast between two glass plates to separate fragments according to size
- » After run gel exposed to film and developed to reveal image

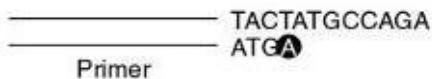
# Sequencing gel autorad



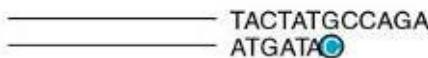
# Fluorescent Terminators

Primer extension reactions:

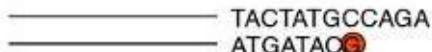
ddA reaction:



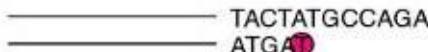
ddC reaction:



ddG reaction:

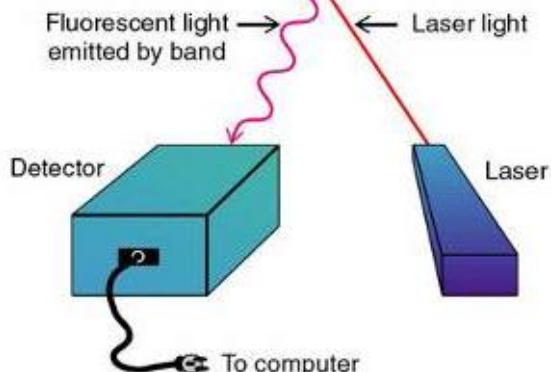


ddT reaction:

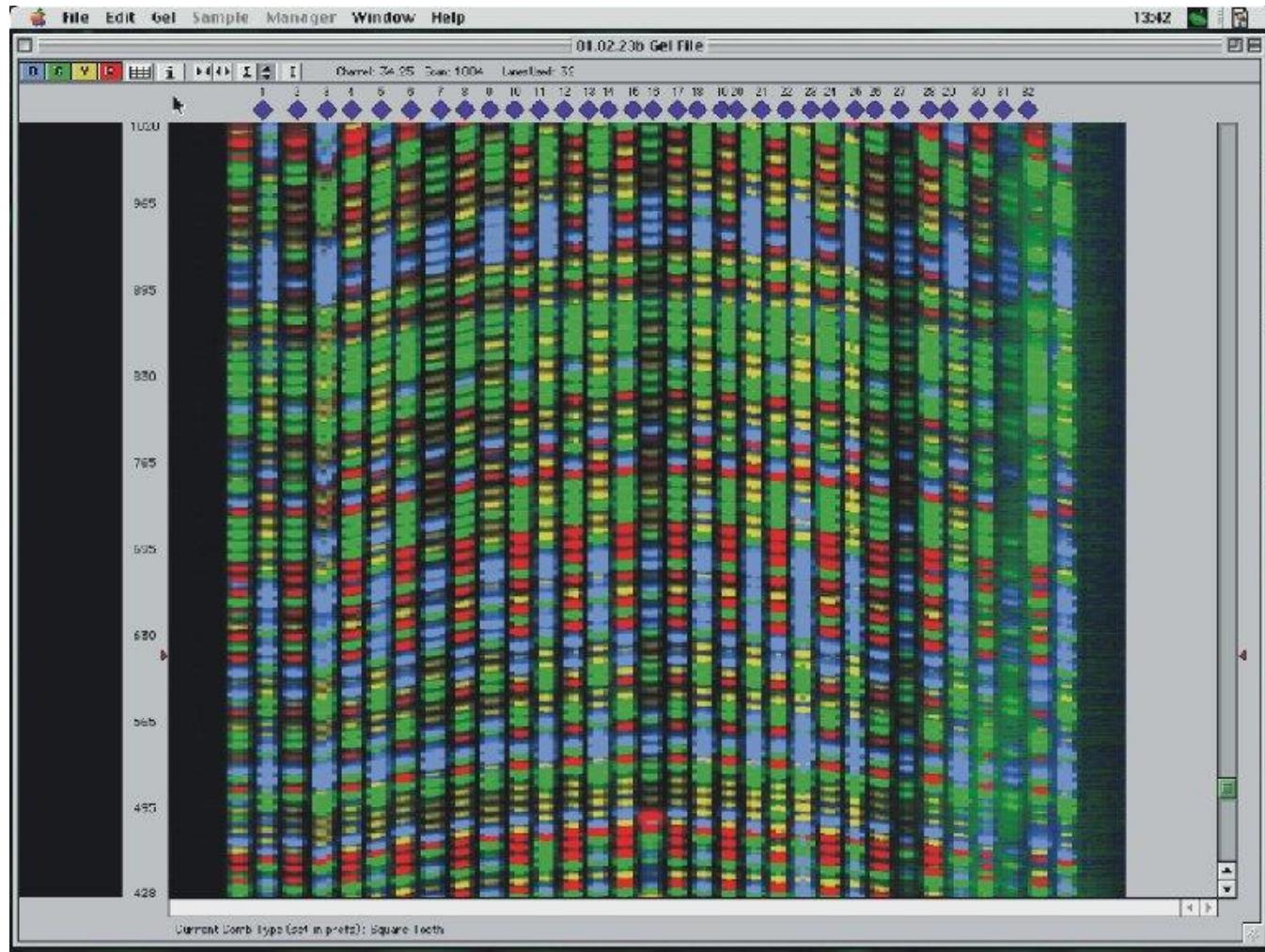


Electrophoresis:

A  
G  
A  
C  
C  
G  
T  
A  
T  
C  
A  
T



# Fluorescent Gel Sequencing

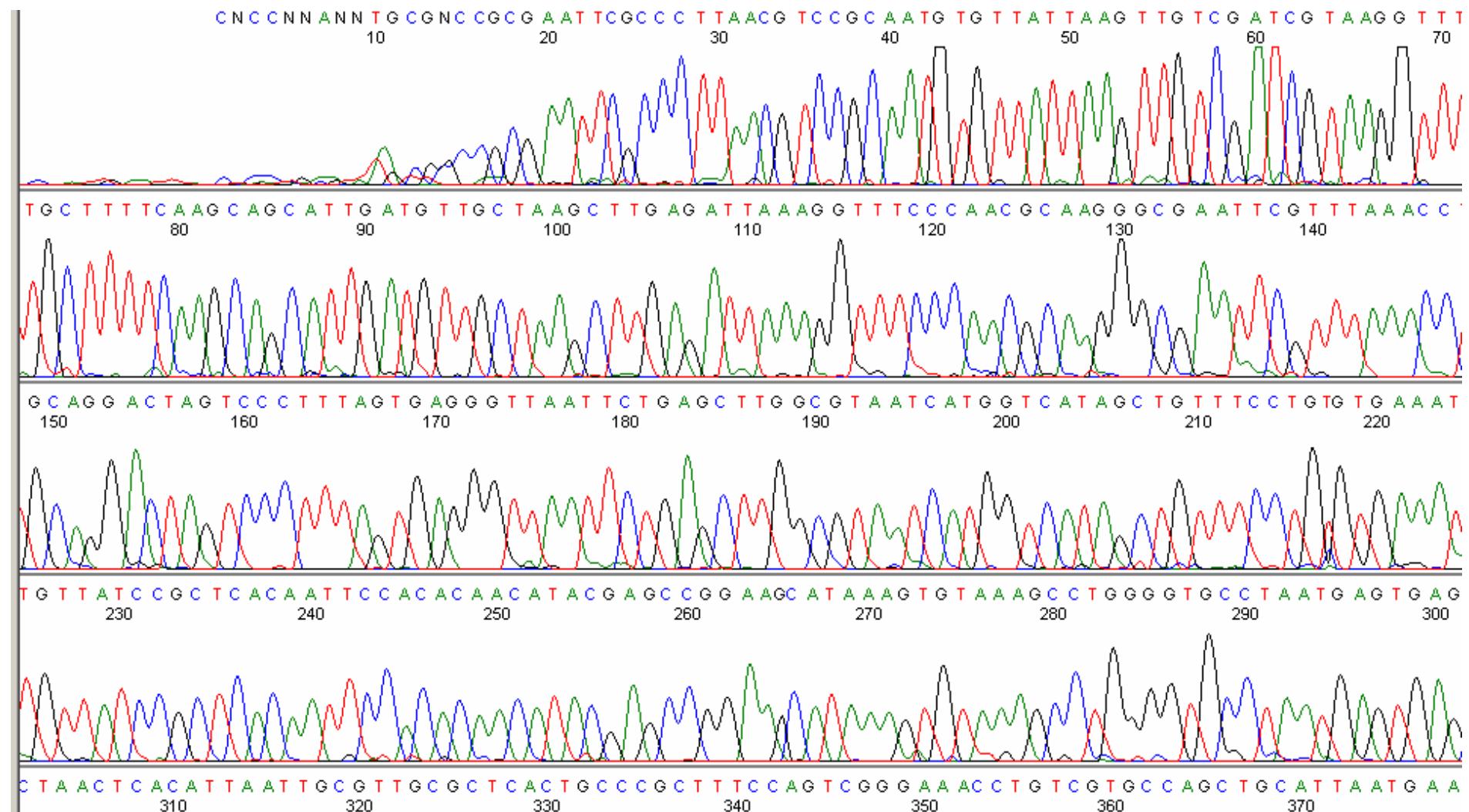




# ABI Capillary 3730



# DNA Sequence Files





# Error

- » Sequence quality Q is reported on a log scale
- » Q<sub>10</sub> is 1 error in 10
- » Q<sub>20</sub> is 1 error in 100
- » Q<sub>30</sub> is 1 error in 1000
- » Q<sub>40</sub> is 1 error in 10000
- » Q<sub>50</sub> is 1 error in 100000

# Capillary sequencing

- » Individual reactions -> 96 capillary array
- » PCR errors
- » Cloning bias
- » 1000-base reads
- » 1-2 hour run time
- » Accurate, Q30
- » Low cost if want 1 read
- » Low throughput, expensive to sequence whole genomes

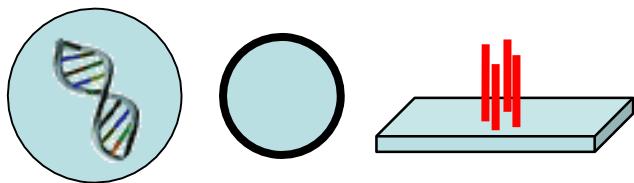




# Next Generation sequencing

Is massively parallel  
Not limited to few reactions per run

# Next-Generation Sequencing



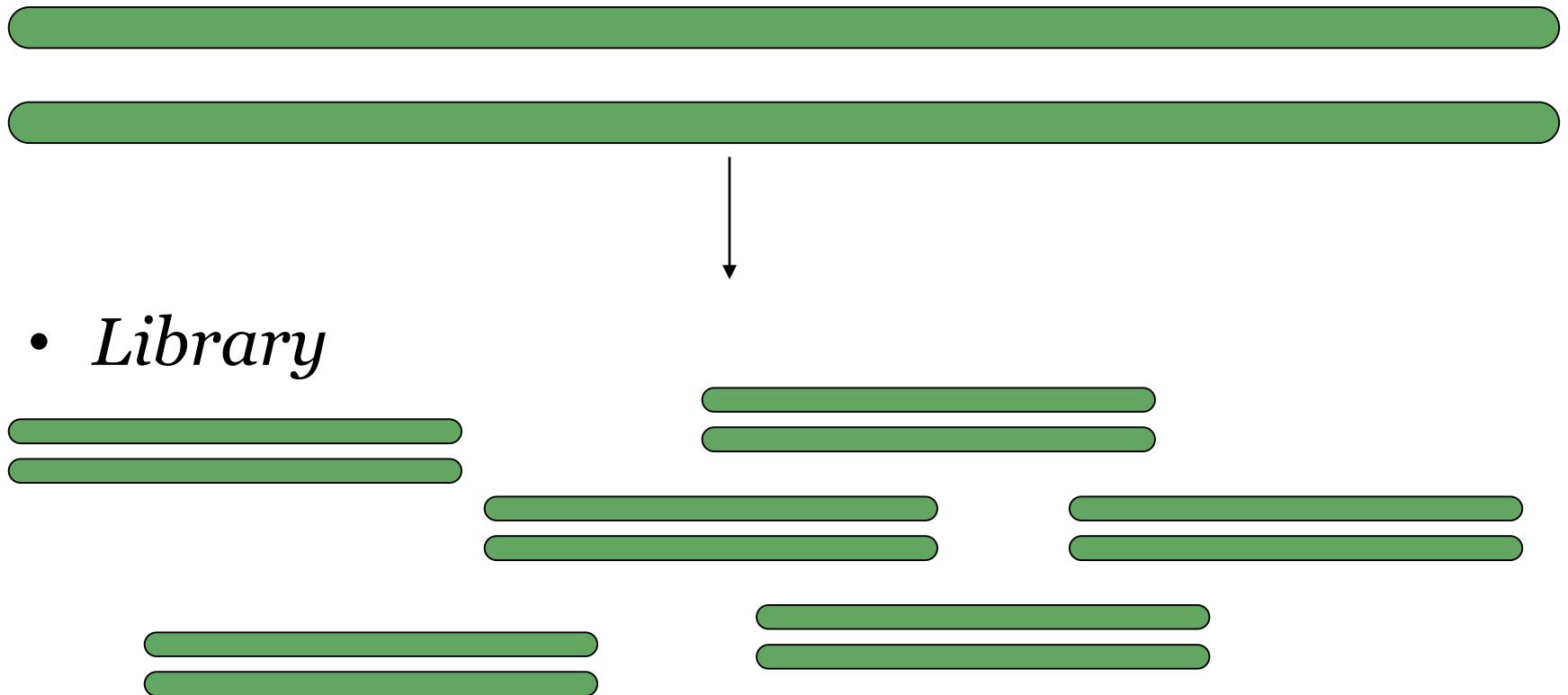
1 feature  
1 template



1 chip, thousands or  
millions of features  
Output Mb-Tb

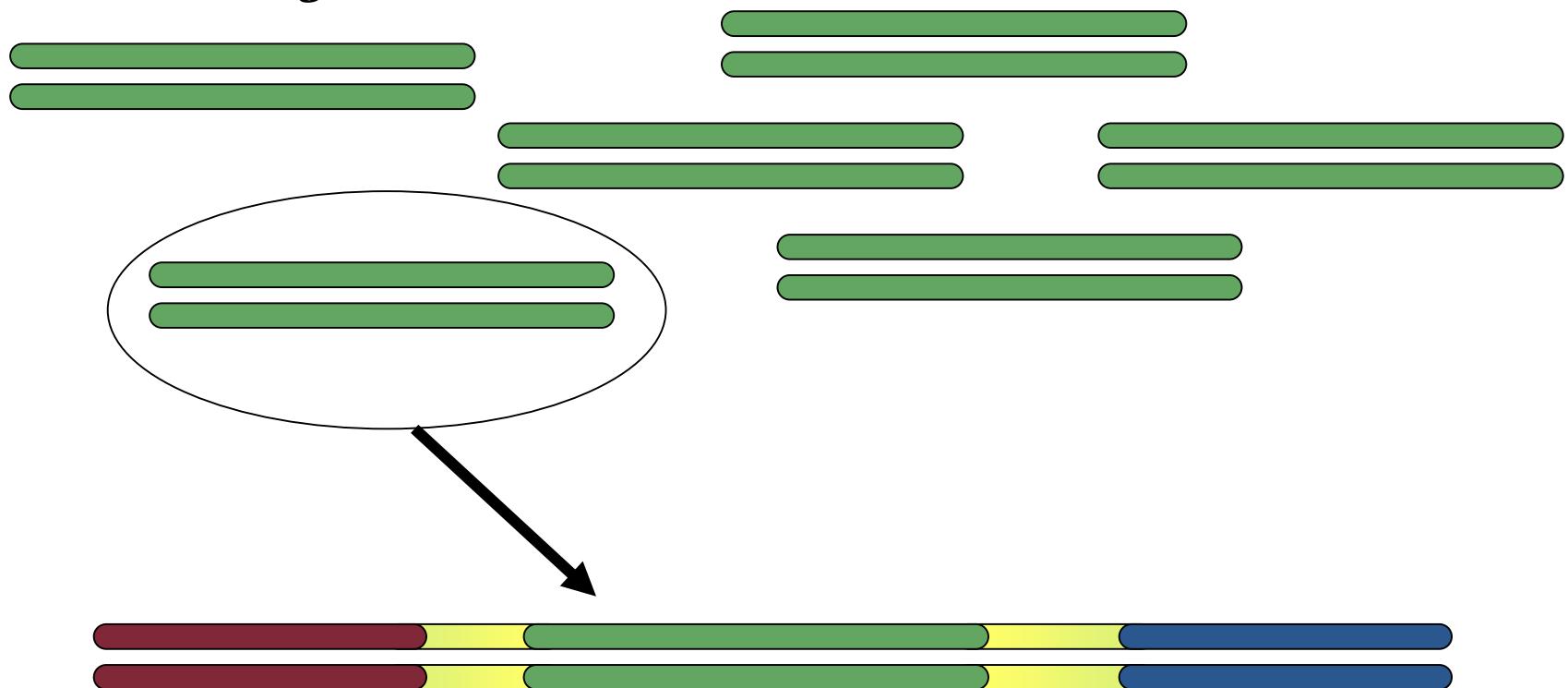


# Library prep



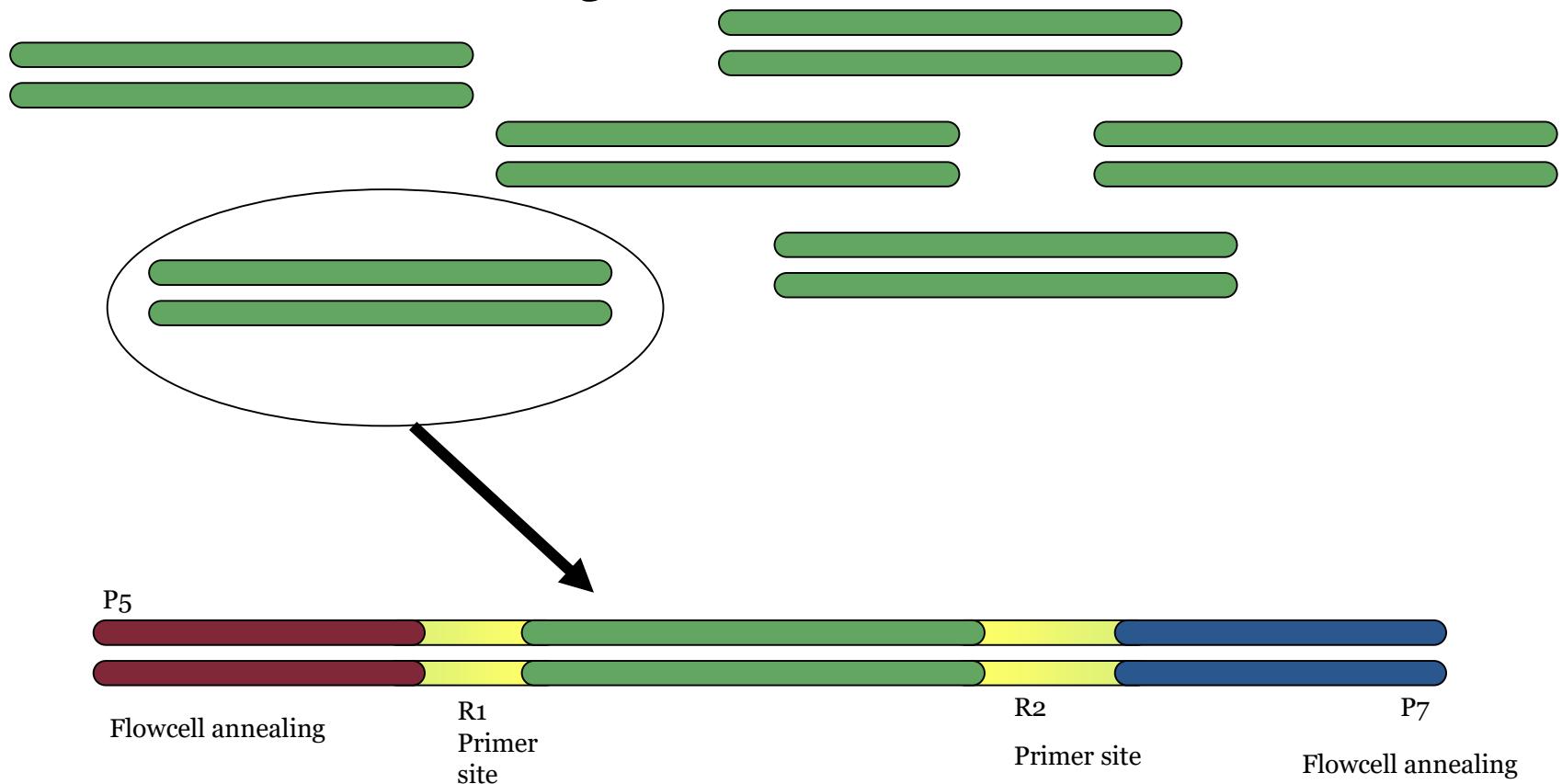
- *Library*

- *Library*



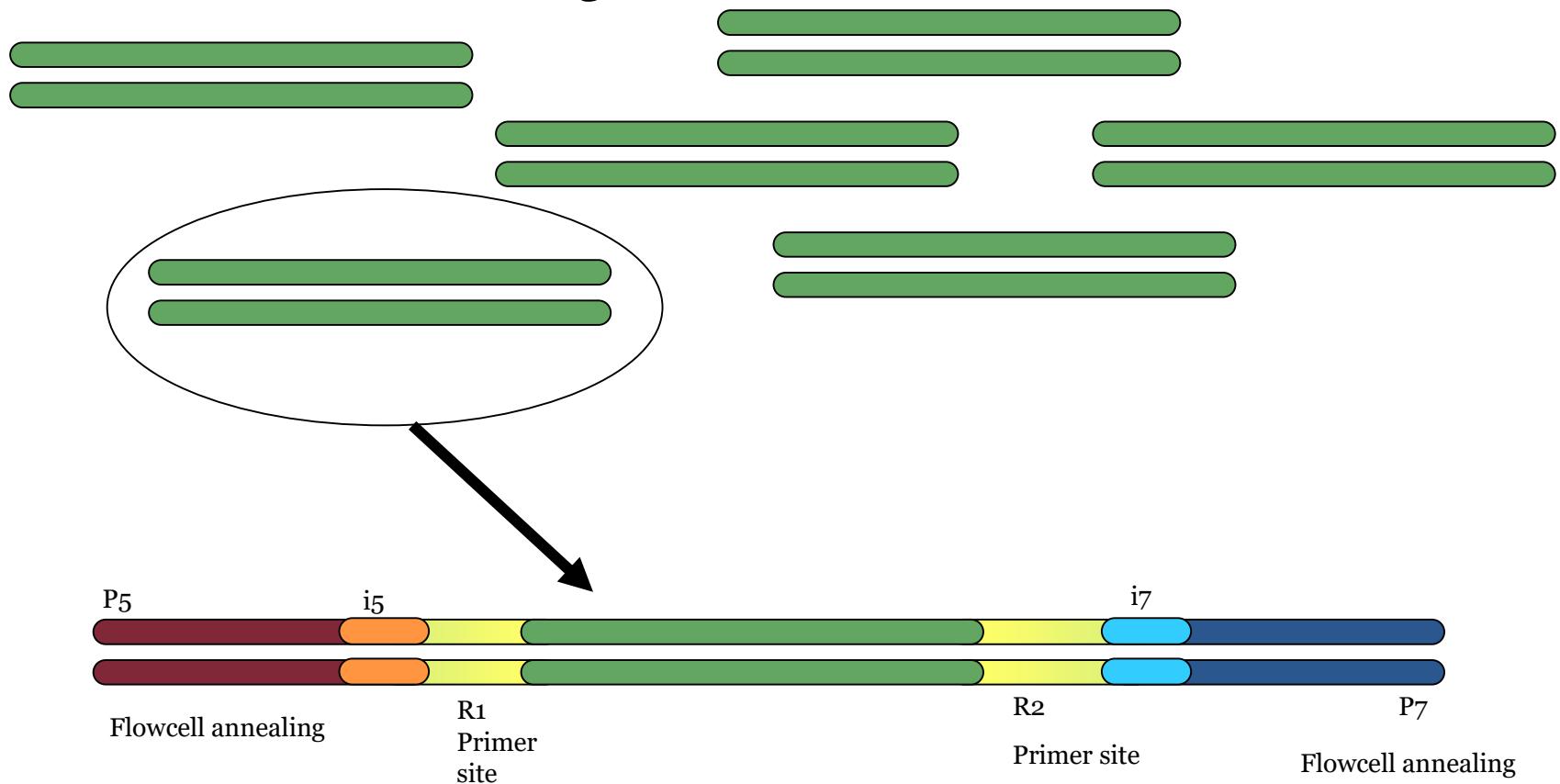


- *Illumina Library*





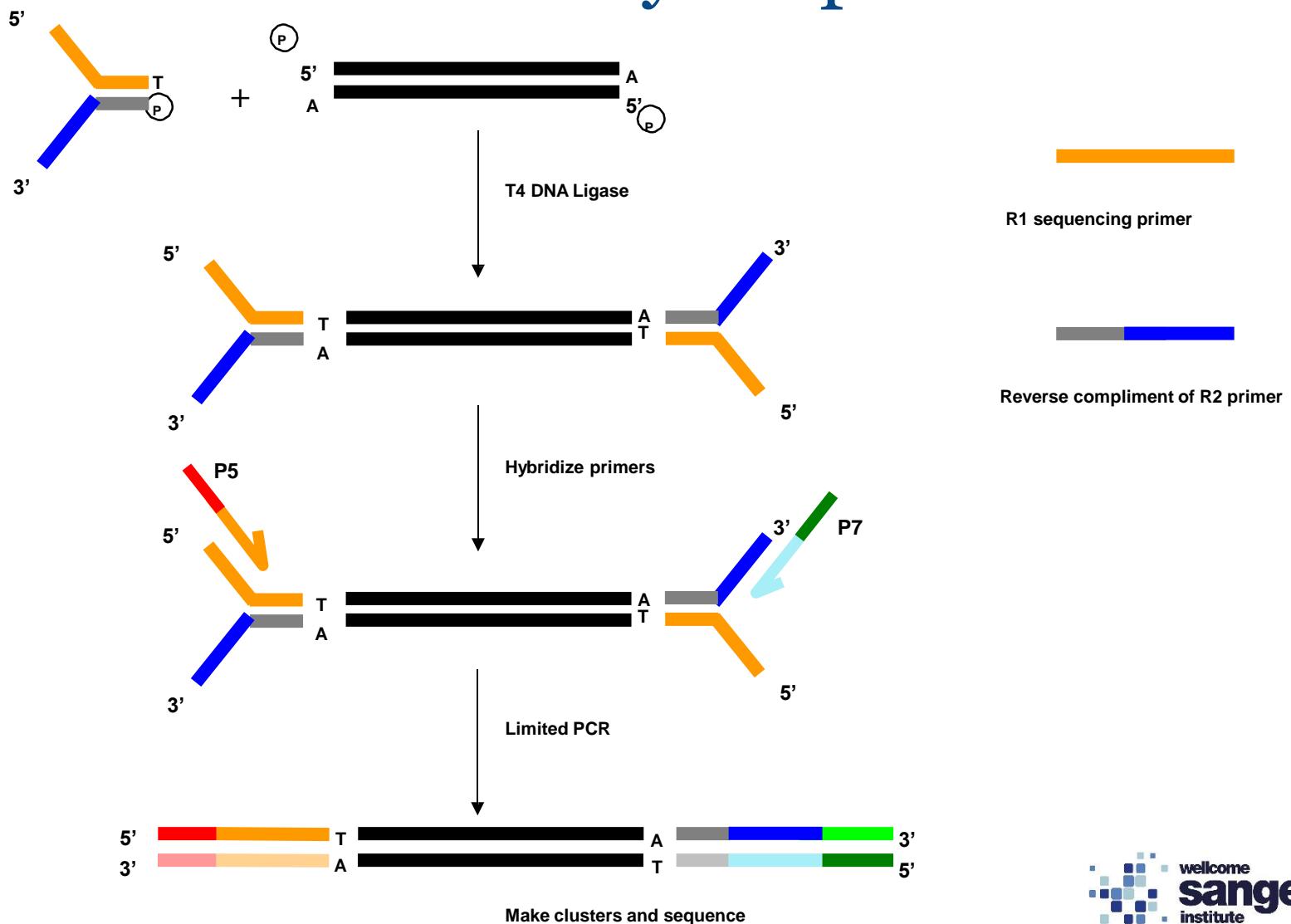
- *Illumina Library*



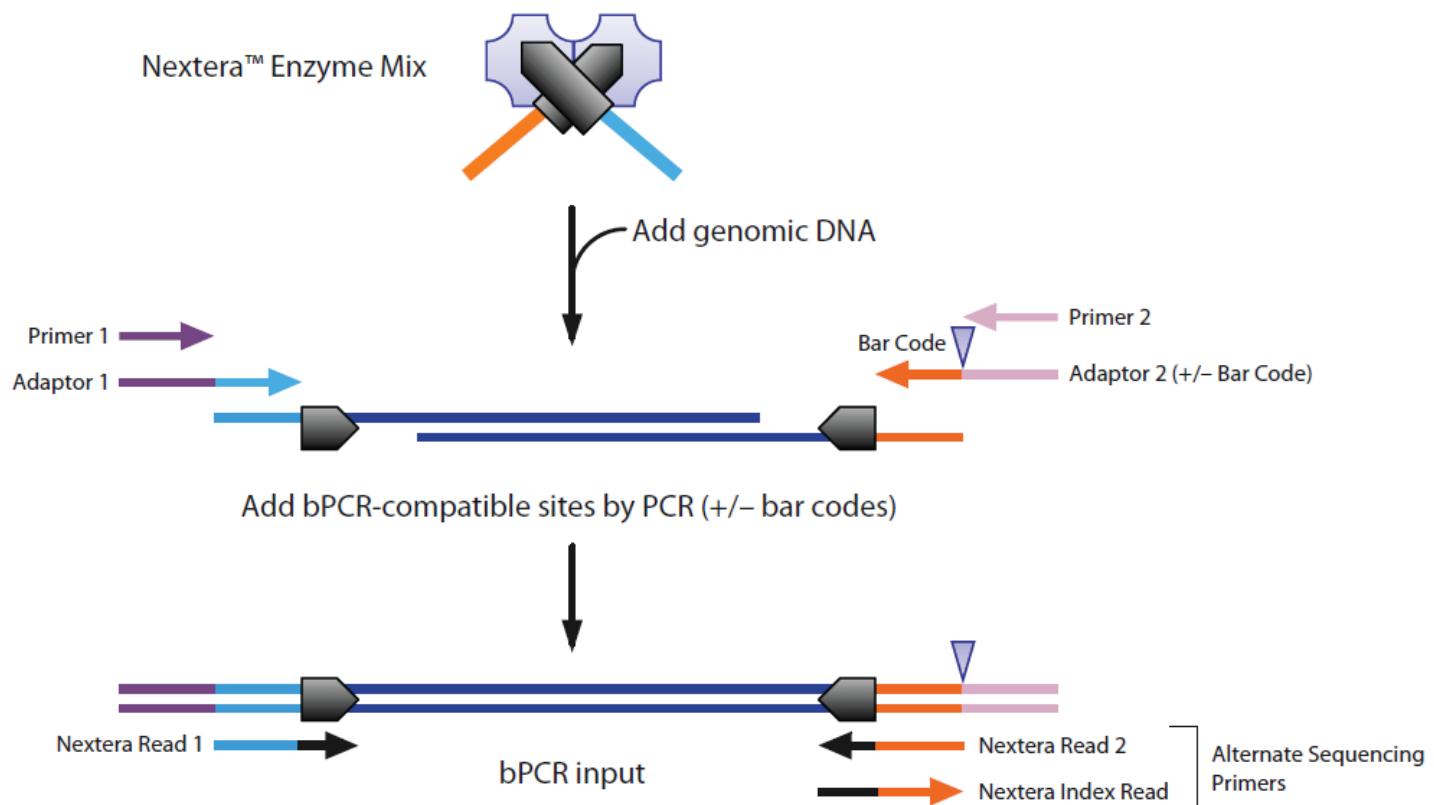
# Library Prep Approaches

1. Classical adapter ligation method
2. Transposon mediated
3. Inclusion of adapter sequences during PCR

# Illumina Paired End Library Prep

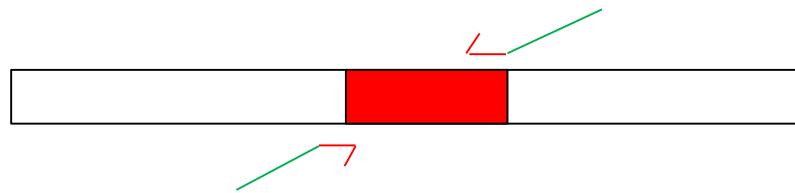


# Nextera





# Amplicon sequencing



# 454



# 454

- » Started NGS (2005)
- » First massively parallel sequencer
- » Bought by Roche in 2007. Now discontinued
- » Based on pyrosequencing of bead-bound DNA in microwells
- » Fore-runner of Ion Torrent and Genapsys



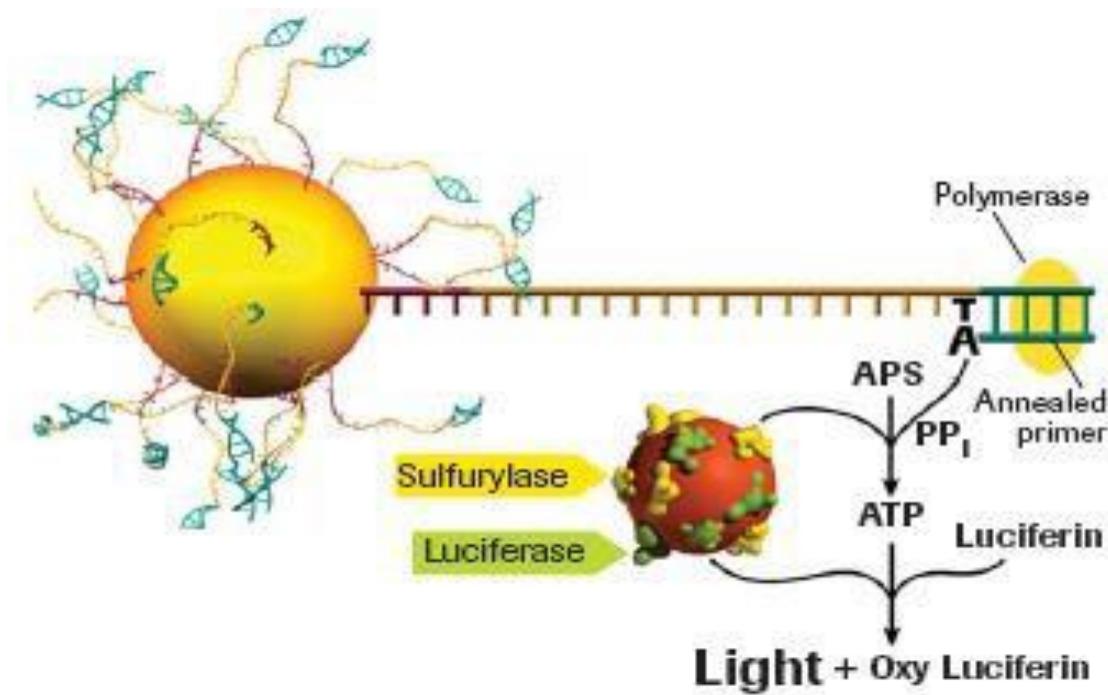
# 454/Roche Summary

- » .7 Gb / run
- » 700 base reads
- » <24 hour run time
- » \$7,000 / Gb



Roche discontinued in 2016 – Too expensive

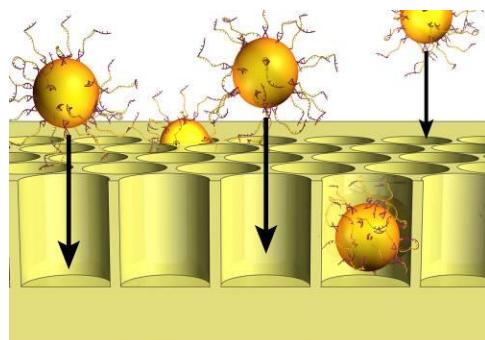
# Pyrosequencing



- » The incorporation of new bases releases inorganic pyrophosphate
- » A chemical cascade converts luciferin to oxy-luciferin + light

# Depositing DNA Beads into the PicoTiter™Plate

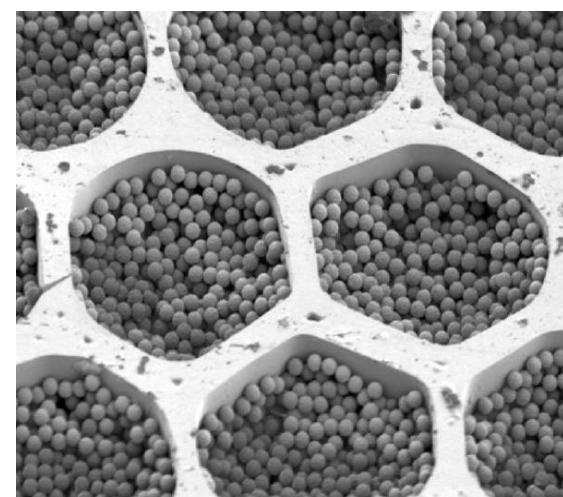
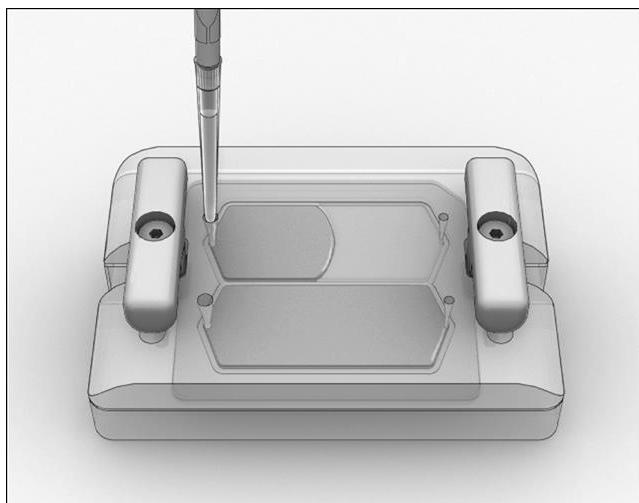
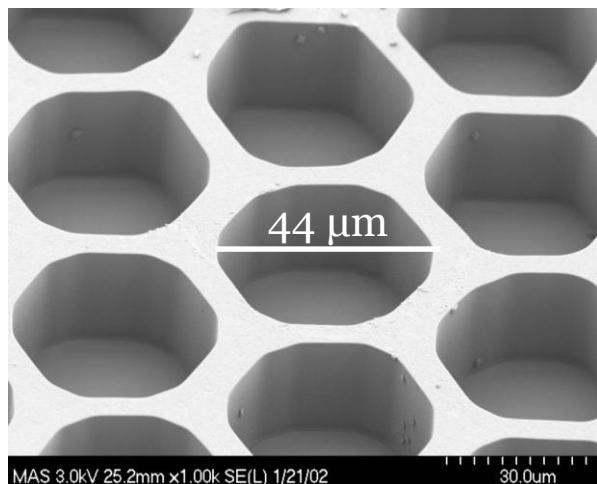
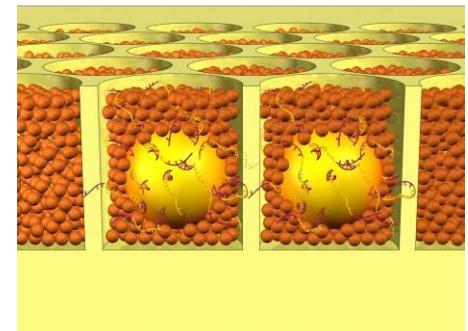
Load beads into  
PicoTiter™Plate



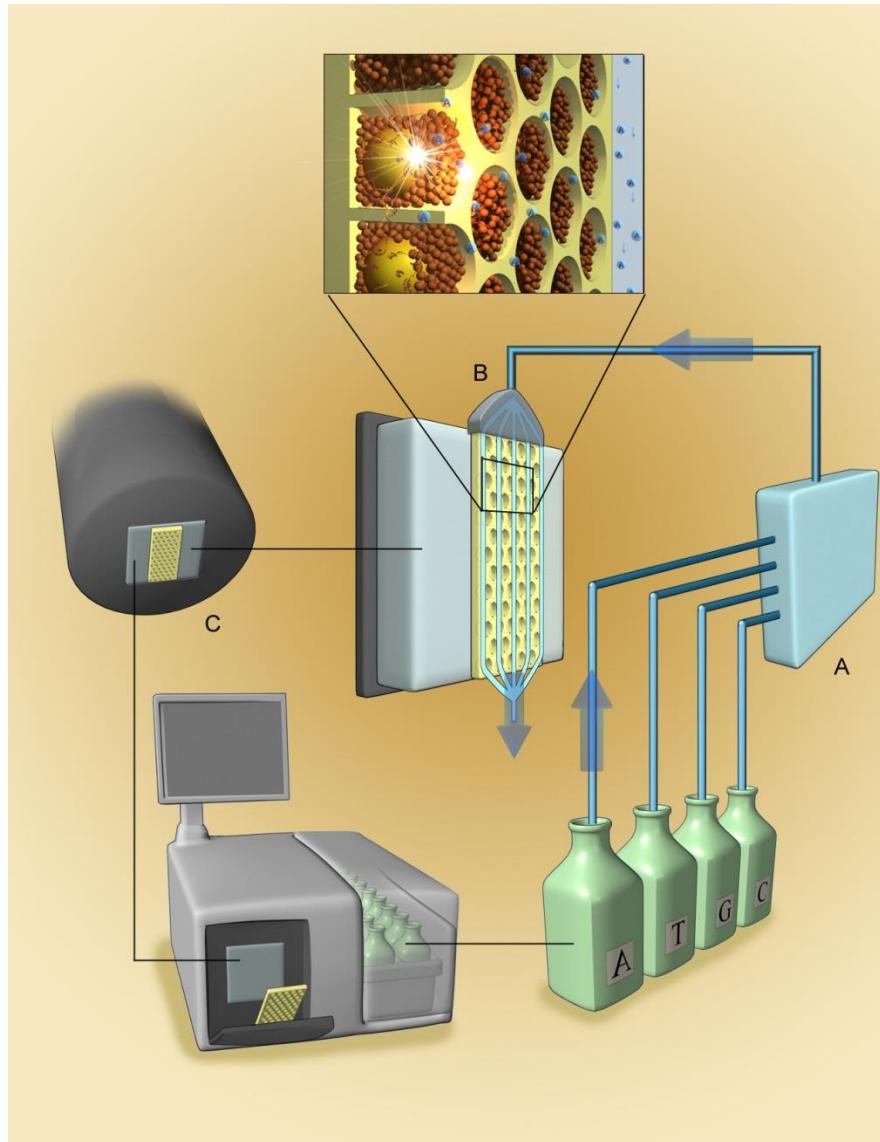
Load Enzyme Beads



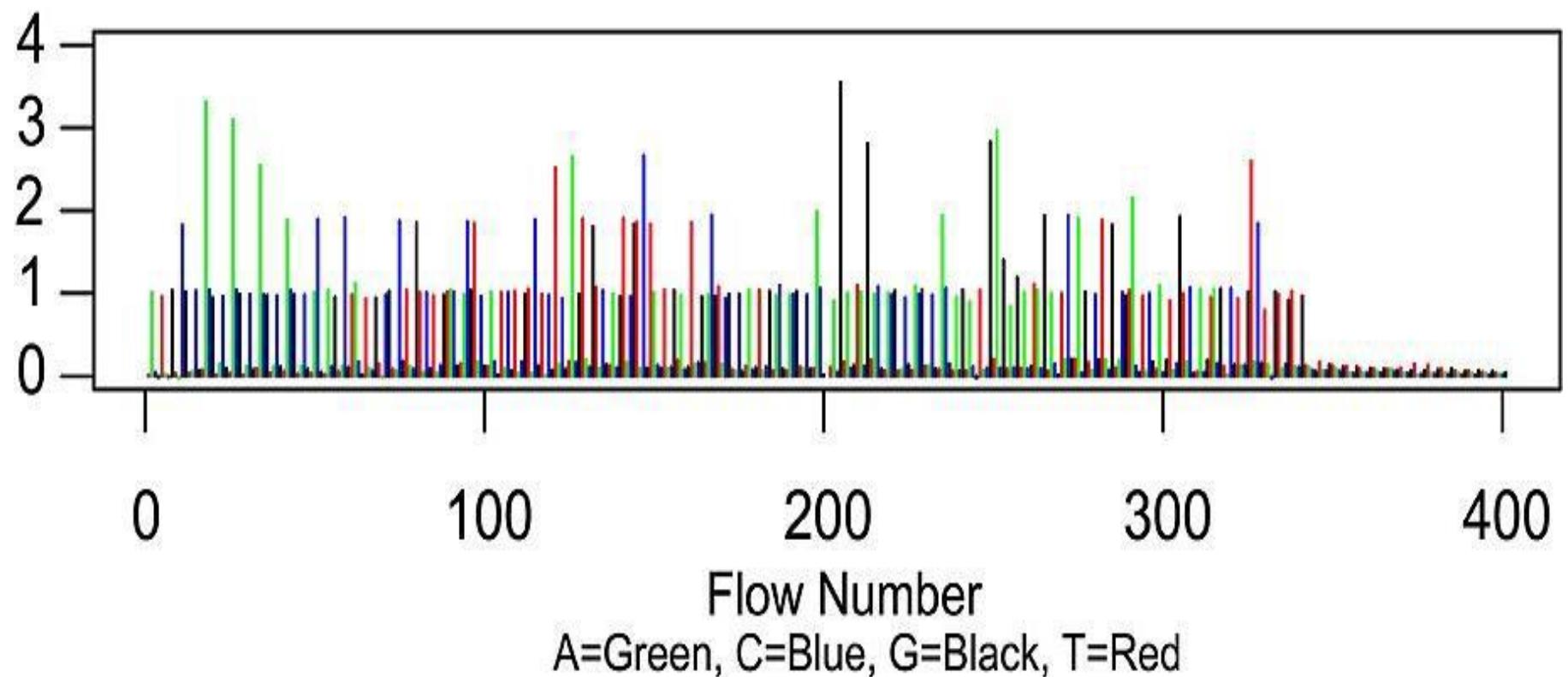
Centrifugation



# 454 Technology - Sequencing Instrument



# 454 Data Example



# 454/Roche Summary

- » Long read lengths – good for amplicons and *de-novo* sequencing
- » High error rate near homopolymers
- » Single end only
- » .7 Gb / run
- » 700-1000 base reads
- » <24 hour run time
- » \$7,000 / Gb



Roche discontinued in 2016 – Too expensive

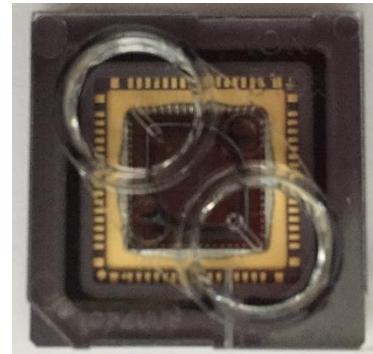
# Other Flow technologies

- » Ion Torrent. As 454 but detects the H<sup>+</sup> released as a base is incorporated.
- » Genapsys. As 454 but detects increase in impedance as a base is incorporated.

# Ion Torrent's PGM



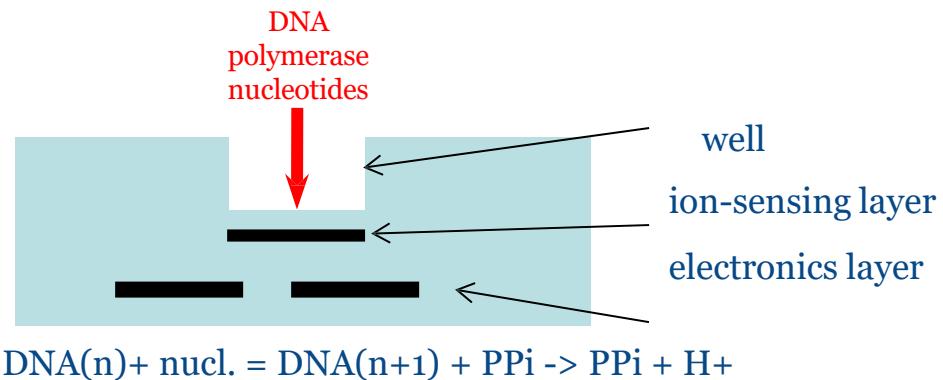
Capital cost \$50,000



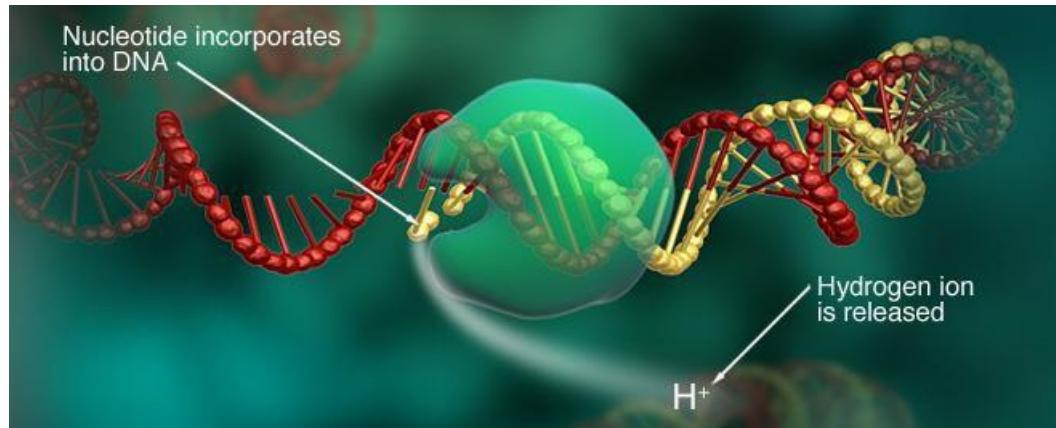
# Ion Torrent

- » Similar to 454 but detects H<sup>+</sup> released as a base is added
- » Prone to errors near homopolymers
- » Not good for whole genome sequencing but useful for targeted sequencing as run times are short
- » Used in a lot of clinical settings for disease panel sequencing

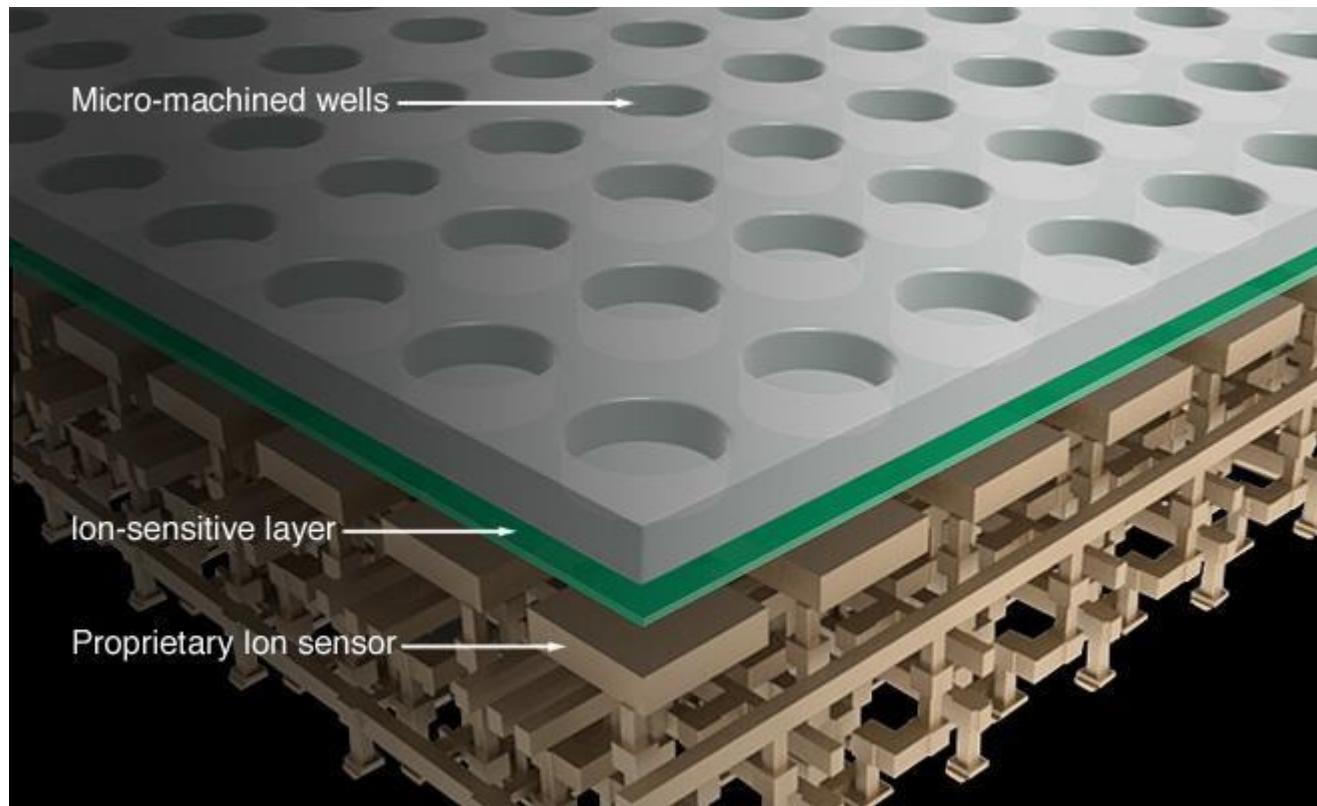
# Ion Torrent's Technology



Flow through A then T then ... like 454



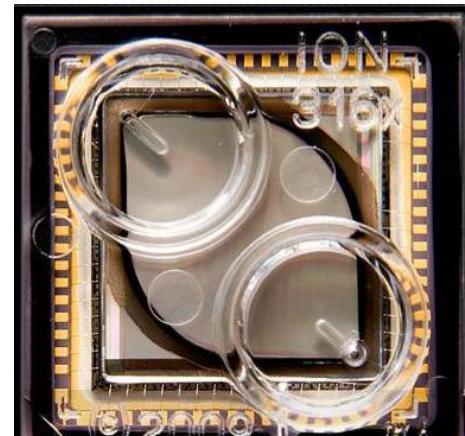
# Ion Torrent Chip Drawing





# Ion Torrent

- » Library prep like original 454
- » Amplification on beads by emPCR
- » CMOS chip detection
- » Cyclic addition sequencing - pH changes
- » Not single-molecule





## Ion AmpliSeq™ technology: As Simple As PCR Your Targets, Your Genome, Your Panel

The most comprehensive gene coverage  
with the lowest amount of DNA or RNA Input

### Simple

- 10 ng of DNA per pool
- FFPE-compatible
- PCR-based target selection



### Scalable

- Up to 24,000 primers per pool
- 1–1000s of genes
- 96 barcodes for multiplexing



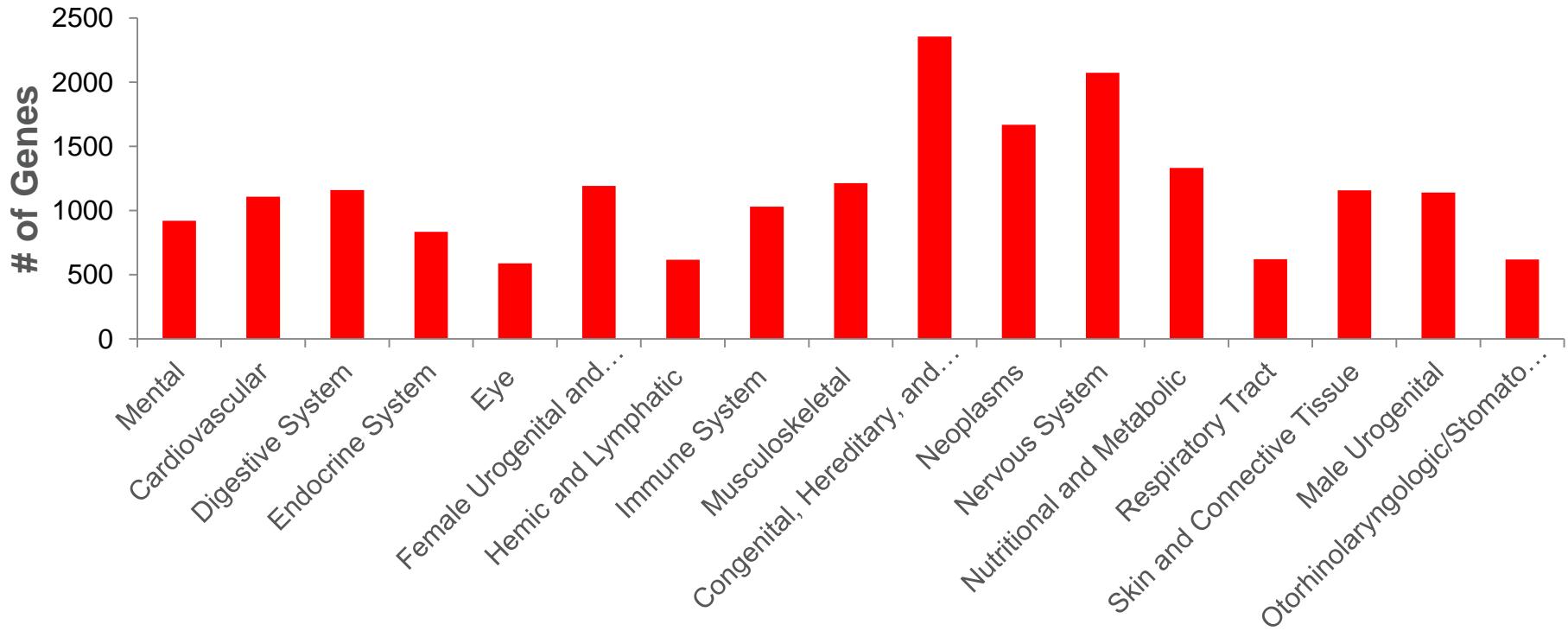
### Fast

- 1 day from DNA to results
- 2 hours to design custom panels
- 3.5 hours for target selection and library preparation



Ion AmpliSeq On-Demand Panels | Now 5,000 Pre-Designed, Pre-Tested Genes Available

## Expanded Gene Content Across Disease Research Areas



Average of 1154 genes per major UMLS disease research area category

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

# Ion GeneStudio S5 Series | Flexible Portfolio Configurable to Your Needs



Ion GeneStudio™ S5



Fast.



Ion 510™  
Chip  
2–3 M reads  
Up to 400 bp



Ion 520™  
Chip  
3–6 M reads  
Up to 600 bp

Ion GeneStudio™ S5 Plus



New

Flexible.

Ion GeneStudio™ S5 Prime



New

Powerful

	New	New
	Ion 530™ Chip 15–20 M reads Up to 600 bp	Ion 550™ Chip 100–130 M reads Up to 200 bp

For Research Use Only. Not for use in diagnostic procedures. \* Throughputs based on 200bp sequencing

# Output and Turn-Around Time to Meet Your Lab's Peak Volume Needs



**Ion GeneStudio™ S5**



**Ion GeneStudio™ S5 Plus**



**Ion GeneStudio™ S5 Prime**



<b>Speed*</b>	19 hrs	10 hrs	6.5 hrs
<b>Output (max/day):</b>	15 Gb/80 M	30 Gb/160 M	50 Gb/260 M
<b>Chips (max/day):</b>	1 x 540	<u>2 x 540</u> or 1 x 550	2 x 550

\* Based off 540 chip – sequencing (2.5 hours) and analysis (varies) time

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

## Genexus System—Tomorrow's Specimen-to-Report NGS Workflow

### Genexus Software

\$299k  
12-15M reads

- FFPE tissue
- Frozen tissue
- Bone marrow
- Whole blood
- PBL
- Urine
- Saliva

#### Nucleic acid purification and quantitation\*

**Ion Torrent™ Genexus™**  
Purification System (Available 2020).



Up to 32 FFPE tissue samples with DNA OR RNA only input

2 hr TAT

#### Library preparation to variant interpretation

**Ion Torrent™ Genexus™**  
Integrated Sequencer (Available Nov 2019)

**Ion Torrent™ GX5™ Chip:**  
12–15M reads/lane



14 hours for a single-lane run (approx. 24 to 30 hours for full chip)  
Up to 32 Samples per run

- \* Specimen-to-report workflow available after Ion Torrent Genexus Purification System launches in 2020.  
The content provided herein may relate to products that have not been officially released and is subject to change without notice.

## Oncomine Precision Assay on Ion Torrent Genexus System

Maximizes your ability to detect relevant variants

### Curated pan-cancer content



- Mutations, CNVs, and fusion variant types across 50 key genes
- Tumor suppressors, drivers, and resistance variants

### Tissue and plasma samples



- One test, one workflow, multiple sample types
- Maximizes the number of tumors that can be profiled

### Molecular tagging



- Enhanced low-level variant detection
- Key for liquid biopsy testing

### FusionSync™ Detection Technology



- Sensitive and specific—targeted isoform designs
- Novel fusion detection

The content provided herein may relate to products that have not been officially released and is subject to change without notice.



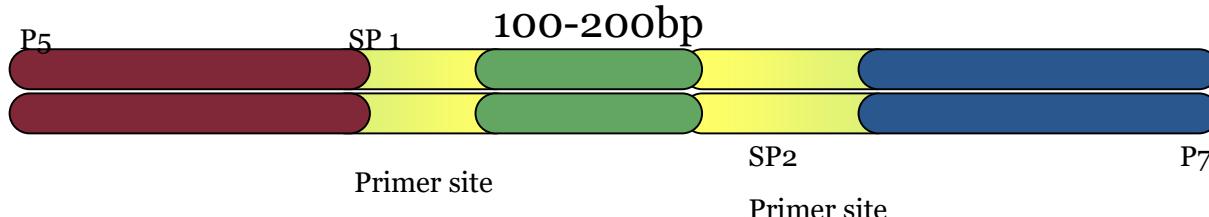
# Sequencing by Synthesis with fluorescence detection



# Illumina

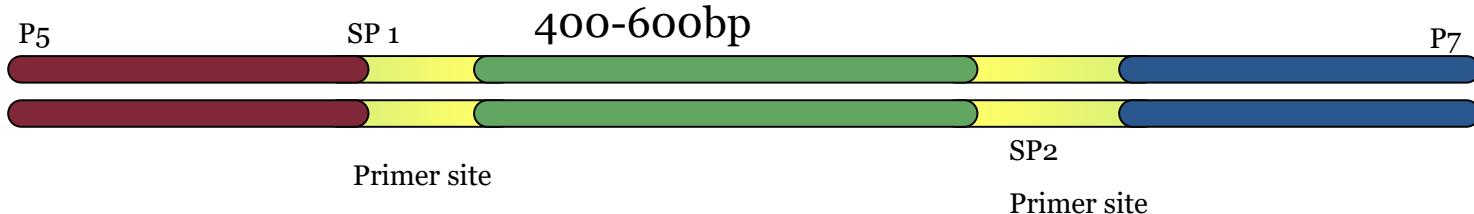
# Illumina Library Insert sizes

*Exome, targeted, ChiP, ATAC*



Reads need to be just long enough to map and ideally not to overlap exon boundaries

*Whole Genome*



Fragments need to be just long to span common repeat elements eg AluI with unique sequence on either side so can map

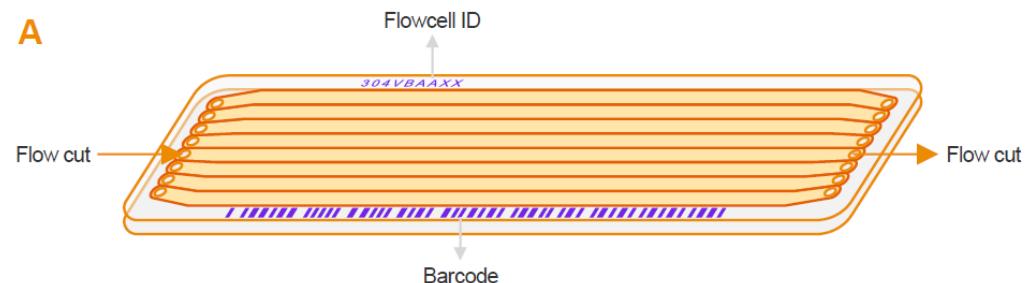


# Recall: We Prepared Our Samples So That Nucleotide Fragments Bind a Flow Cell, a Device Where Sequencing Occurs on an Illumina Platform

DNA is hybridized onto the surface of the flow cell

Polymerases extend DNA using fluorescent nucleotides

Cameras detect fluorescent signals across the flow cell



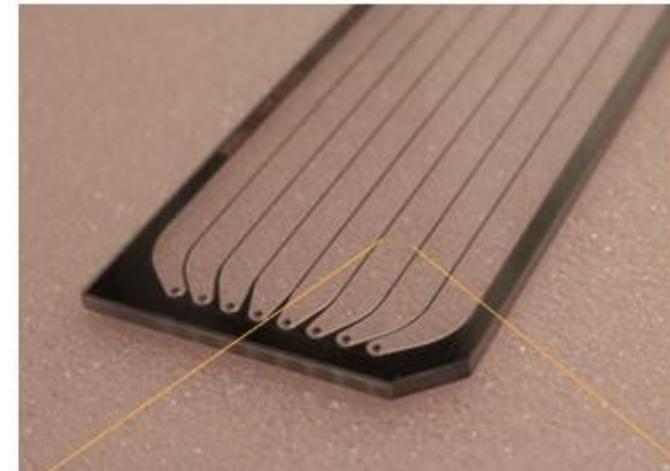
illumina®

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

wellcome  
**sanger**  
institute

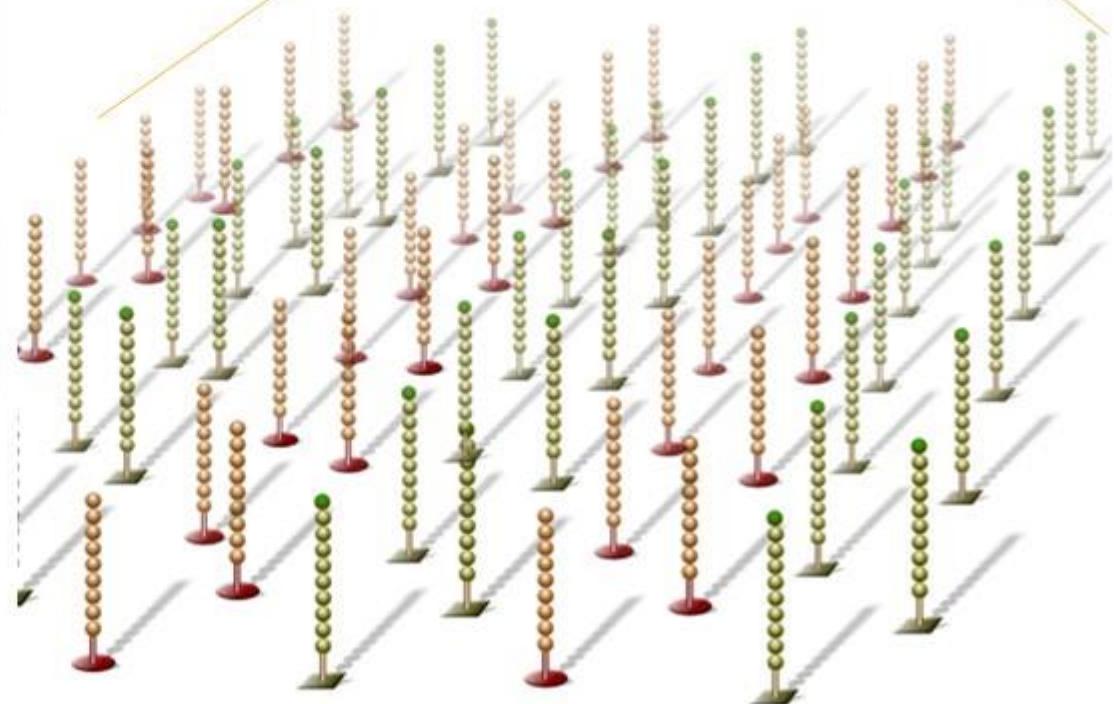
# What is a Flow Cell?

Cluster generation occurs on a flow cell



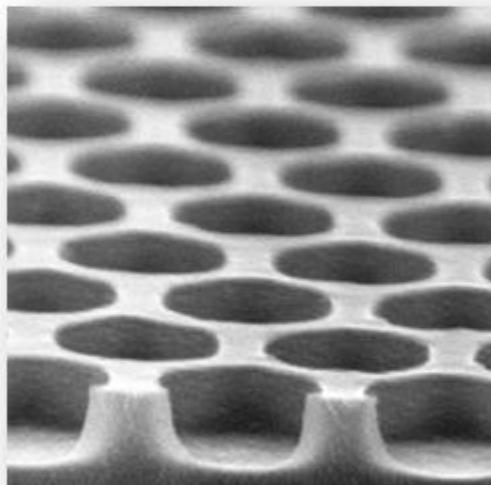
A flow cell is a thick glass slide with channels or lanes

Each lane is randomly coated with a lawn of oligos that are complementary to library adapters



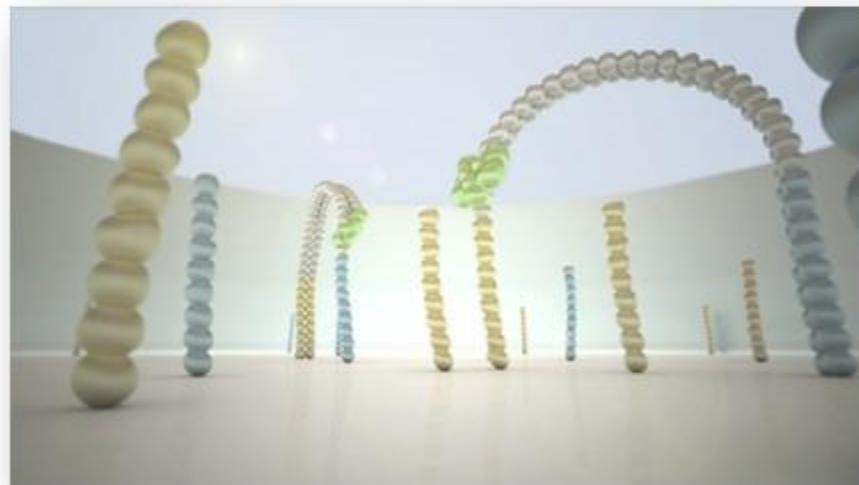
# Patterned Flow Cell Technology

iSeq100, HiSeq4000, Xten, NovaSeq

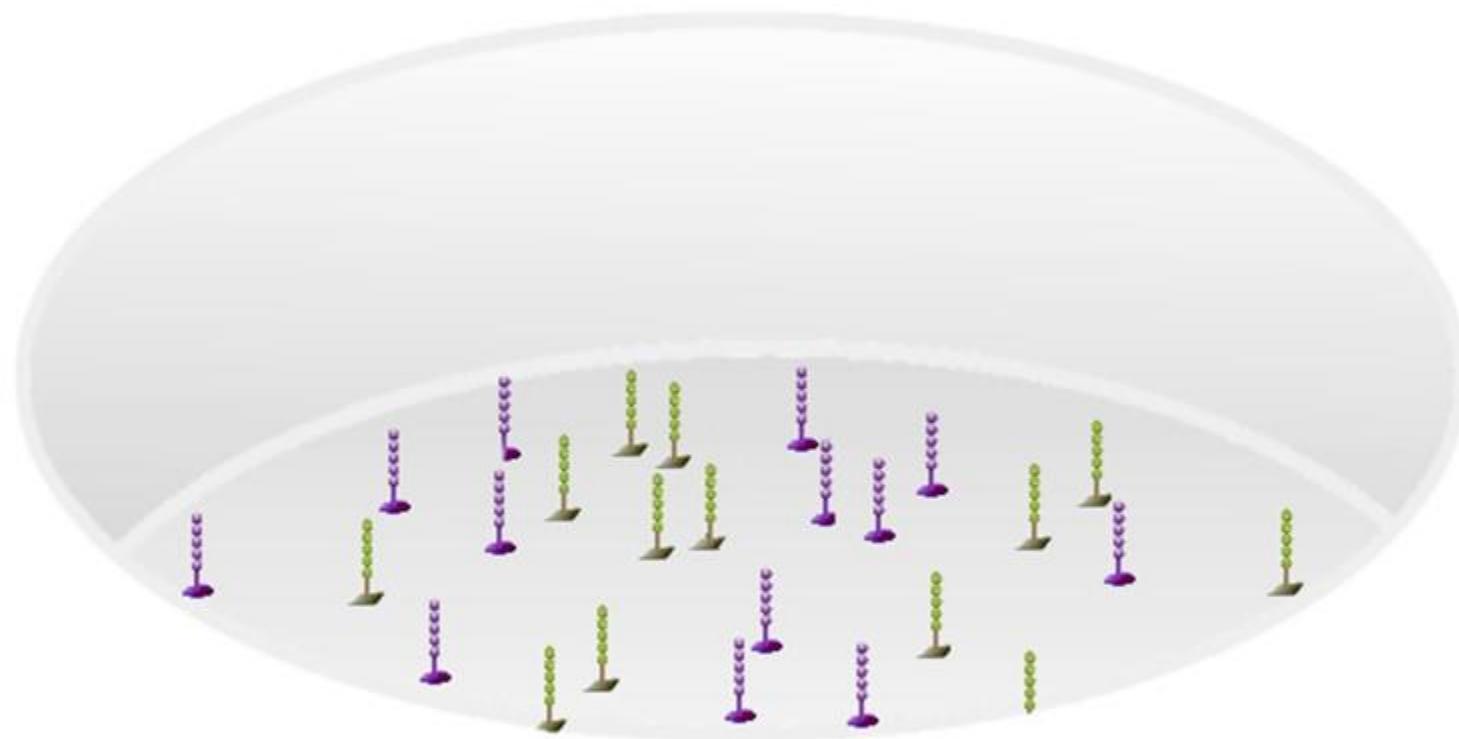


## Billions of ordered wells:

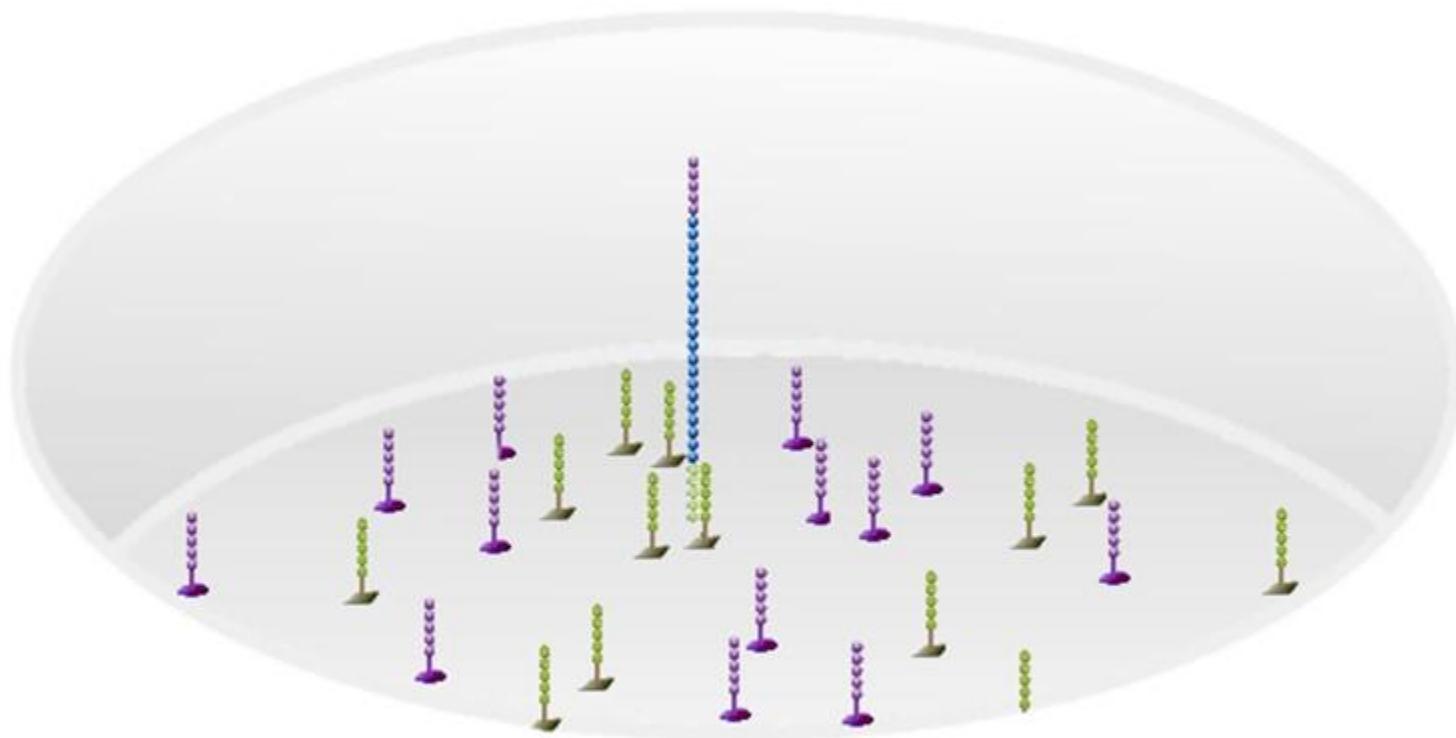
- Clusters contained within ordered wells
- Defined cluster size and spacing
- Increased cluster density
- Simplified imaging
- No pause to complete template generation



**Since 2016 Illumina flowcells have been patterned with clusters formed in a defined array of wells**

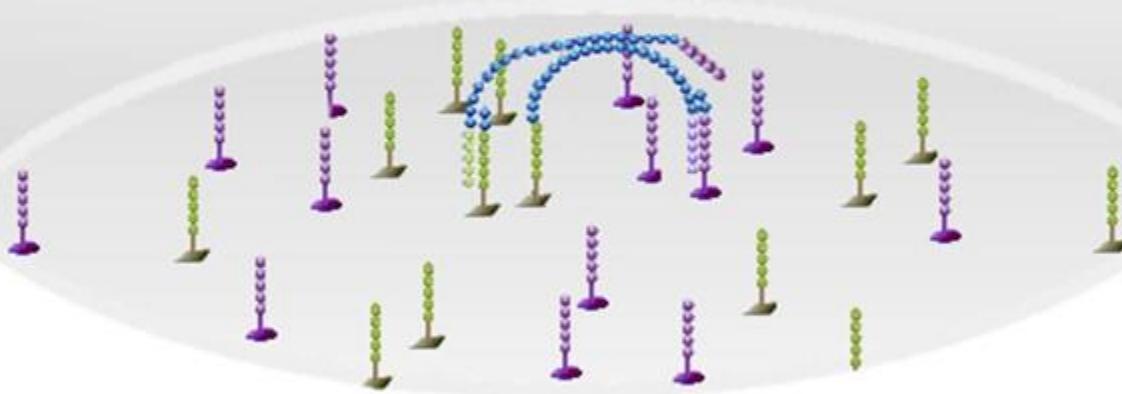


Oligos are not present on surface between wells to control cluster size

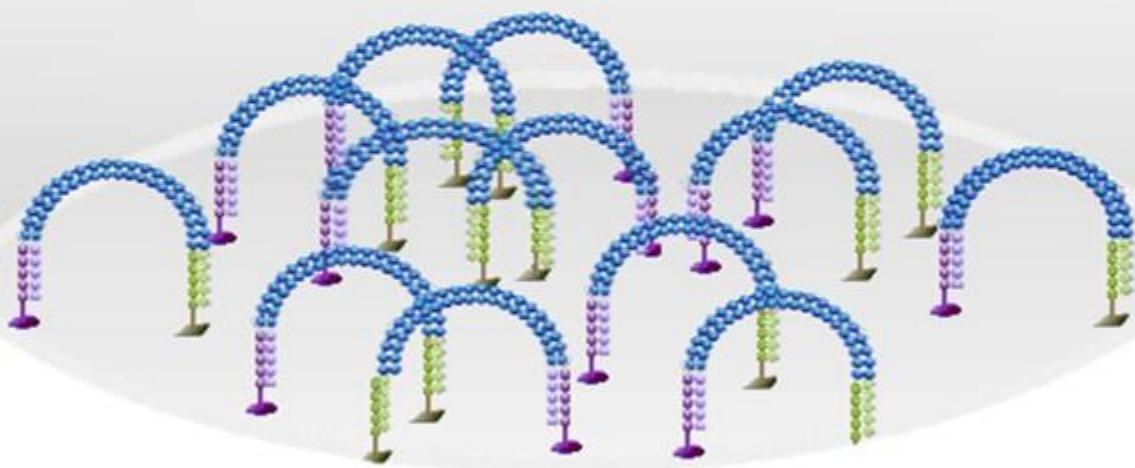


Single stranded DNA is introduced to the flow cell, which hybridizes to an oligo present in the microwell

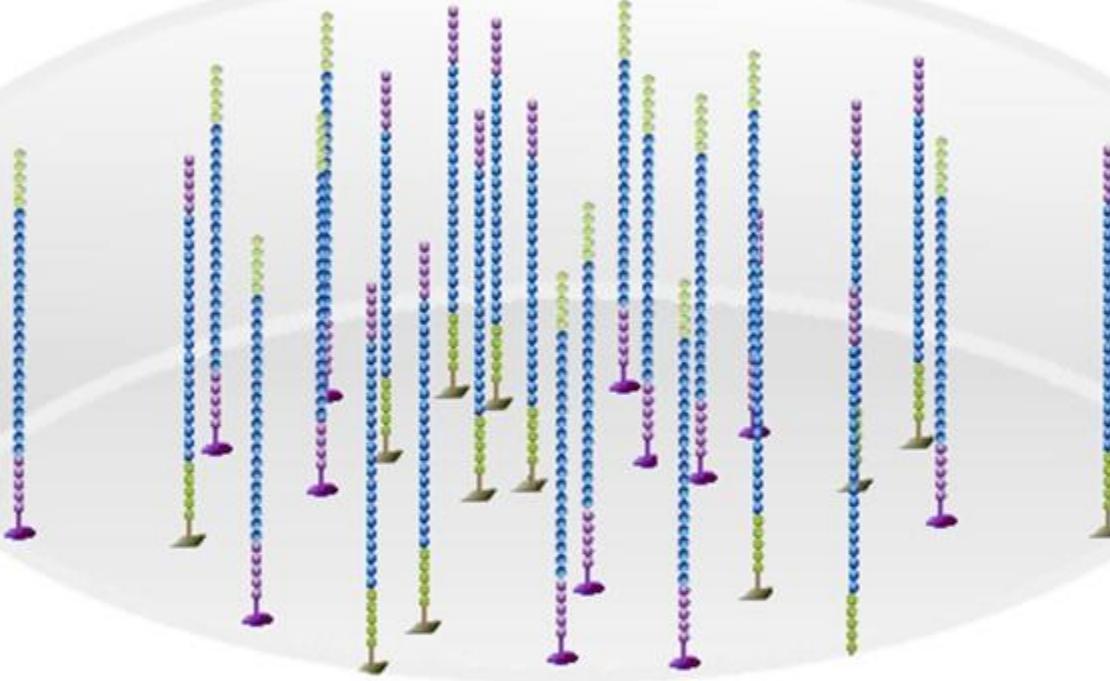
## ExAmp



Amplification occurs on both strands simultaneously

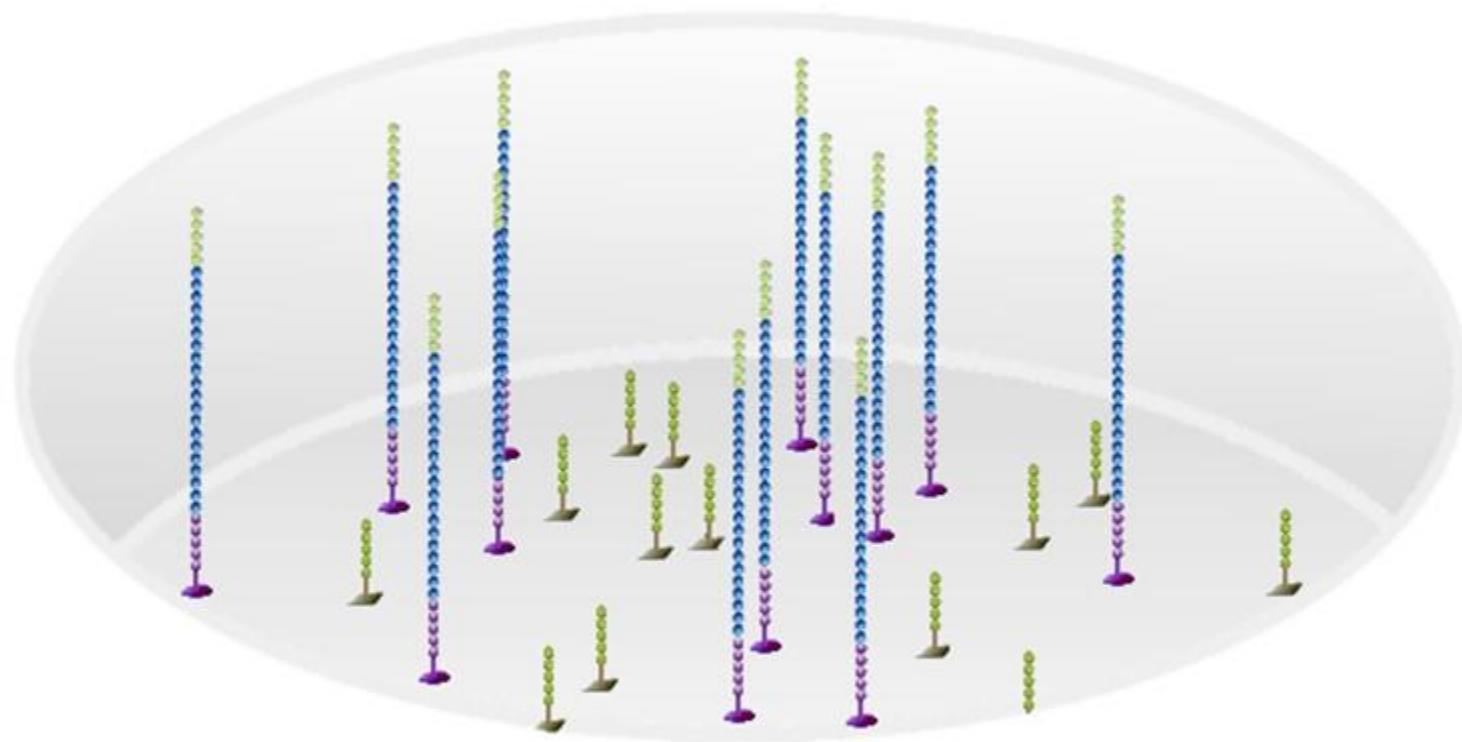


This process continues until no unused oligos are left on the flow cell surface

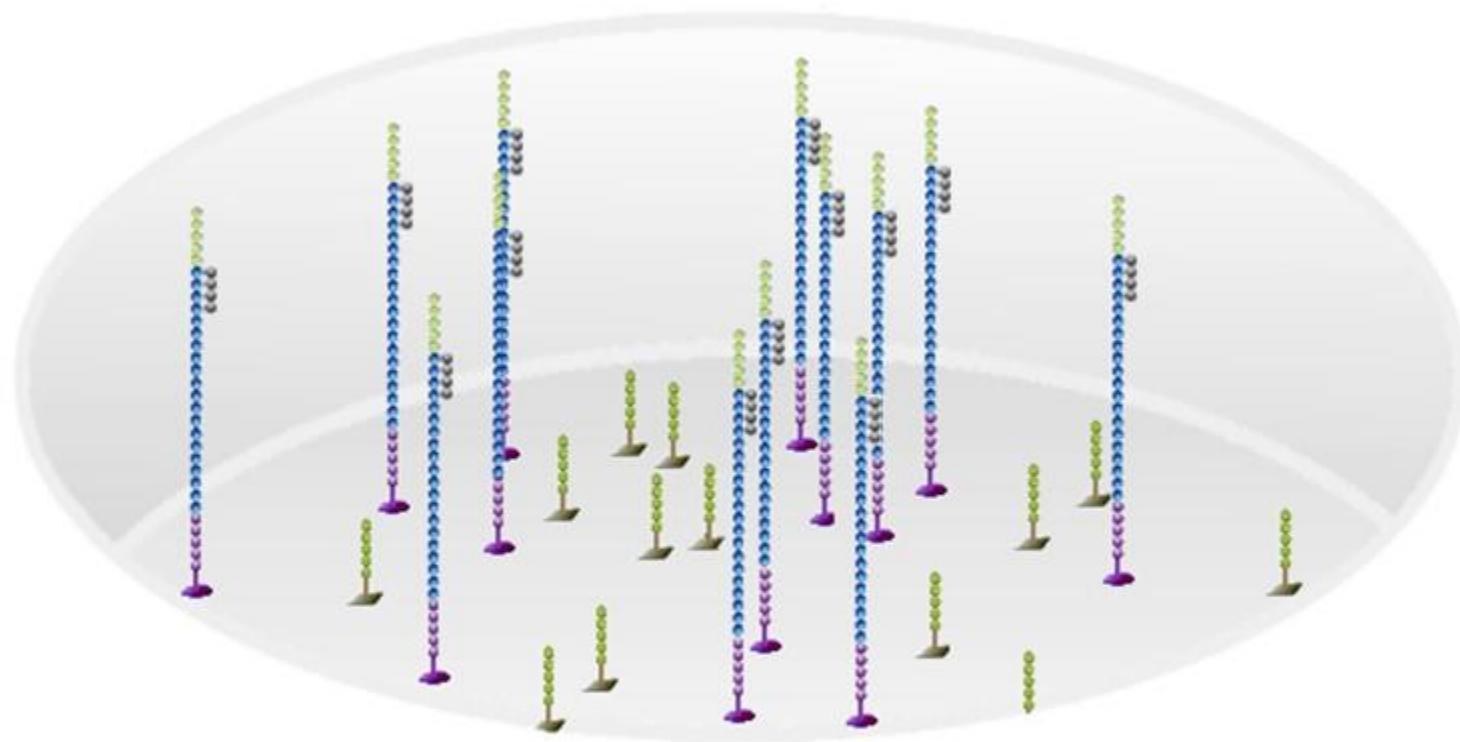


The diagram illustrates a collection of DNA molecules that have been converted into linear strands. These linear strands are represented by vertical lines composed of small, colored circular segments. The segments are colored in four distinct hues: blue, green, yellow, and purple. Each strand begins with a small, dark brown, V-shaped base at its bottom end. The strands are arranged in a somewhat scattered fashion within a large, light gray oval. Some strands are relatively short, while others extend significantly upwards, creating a sense of depth and variation.

To prepare the strands for sequencing,  
linearization is performed

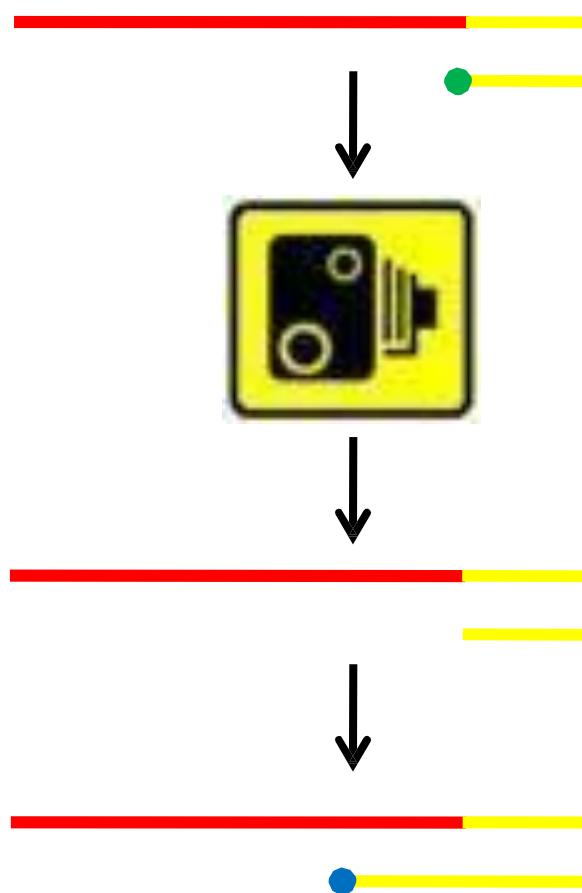


Reverse strands are cleaved



The Read 1 sequencing primer is  
hybridized

# Sequencing by Synthesis

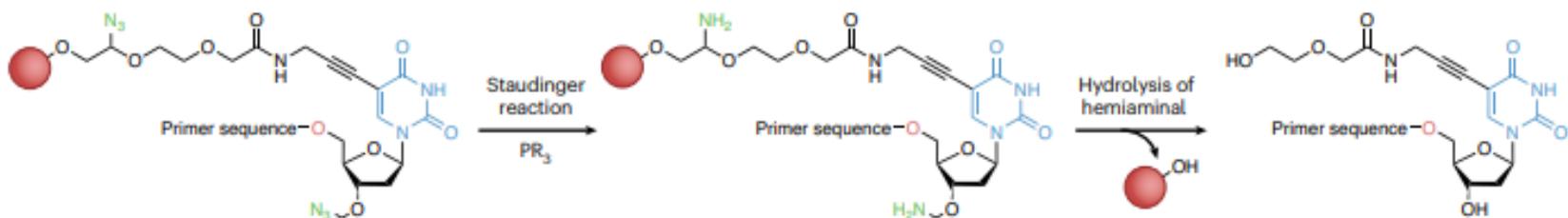
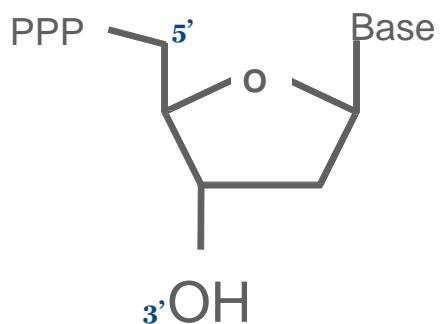


- » Extend by 1 base
- » Image
- » Reverse termination
- » Repeat



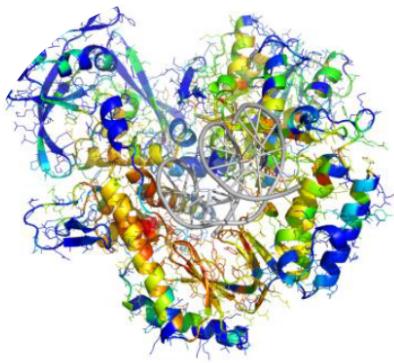
# Illumina Modified Nucleotides

Natural dNTP:



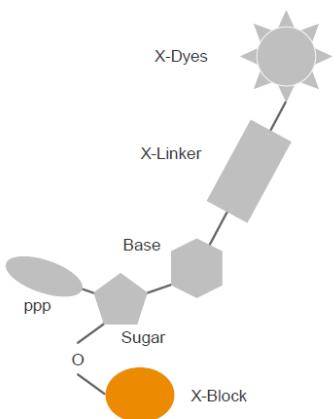
Reversible fluorescent dTTP:

# XLEAP-SBS Chemistry



## Novel Polymerase

Faster incorporation, higher fidelity



## X-Block, X-Linker, X-Dyes

Most resistance to heat, ~50x more stable in solution, ~500x more stable lyophilized, faster block cleave

Enables ambient shipment



## Faster cycle times\*



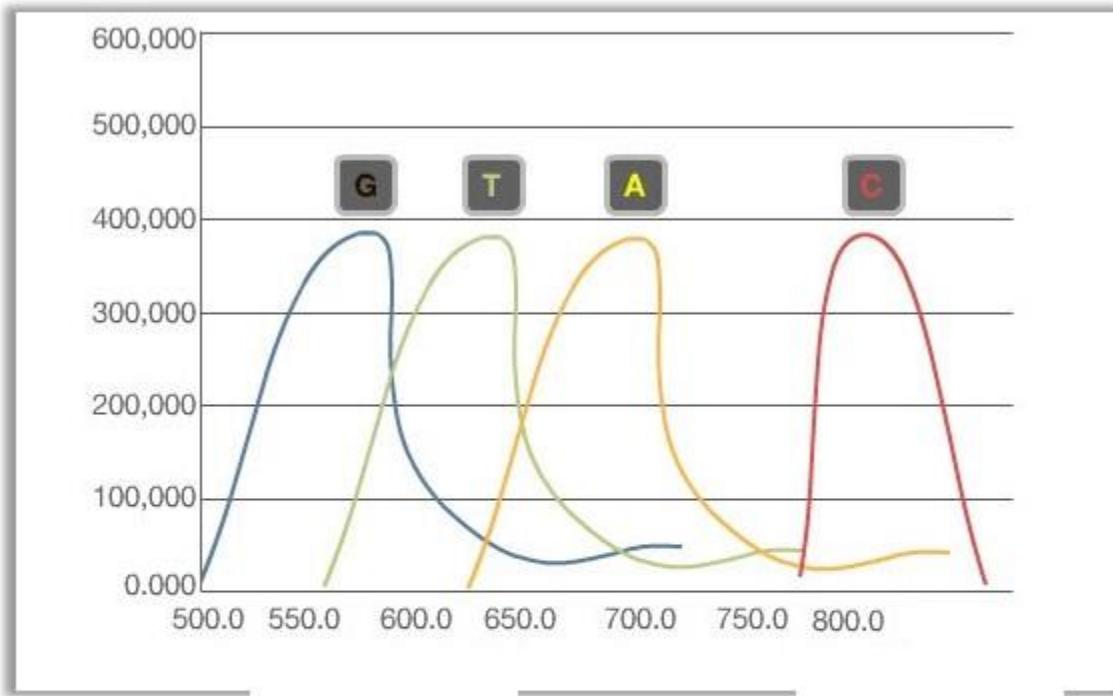
**Greater accuracy\***



More sustainable\*

\* as compared to NovaSeq 6000

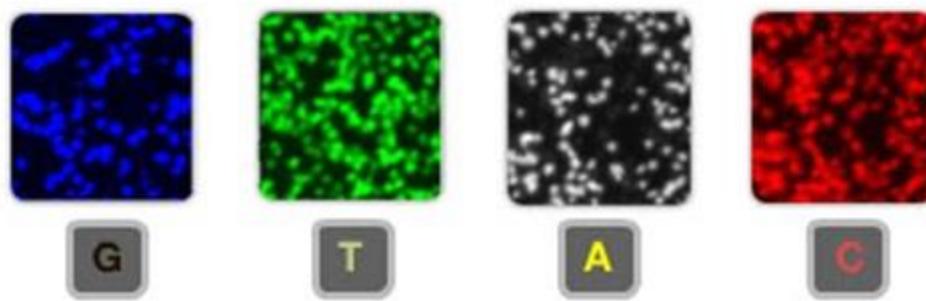
# Four Channel SBS Chemistry: GA, HiSeq, MiSeq



Each of the four DNA bases emit an intensity of a unique wavelength

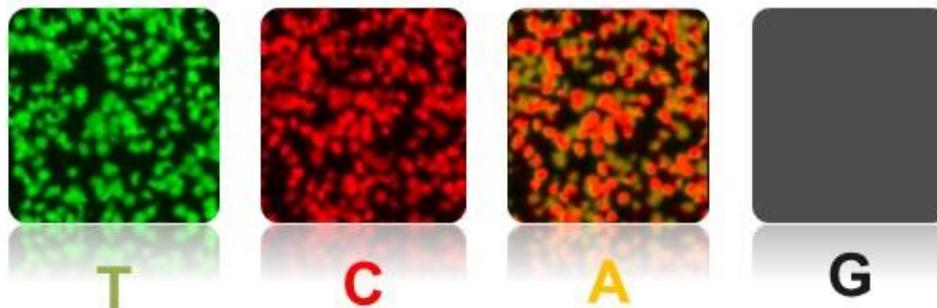
Collects four images:

- During each cycle, each cluster appears in only one of four images



# 2-Channel Sequencing Imaging Cycles

- 2 Channel Sequencing only requires 2 images and hence all the data from the 4 DNA bases are encoded in these 2 images.
- Uses the same sequencing by synthesis (SBS) method as 4 channel sequencing but allows more efficient acquisition of the data.
- Used by NovaSeq, NextSeq and MiniSeq



Channel	Green	Red	Green and Red	Dark - Neither

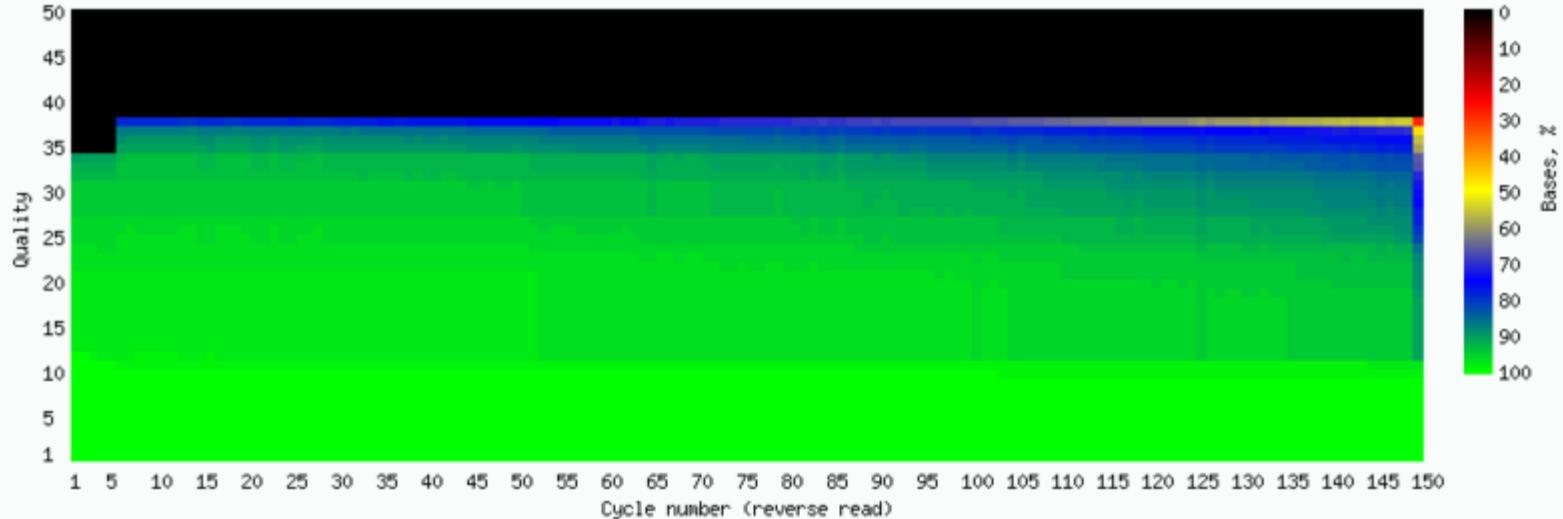
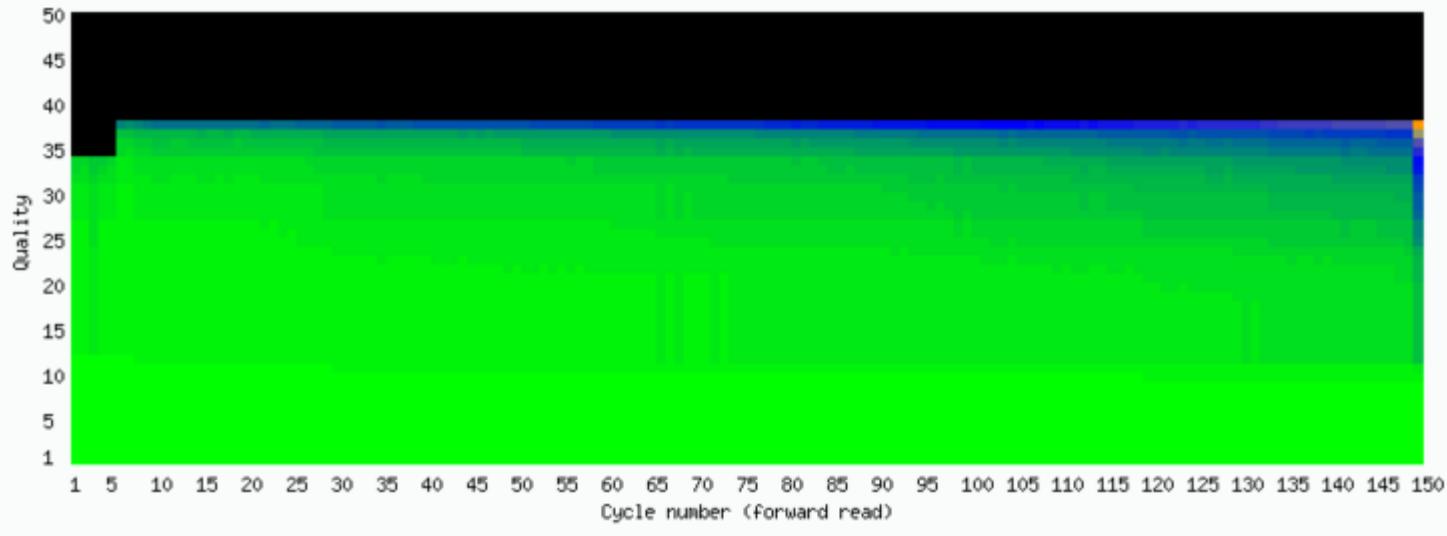
For Research Use Only. Not for use in diagnostic procedures.



# Error

- » Sequence quality Q is reported on a log scale
- » Q<sub>10</sub> is 1 error in 10
- » Q<sub>20</sub> is 1 error in 100
- » Q<sub>30</sub> is 1 error in 1000
- » Q<sub>40</sub> is 1 error in 10000
- » Q<sub>50</sub> is 1 error in 100000

# Error limits read length



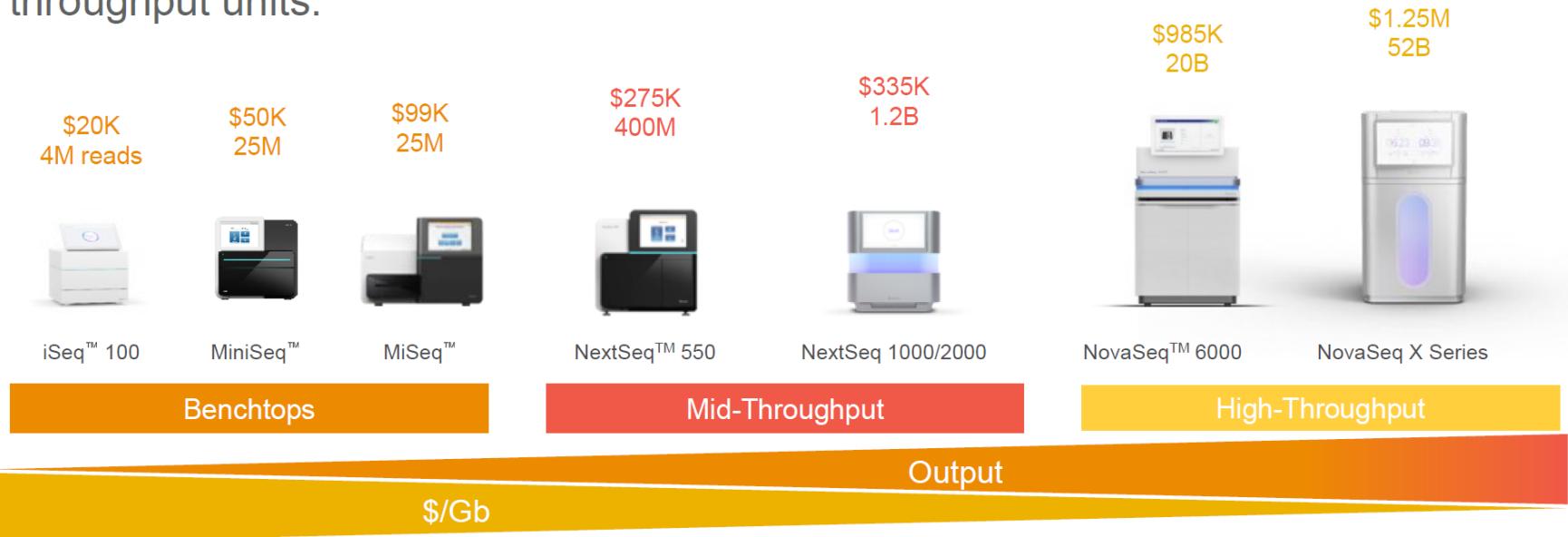


# Illumina sequencing with indexing

With Illumina sequencing you sequence the insert from each end. And you can also perform sequencing reactions that read the sequence of the index in the introduced adapter. So how is this done

# Illumina's Sequencing Portfolio

Sequencers vary a lot in size! From small benchtop platforms to large high throughput units.

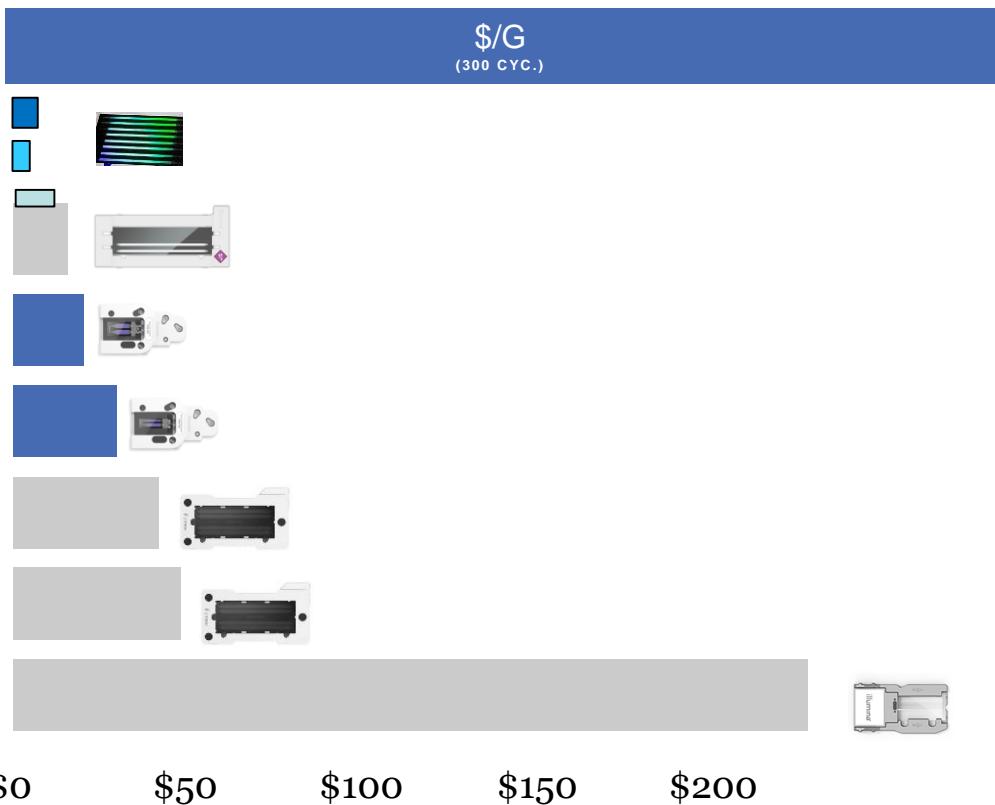


► The sequencer you use may depend a lot on your experiment!

► 300,000 and growing peer-reviewed publications

# Economics of Illumina's Sequencing Portfolio

Platform	Flow Cell	\$/G (300 CYC.)
<b>Novaseq X/X+</b>	10B 25B	\$3 \$2
<b>NovaSeq 6000</b>	S4 SP	\$5 \$15.40
<b>NextSeq 2000</b>	P3	\$20.00
<b>NextSeq 1000/2000</b>	P2	\$29.50
<b>NextSeq 550</b>	HO	\$41.38
<b>NextSeq 550</b>	MO	\$47.50
<b>MiSeq</b>	v2	\$236.00





## Scale up your way with NovaSeq™ X Series 1.5B, 10B & 25B reagent kits

Now Shipping



### 1.5B | 1.6B Clusters

100, 200, 300 cycles

**165-500 Gb**

$\leq 23$  hrs

Fast turnaround times for  
small batch sizes



### 10B | 10B Clusters

100, 200, 300 cycles

**1-3 Tb**

$\leq 25$  hrs

Flexible multi-project run,  
maximum efficiency



### 25B | 26B Clusters

300 cycles

**8Tb**

$\leq 48$  hrs

Large projects, deeper  
sequencing, lowest cost  
per sample



# MiSeq i100



Height  
65 cm  
(25.6 in)

Depth  
44.8 cm (17.6 in)

Width  
40.2 cm (15.8 in)

## MiSeq i100 Series specifications<sup>a</sup>

Output range<sup>b</sup> 1.5–30 Gb

Paired-end reads per run 10–200M

Max read length 2 x 300 bp

Run time ~4–15.5 hr

a. Specifications based on Illumina PhiX control library at supported cluster densities.

b. Maximum range based on 100M flow cell specifications. The 100M flow cell will be available starting in 2025 for the MiSeq i100 Plus System only.



# Illumina

- » Cheap \$2-\$300/Gb
- » Highly accurate data mostly Q30
- » Massively parallel. Millions/billions of reads
- » Short read



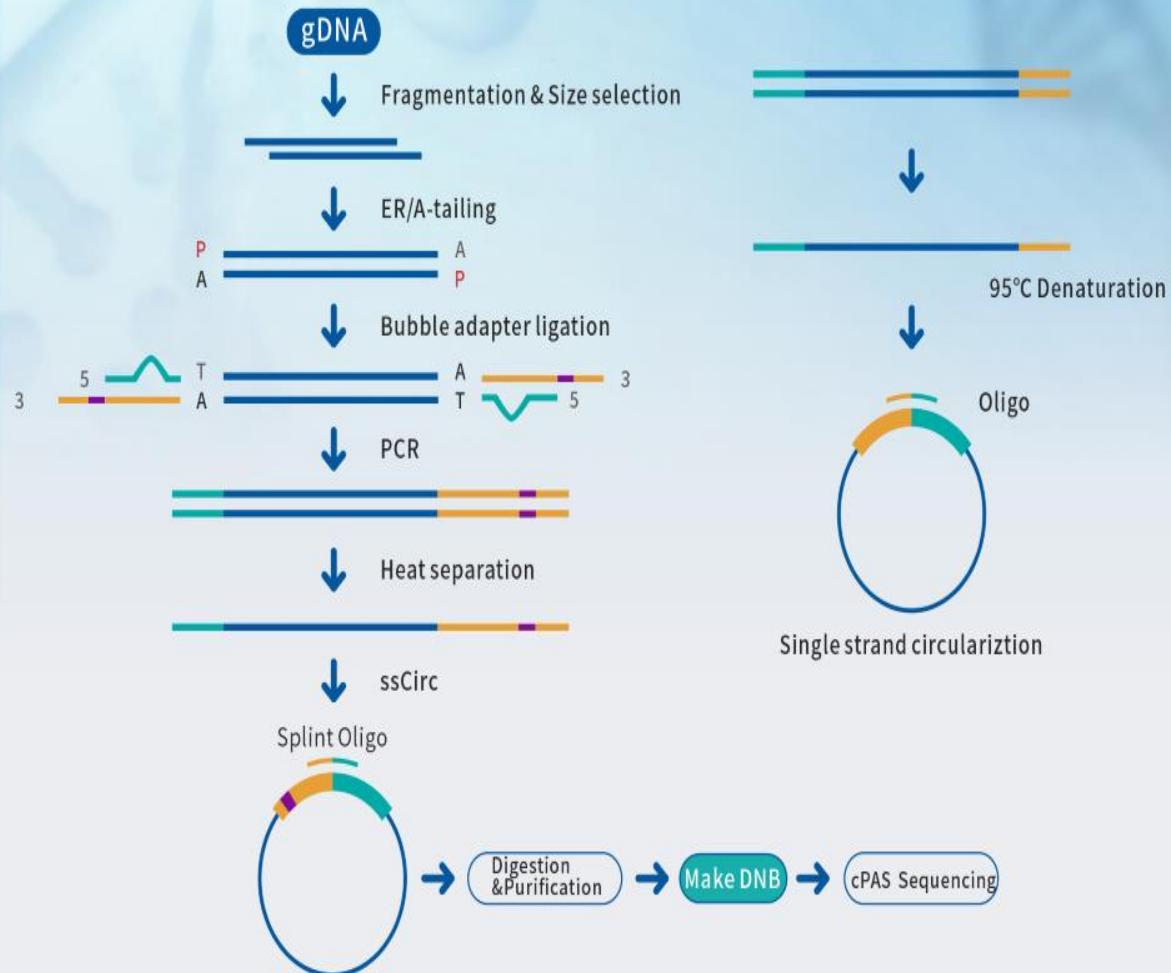
LEADING LIFE SCIENCE INNOVATION

The ownership of this material belongs to MGI, and without the permission of the owner, it shall not be directly released or disclosed.

MGI All Rights Reserved Dissemination, distribution and copying is prohibited without authorization.

# DNBSEQ™ Technologies: Library Preparation

MGI

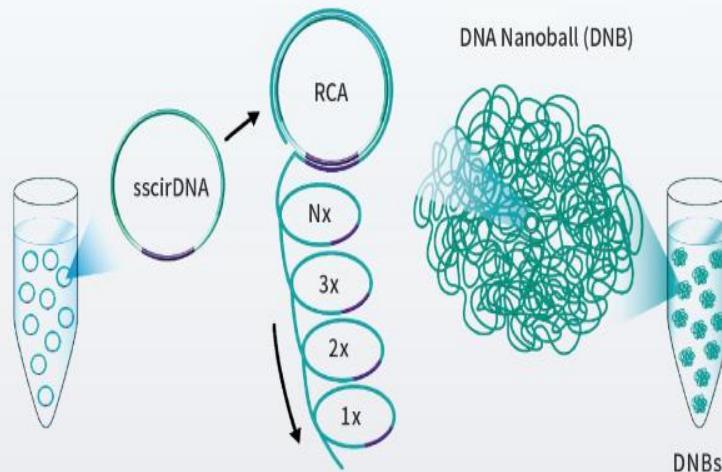


\* Take the preparation of DNA library as an example

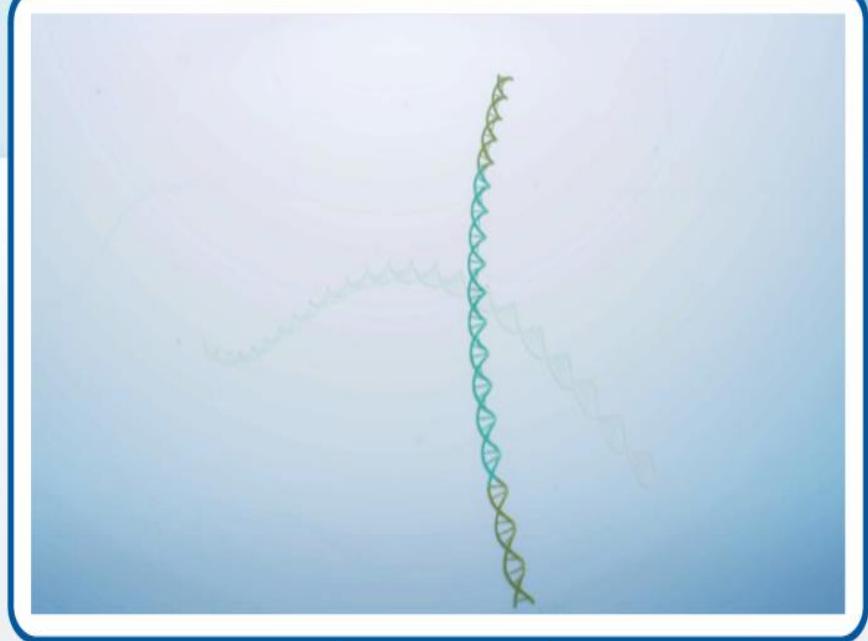
# DNBSEQ™ Technologies: Make DNB

## RCA: Rolling Circle Amplification

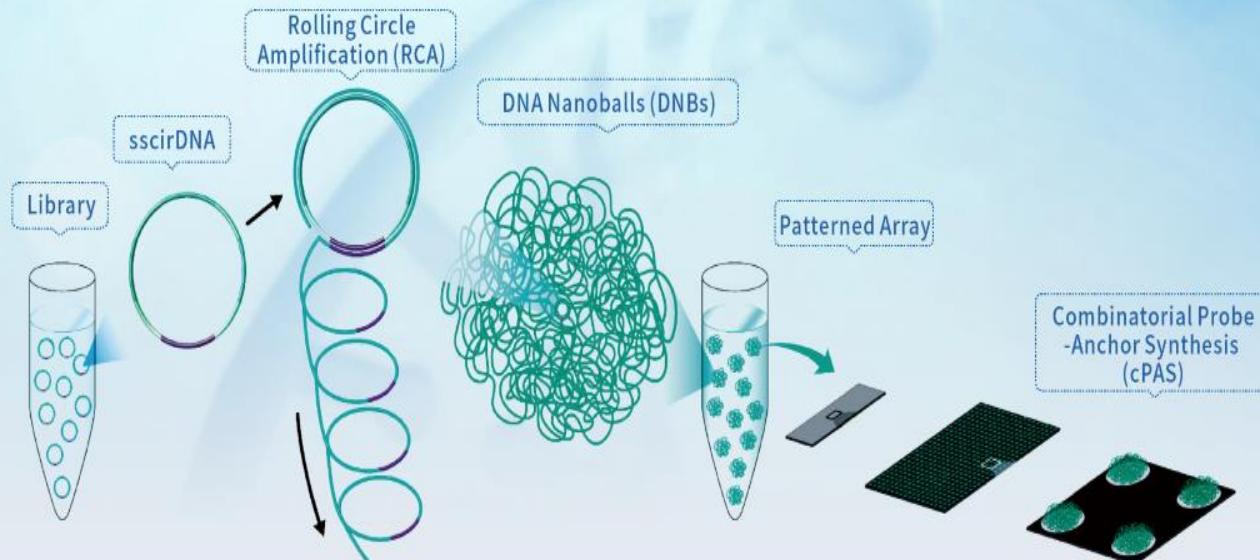
- From sscir DNA to DNBs
- From 1 copy to 300-500 copies



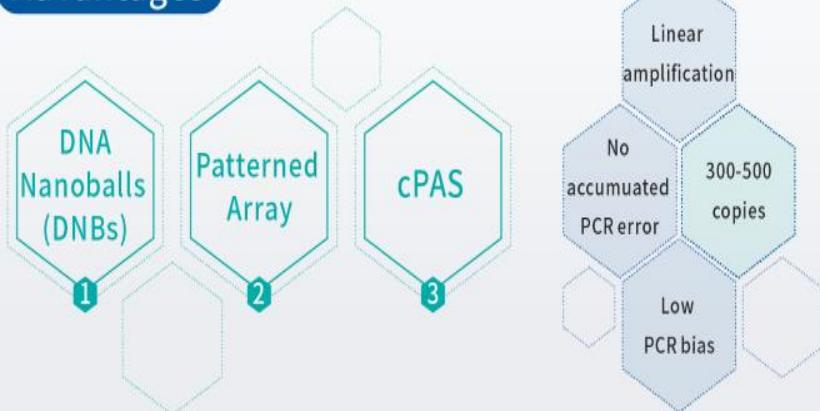
## Make DNB



# DNBSEQ™ Technologies



## Advantages



### • MPS

Massively parallel sequencing

#### -StandardMPS

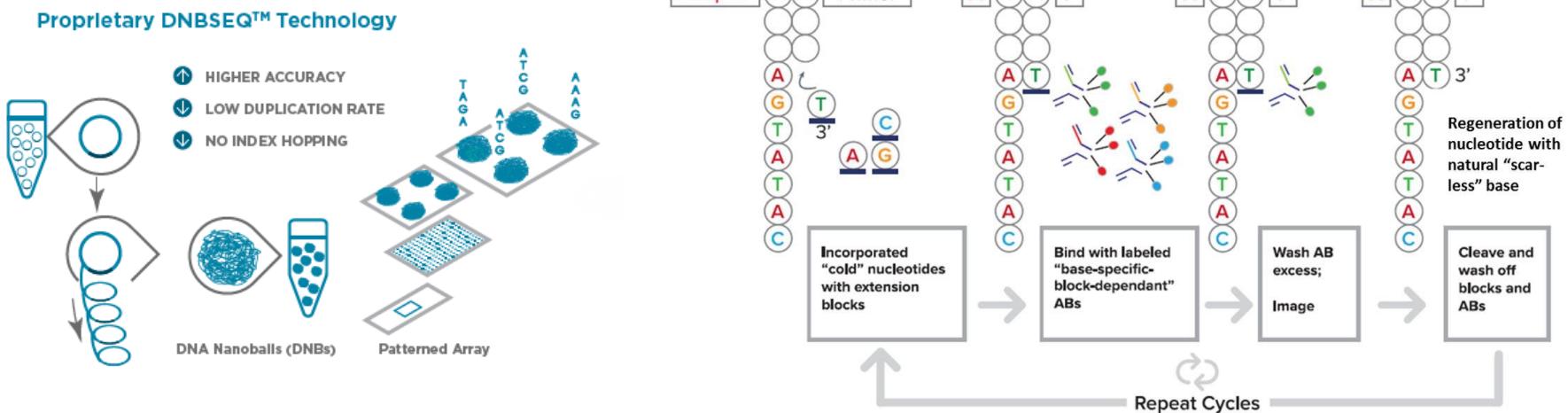
Standard biochemistry with fluorescent groups labelled to dNTPs:  
e.g. DNBSEQ-G400RS High-throughput Sequencing Set (FCL PE150)

#### -CoolMPS

A novel antibody-based sequencing chemistry with fluorescent groups labelled to the antibodies:  
e.g. CoolMPS High-throughput Sequencing Set (DNBSEQ-G400RS FCL PE100)

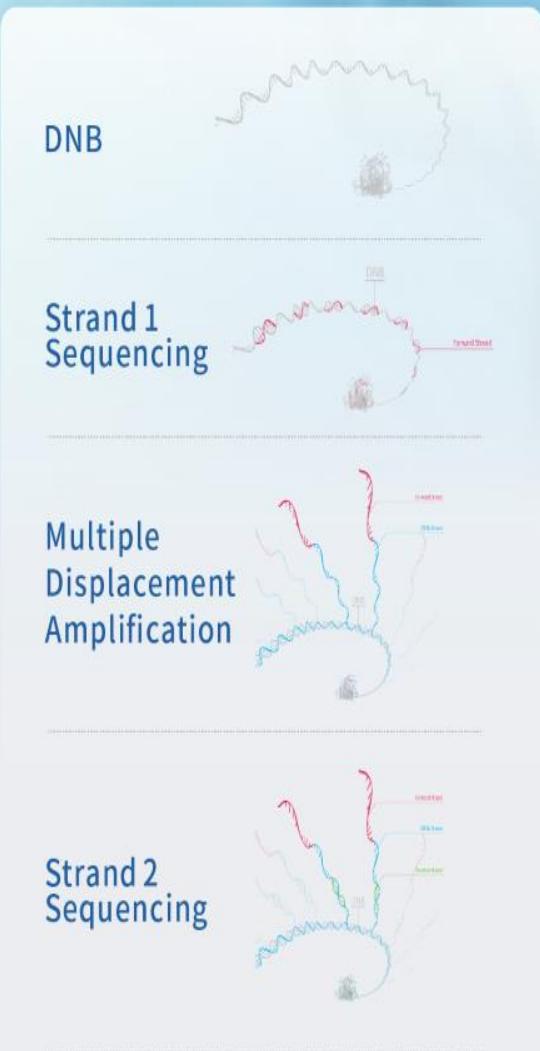
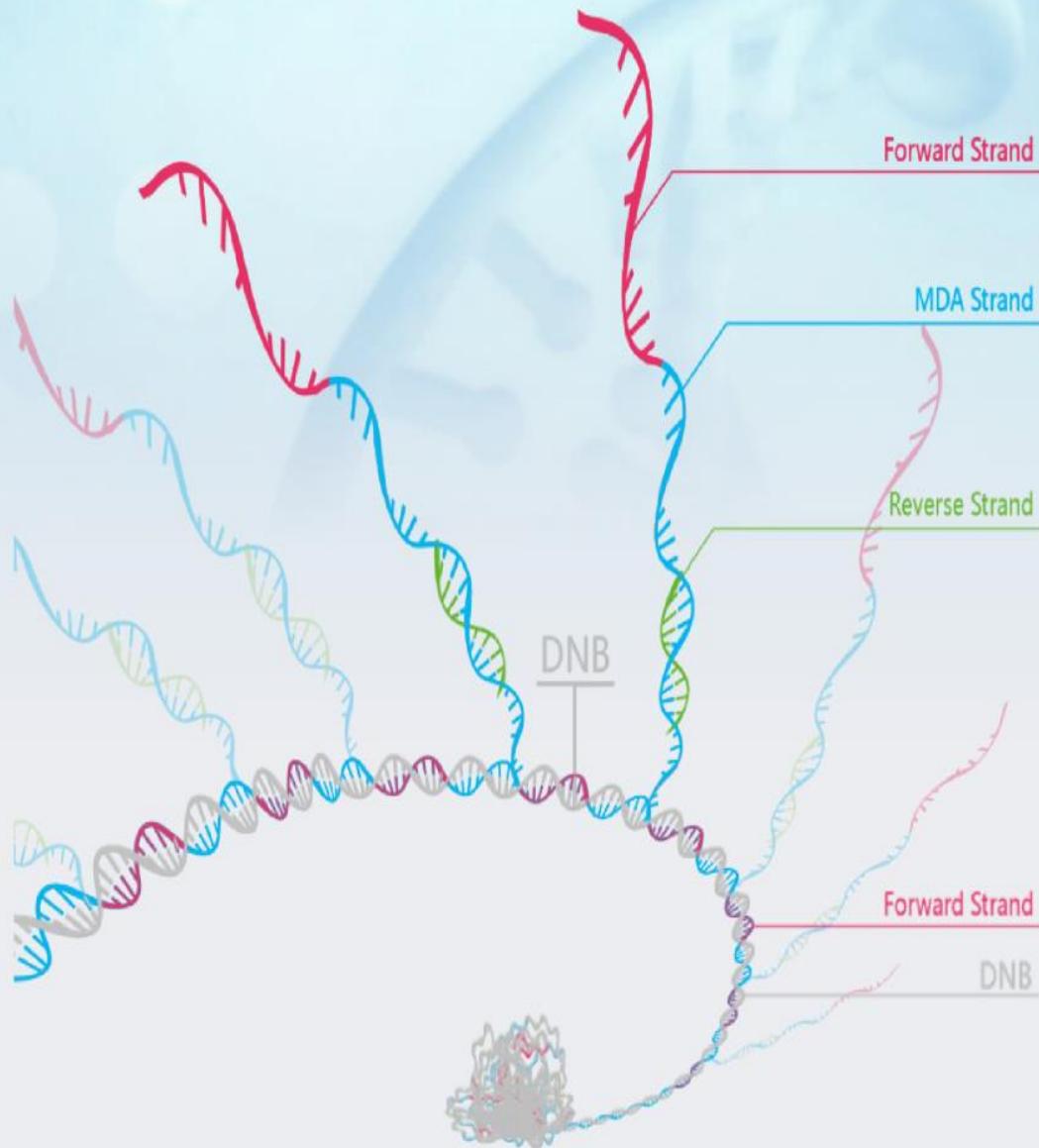
# MGI sequencing chemistry

## DNA nanoballs and cool MPS



# DNBSEQ™ Technologies: Pair End (PE) Sequencing

MGI



# Genetic Sequencer Parameters



	DNBSEQ-T20x2	DNBSEQ-T7	DNBSEQ-G400	DNBSEQ-G50	DNBSEQ-G99	DNBSEQ-E25	
Product Features	Ultra-high Throughput Platform for Population Genomics	Ultra-High Throughput Ultra-High Speed Ultra-Low cost	Comprehensive High Quality	Compact Flexible	Rapid Flexible	Portable Stackable	
Flow Cells/Run	6	4	2	1	2	1	
Flow Cell Type	Slide	FC	FCS	FCL	FCS	FCL	
Lane / Flow Cell	1	1	2	4	1	1	
Effective Reads / Flow Cell	40B	5800M	550M	1500-1800M	100M	500M	
Max Read Length	PE150*	PE150	PE300	SE400 PE200	PE150	PE300	
Min Read Length	/	SE50	SE100	SE50	SE100	SE100	
Data Output/Run	72T	7T	330G	1440G	30G	150G	
Q30 (Max/Min Read)	≥85%	PE150>85% SE50>85%	PE300>80% PE150>85% SE100>85%	PE150>85% PE100>85% SE50>90%	PE150>80% SE100>80%	SE100, PE50>90% PE150>85% PE300>85%	PE150>80% SE100>90%
Run Time	80h	5-24h	13-37h	14-107h	10-28h	7-40h	5-30h
Applications	WGS, cWGS, stLFR WGS, WGBS Single-cell Sequencing, Stereo-seq	WGS, WES, metagenomics sequencing, oncology panel sequencing	Transcriptome sequencing, WGS, WES, Molecular breeding, pathogen rapid detection, liquid biopsy, precision medicine, genetic testing early screening of tumor, individual identification	liquid biopsy, precision medicine, genetic testing, early screening of tumor, individual identification, SARS-CoV-2 identification	targeted oncology panel sequencing, small whole-genome sequencing, low-depth whole genome sequencing	Pathogen rapid detection Science education	

# DNBseq-T1+



12Tb in 22 hours  
Approx \$2/Gb  
Upto PE300



# New Sequencers



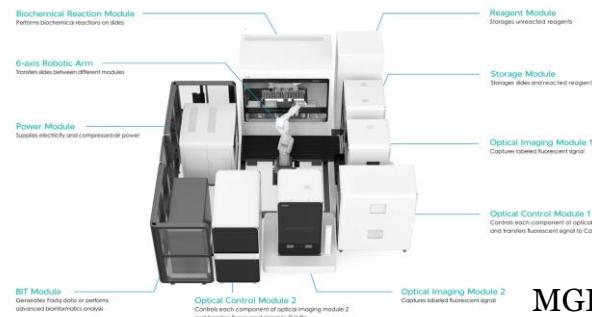
Onso



Element



Ultima UG100



MGI



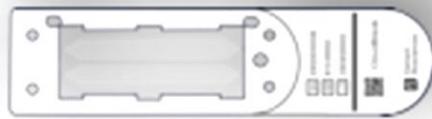
Singular S4

# Comparison Table

	Illumina Novaseq	Illumina Nextseq	Ultima UG100	Element Aviti	Singular G4	PacBio SBB
Instrument	\$1.25M	\$335k	\$1.5M	\$289k (\$249K)	\$350k	\$99K
\$/Gb	3 (10B), 2 (25B)	20 (9 in 2024)	1	2-5	8-10	4-12
Accuracy	QQQ	QQQ	QQQ	QQQQ	QQQ	QQQQQ
Max read length	2x150 (SP)	2x300	300 SE	2x300	2x150	200SE
Run Time for 2x150 or 1x300	24 hrs	48 hrs	18 hrs	48 hrs	19 hrs	48hrs
Flowcells	2	2	2	2	4	2
Yield/run	6000Gb, 16000Gb	30-360Gb	>4000Gb	480Gb	360Gb	150Gb
Other	Infinity long reads	Infinity long reads		Non patterned, loop long reads		Non patterned
Use case			Cheap sequence for big data applications,	-high plex levels -low complexity -long reads	Flexibility via independent flowcells	High accuracy applications, high plex potential

# Element AVITI™

- ~\$335K. \$249K each if you order 3
- \$2-\$5/Gb. \$1680 for 200-300Gb
- 2 flowcells. 2 x 150. 90% >Q40
- 2 x 300 available
- Sequencing by binding on RCA amplified template
- Loop synthetic long read
- Low duplicates
- No index hopping allowing for combinatorial indexing
- Better results with low complexity sequencing
- Can have long inserts >2kb

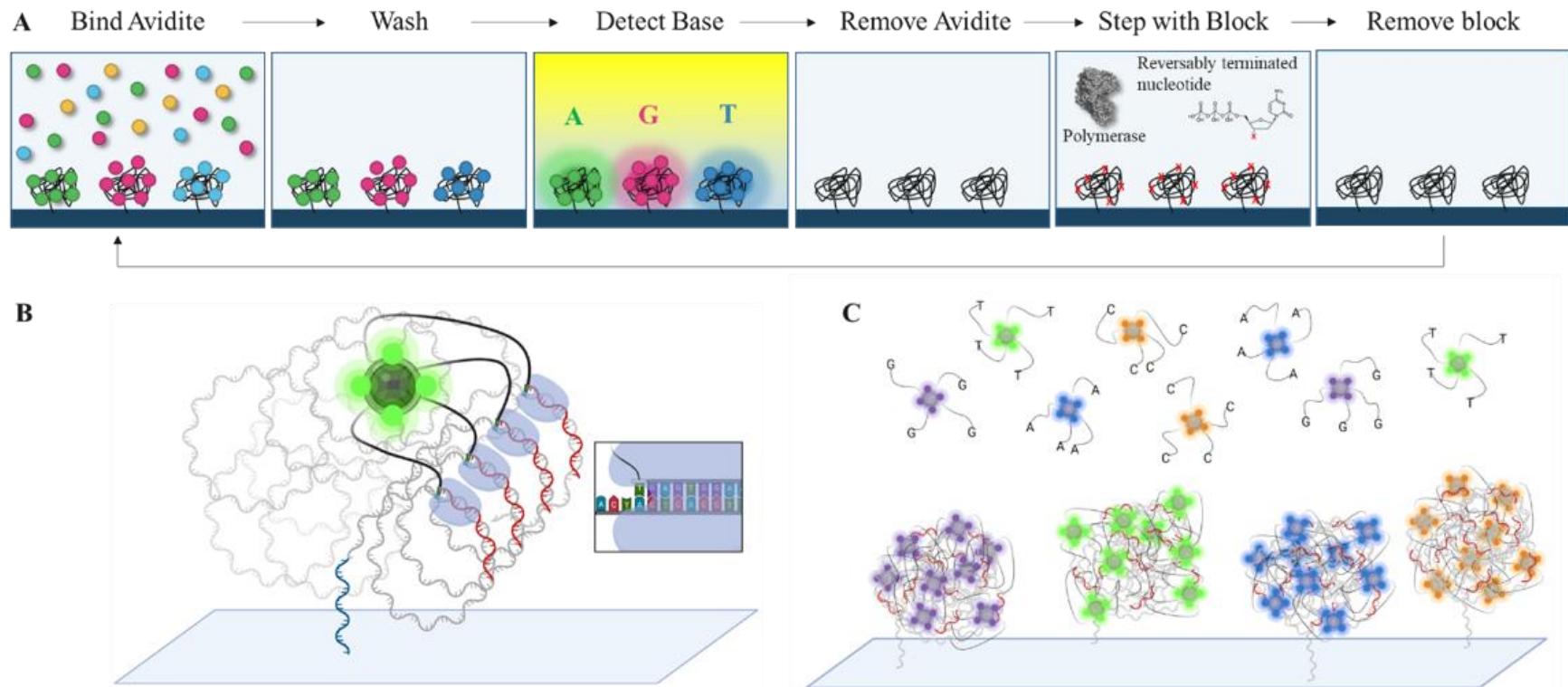


 Element  
Biosciences

# Element Kits

Kit type	2X150 High Output	2X150 Medium Output	2X150 Low Output	2X75 High Output	2X75 Medium Output	2X300 High Output	2X300 Medium Output
Part number	860-00013	860-00012	860-00011	860-00015	860-00014	860-00017	860-00016
Price USD	1377.60	885.60	721.60	885.60	680.00	1541.60	1049.60
USD/Gb	4.59	5.90	9.62	5.90	9.07	8.56	17.49
USD/Million reads	0.84	1.08	1.76	0.54	0.88	4.07	8.4
Total reads (Billion)	2	1	0.5	2	1	0.6	0.2
total Output (Gb)	300	150	75	150	75	180	60
Run Time (Hours)	38	31	27	24	20	60	51
Lanes	2	2	2	2	2	1	1
approx GBP/lane	475.03	305.38	248.83	305.38	248.43	1063.17	723.86

# Avidity base chemistry (ABC)



# AVITI System Architecture Enables Maximum Run Flexibility

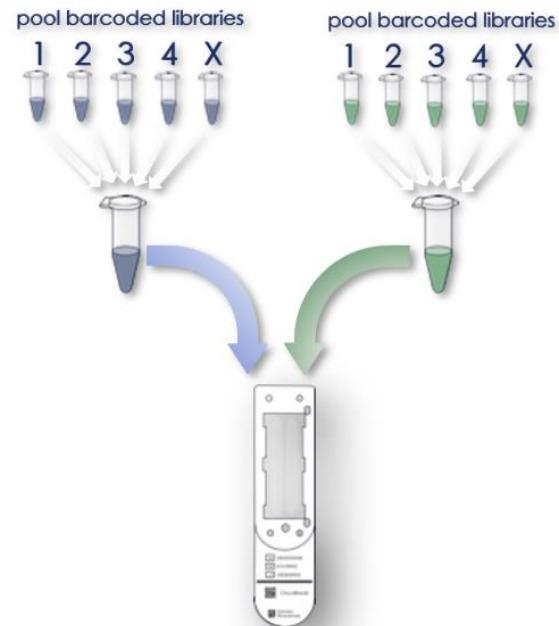


## Dual Independent Flow Cells



Perform two parallel runs with different run parameters

## Individually Addressable Lanes



Sequence separate libraries or pools on a single flow cell



# Cloudbreak Freestyle

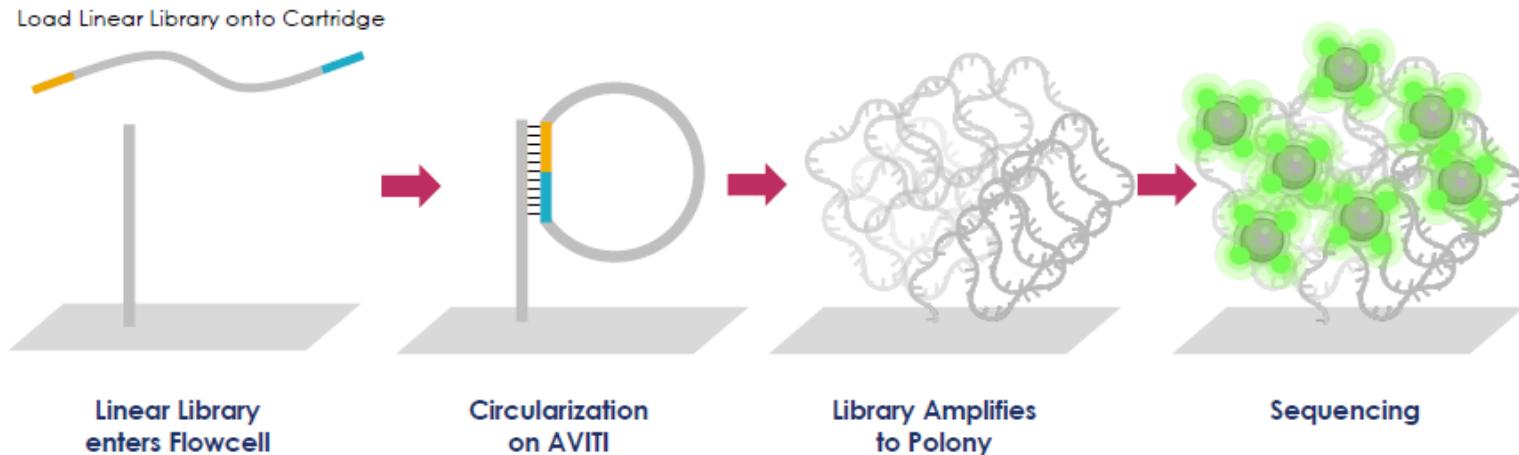
Cloudbreak Freestyle **Enables Linear Library Loading** of your favorite existing library prep solution **Without Circularization**

Saves 1-3 hours of prep time

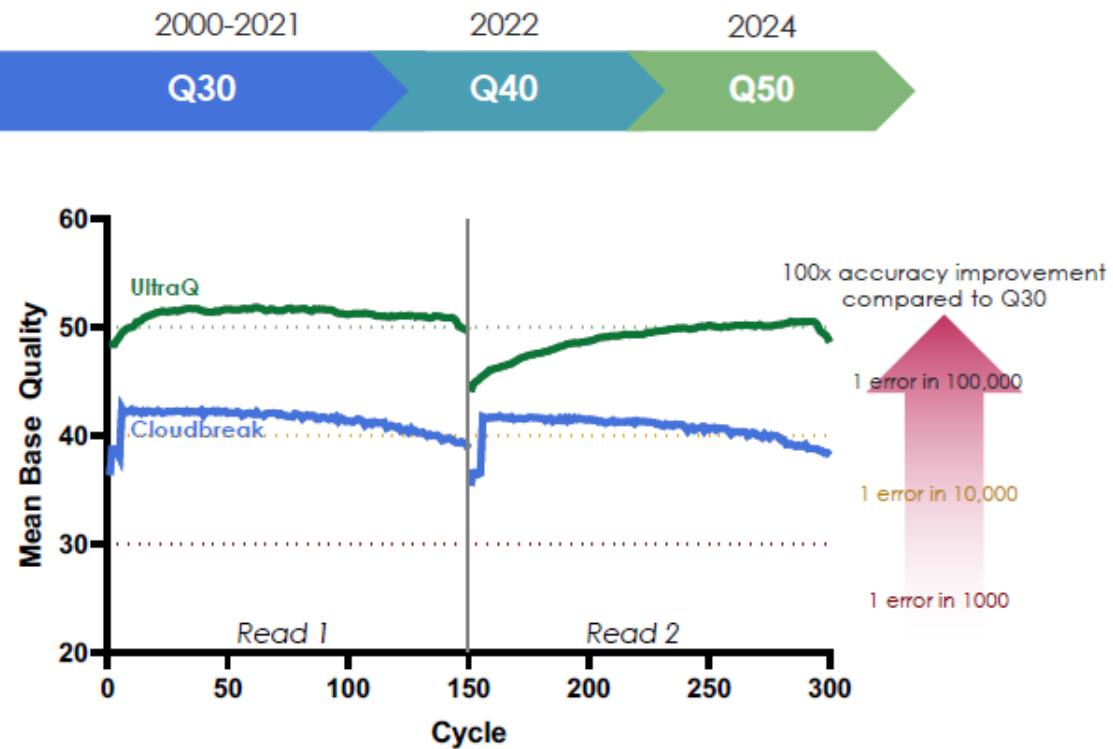
Universally compatible

Maintain high data quality

Automatically On AVITI



# Achieving Q50 with UltraQ



Achieved by innovations in sequencing chemistry & library prep

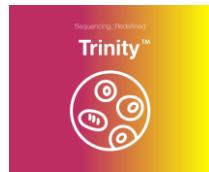


Elevate™ Library prep regents and methods to improve quality



AVIT™ Cloudbreak UltraQ Sequencing Reagents

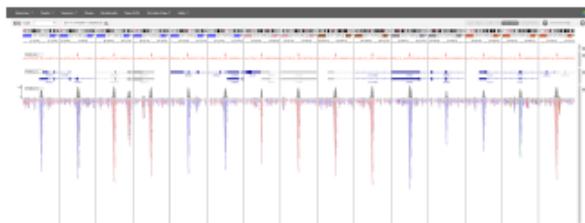
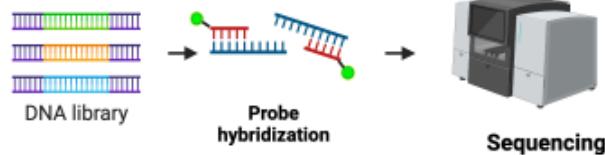
Delivers highest commercially available data quality for challenging applications



# Trinity: On Flow cell hybrid capture

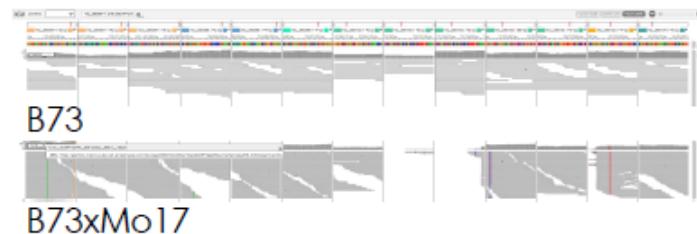
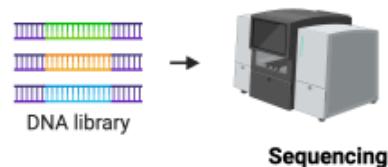
## Two Workflows:

Trinity



- Exome release Q3 2024
- High specificity and sensitivity
- Compatible with 3<sup>rd</sup> party content
- Fast and simple workflow

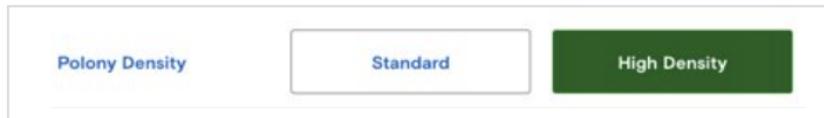
Trinity Plus



- Release after Trinity
- Tunable specificity and sensitivity
- On-flowcell capture

# Expert Mode HD: 20-70% more data for FREE with higher polony density

Click "High Density" on UX



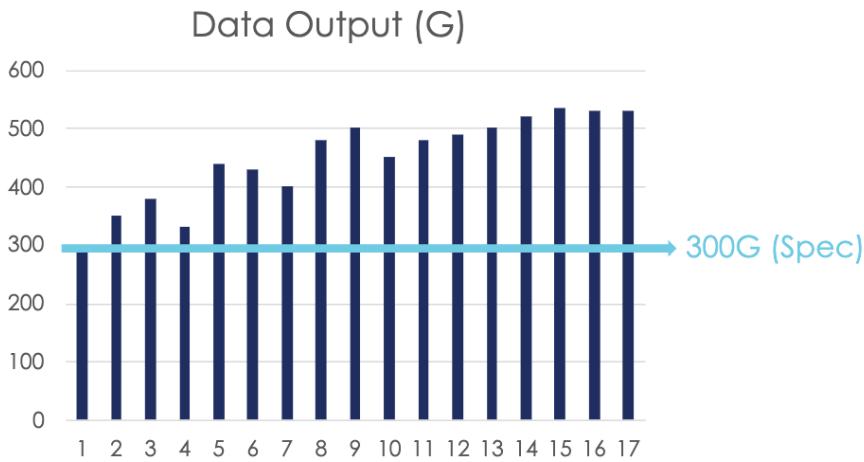
> 90%  
Q30 \*

\$0.72/  
MR \*\*

\$4.8/  
G \*\*

Counting applications  
will benefit the most

Customer reports 1.7x **higher** data output



# Singular Genomics

- \$10-\$15/Gb
- 2 x 150 in 19 hours
- 4 x 4 lane flowcells. Each flowcell can be run and loaded independently.
- Uses similar SBS and clustering to Illumina
- No index hopping. No issue with low complexity





# Ultima



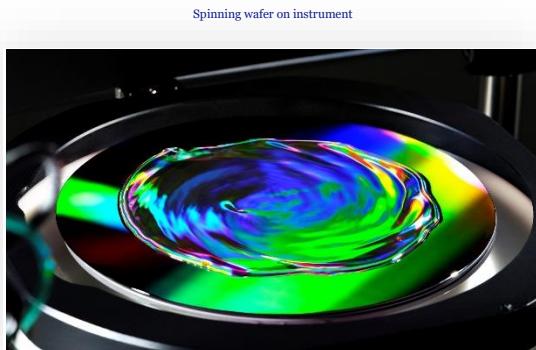
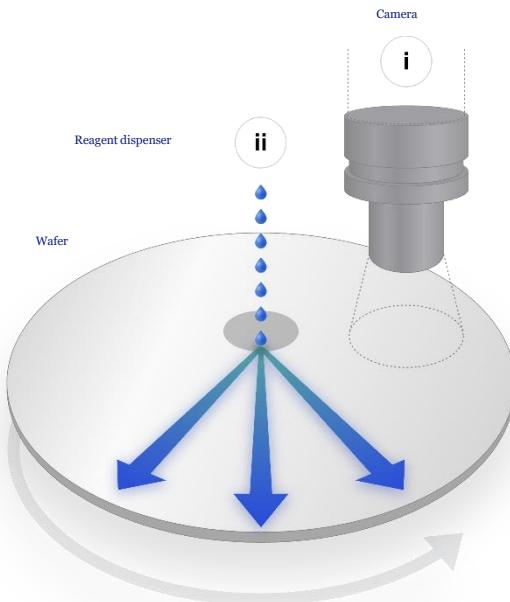
# Ultima

- \$100 genome. \$1/Gb
- SE 300 in ~14hr
- Flow based.
- Some indel errors around homopolymers
- 89% Q30
- Flow based sequencing on a 10" spinning disc





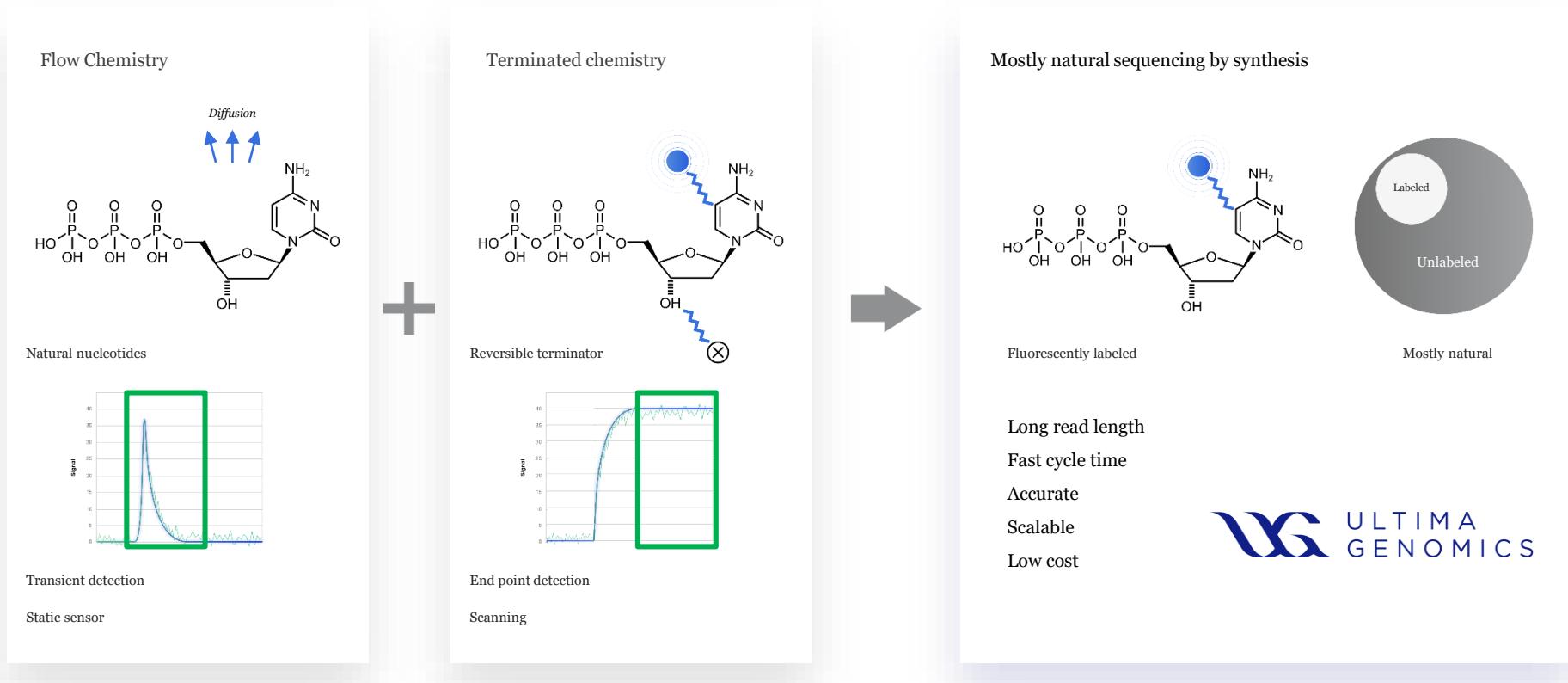
## Revolutionary new sequencing hardware



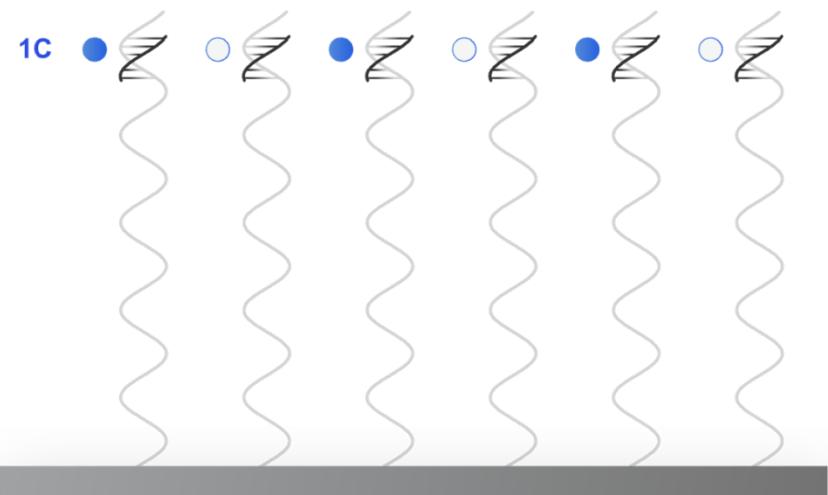
- Low cost (standard substrate)
- ~10B reads per wafer (x2)
- Fast and efficient reagent delivery
- “Air gap” minimized contamination
- Rotational optical scanning

Proprietary and Confidential

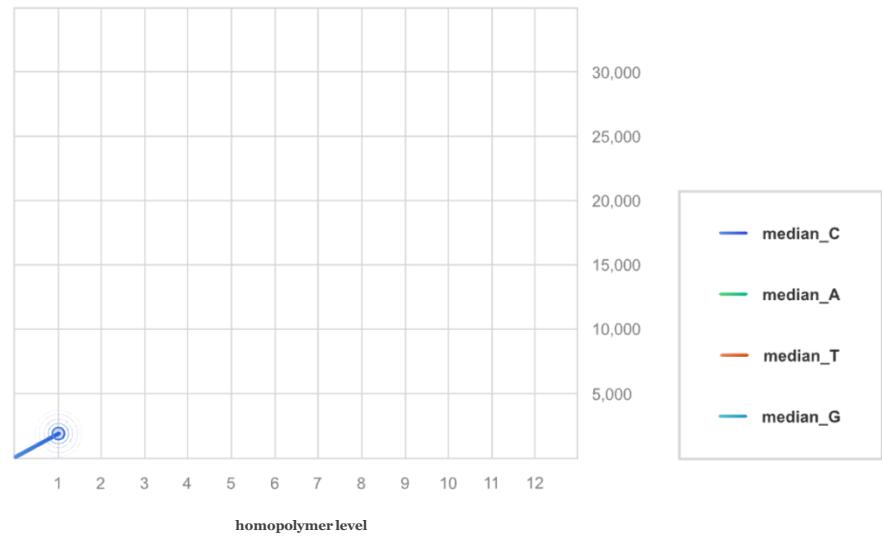
# A novel spin on sequencing chemistry mnSBS



# Mostly natural chemistry combines advantages of flow chemistry with optical scanning

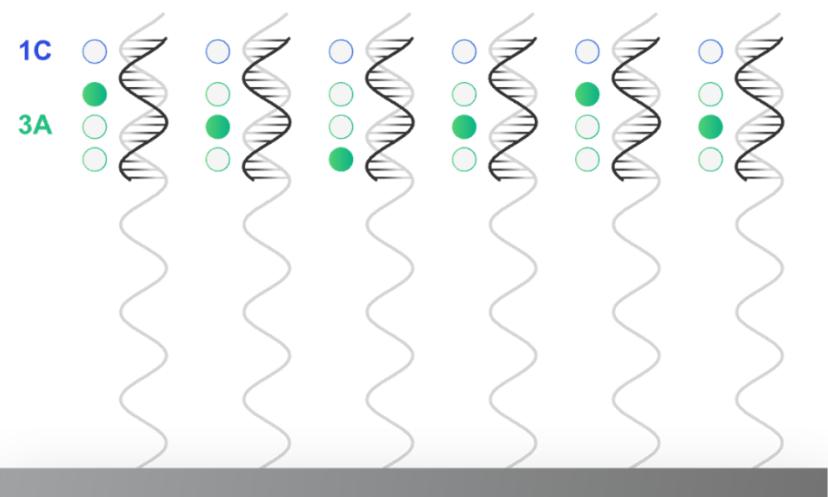


Median signals vs. homopolymer

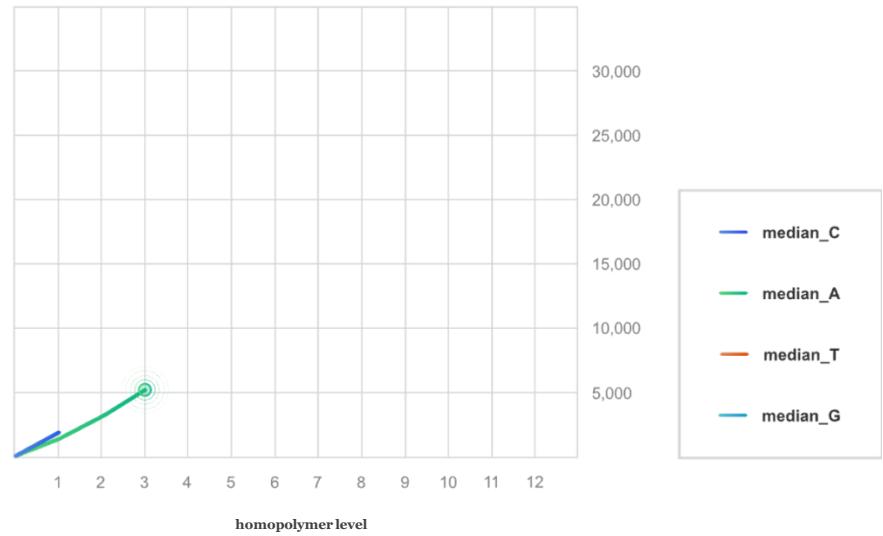


- Majority nucleotides unlabeled to avoid quenching
- Minimal scarring supports longer reads
- Faster runs via 2min wash->image->cleave cycle
- Endpoint detection significantly improves accuracy
- Maintain signal linearity to at least homopolymer length 12
- Machine learning accounts for sequence context

# Mostly natural chemistry combines advantages of flow chemistry with optical scanning



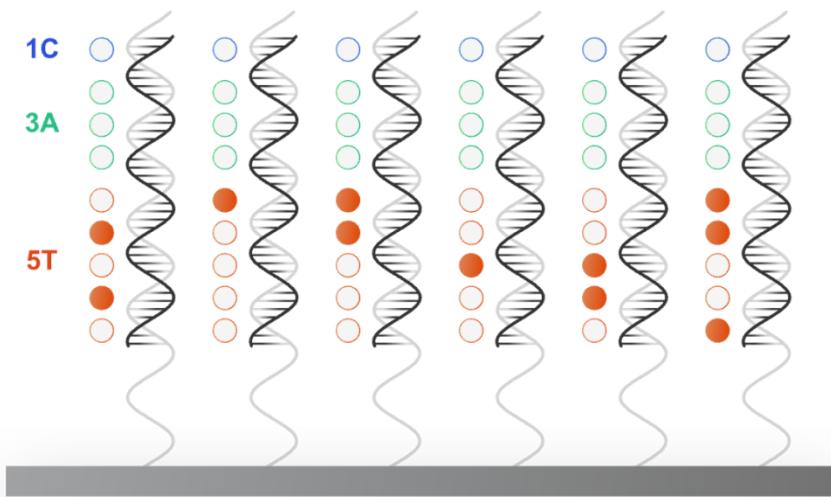
Median signals vs. homopolymer



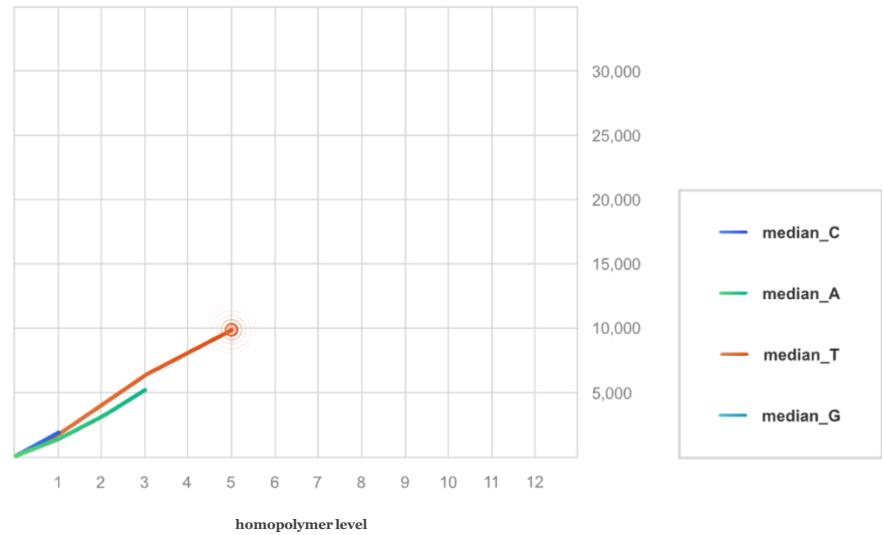
- Majority nucleotides unlabeled to avoid quenching
- Minimal scarring supports longer reads
- Faster runs via 2min wash->image->cleave cycle
- Endpoint detection significantly improves accuracy
- Maintain signal linearity to at least homopolymer length 12
- Machine learning accounts for sequence context

Proprietary and Confidential

# Mostly natural chemistry combines advantages of flow chemistry with optical scanning



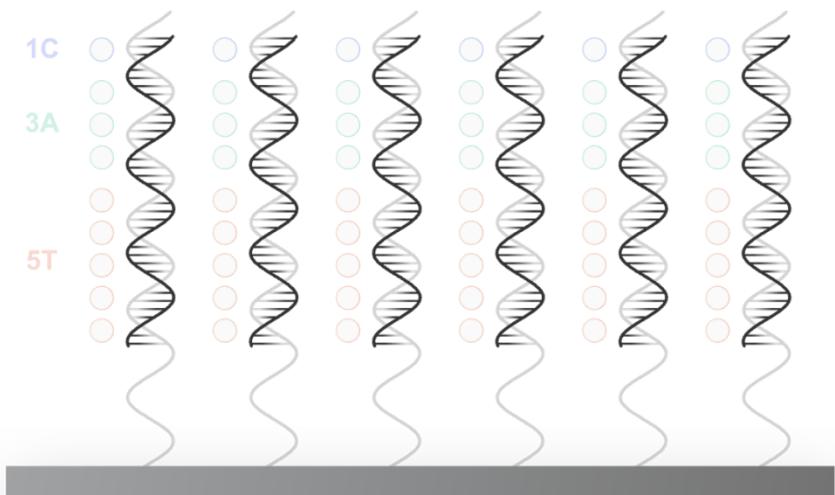
Median signals vs. homopolymer



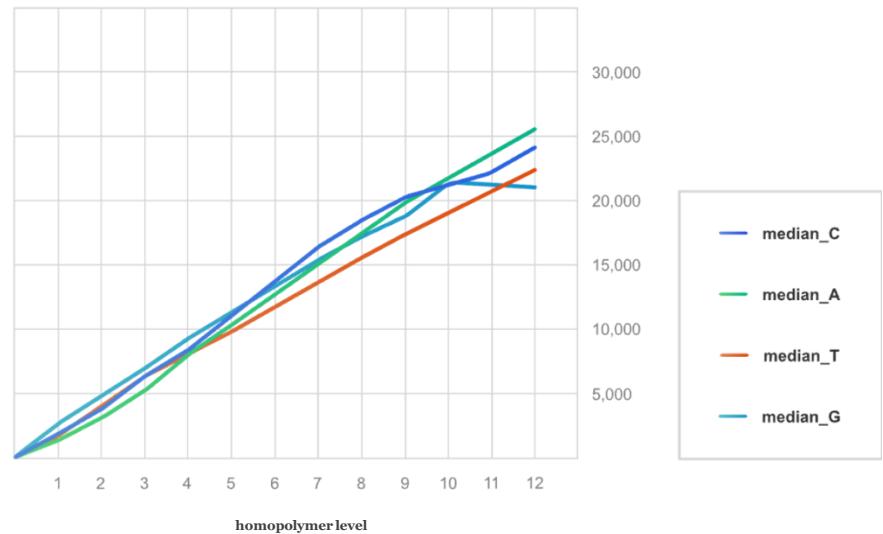
- Majority nucleotides unlabeled to avoid quenching
- Minimal scarring supports longer reads
- Faster runs via 2min wash->image->cleave cycle
- Endpoint detection significantly improves accuracy
- Maintain signal linearity to at least homopolymer length 12
- Machine learning accounts for sequence context

Proprietary and Confidential

# Mostly natural chemistry combines advantages of flow chemistry with optical scanning



Median signals vs. homopolymer



- Majority nucleotides unlabeled to avoid quenching
- Minimal scarring supports longer reads
- Faster runs via 2min wash->image->cleave cycle
- Endpoint detection significantly improves accuracy
- Maintain signal linearity to at least homopolymer length 12
- Machine learning accounts for sequence context

Proprietary and Confidential

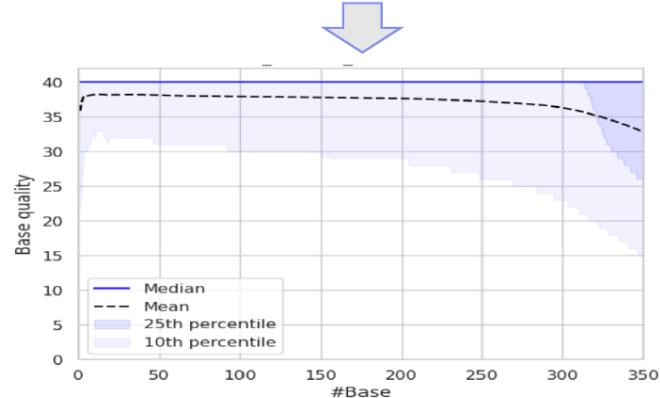
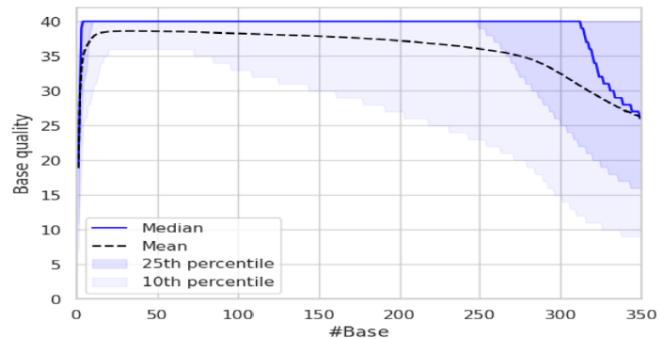


# Continuously improving our performance and data quality

System performance demonstrated on GIAB reference samples:

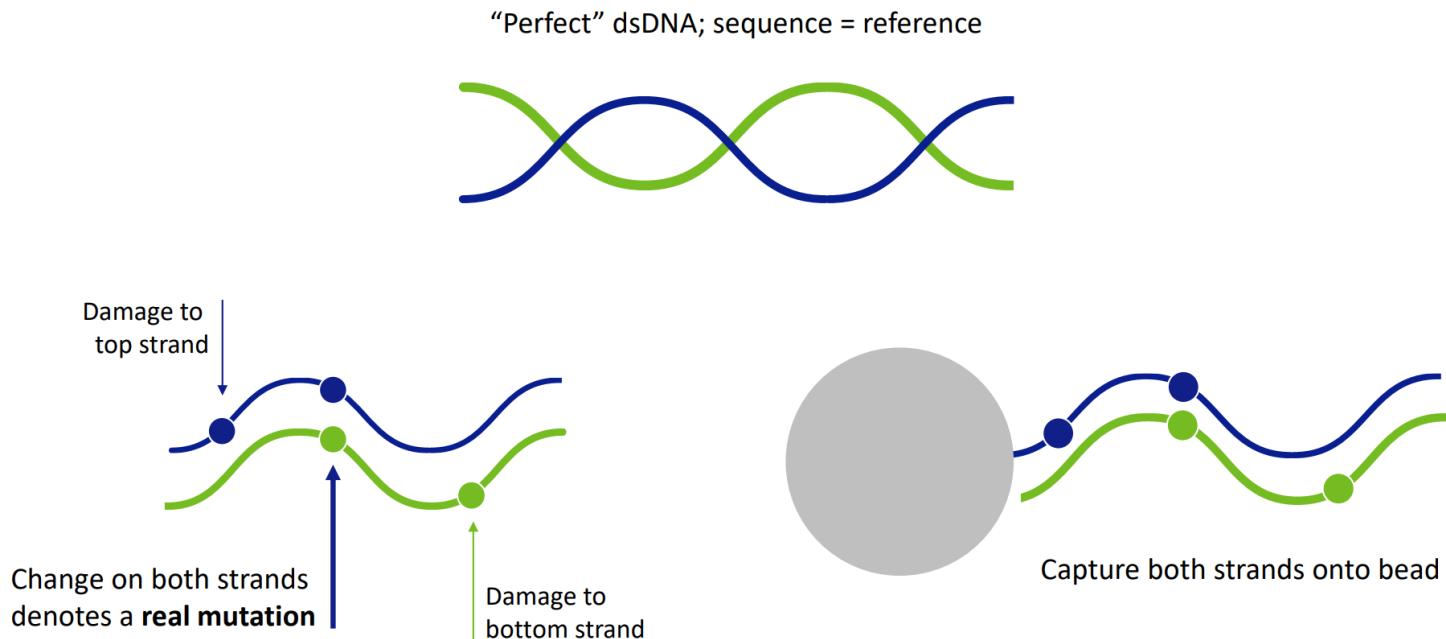
		June '22	February '23	October '23
Read length	Mean	282	294	292
	Mode	310	321	322
Variant calling accuracy* (F1)	SNP	99.6%	99.8%	99.85%
	Indel	96.4%	98.6%	99.06%
Coverage uniformity	F95 Mean	1.8 40X	1.5 39X	1.5X 35X

\* Evaluation region: GIAB v4.2.1  
excluding homopolymer regions of length  $\geq 11$  (0.3% of HCR)  
excluding low complexity regions (1.5 % of HCR)



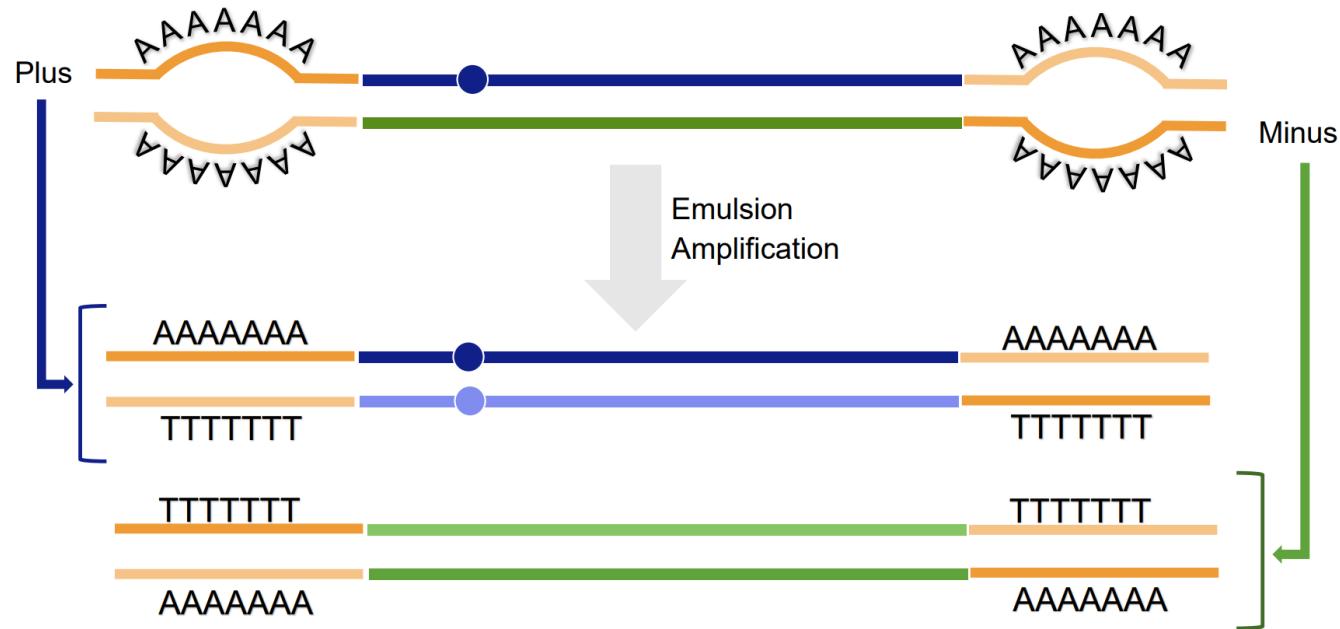
# PPM seq – 1 in a million VAF

## Introducing ppmSeq: paired plus-minus Sequencing



# PPM seq- library prep

Minor adapter change distinguishes strands



# Roche SBX

- Nanopore sequencer enabled by acquisition of Genia and Stratos
- Launch 2026
- 500Mb/sec
- 15B reads in 4 hours
- 7B in 1 hour – fast mode
- Q39 for duplex libraries (200-300bp)
- Q23 for simplex (200-1500bp)
- Instrument \$400-\$600k?
- Consumable pricing = competitive
- Read the paper  
DOI: 10.1101/2025.02.19.639056



## SBX Technology

Designed for Flexibility and Performance with headroom to efficiently Scale into the future



### FLEXIBLE OPERATION

Tunable to sample needs  
Workflow, Throughput, Scale



### READ LENGTH

Duplex: 150 – 350bp (insert)  
Simplex: 50 – 1000bp +



### ACCURACY

Duplex WGS for HG001  
-> Q39 / F1: 99.80% (SNV) / 99.56% (InDel)



### TIME TO RESULT

Sample to VCF WGS in < 7 hours



### THROUGHPUT

WGS for 7 GIAB in 1 Hour @ >30X  
500M Bases/Second



### COST EFFICIENCY

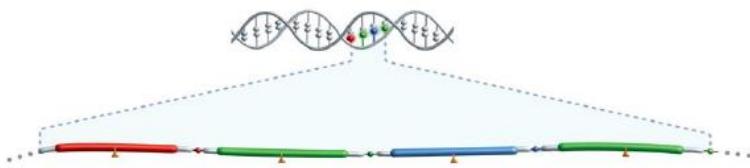
Reusable Sensor Module

*The SBX technology is in development and not commercially available. The content of this material reflects current study results or design goals*

# Roche's High Throughput Sequencing Solution

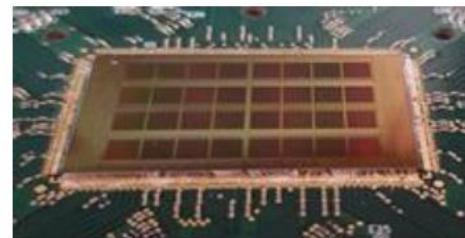
Bringing together two powerful technologies

## Sequencing By Expansion (SBX) Chemistry



S T R A T O S  
genomics inc.

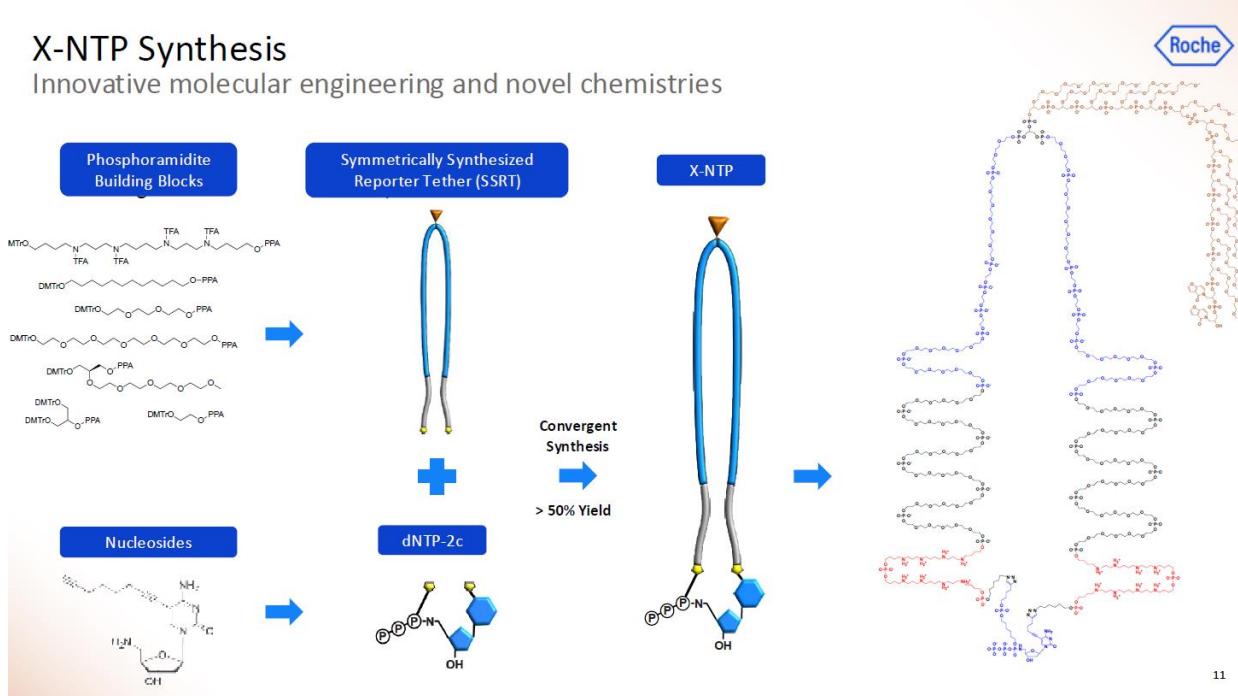
## High Throughput Sensor Array



genia

# Libraries are converted to expandomers Bases copied as XNTPs (50x bigger)

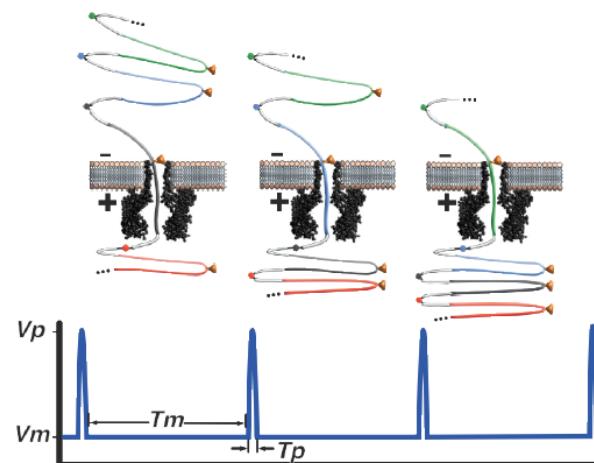
## X-NTP Synthesis Innovative molecular engineering and novel chemistries



11

## Accurate Synchronized Measurement

Efficient and accurate deterministic translocation



Translocation Control Element

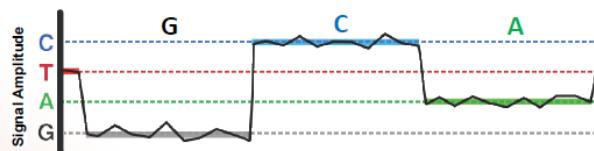
Pauses the reporter code within the nanopore for measurement

Voltage Pulse

Advances Xpandomer one reporter code at a time

Pulsing Rate

Modulates throughput



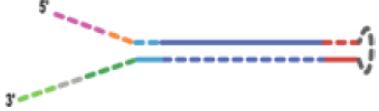
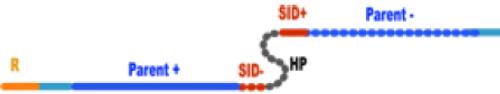
Translated Xp Basecalls



## SBX Library Structures and Workflows

Different library formats, depending on requirements

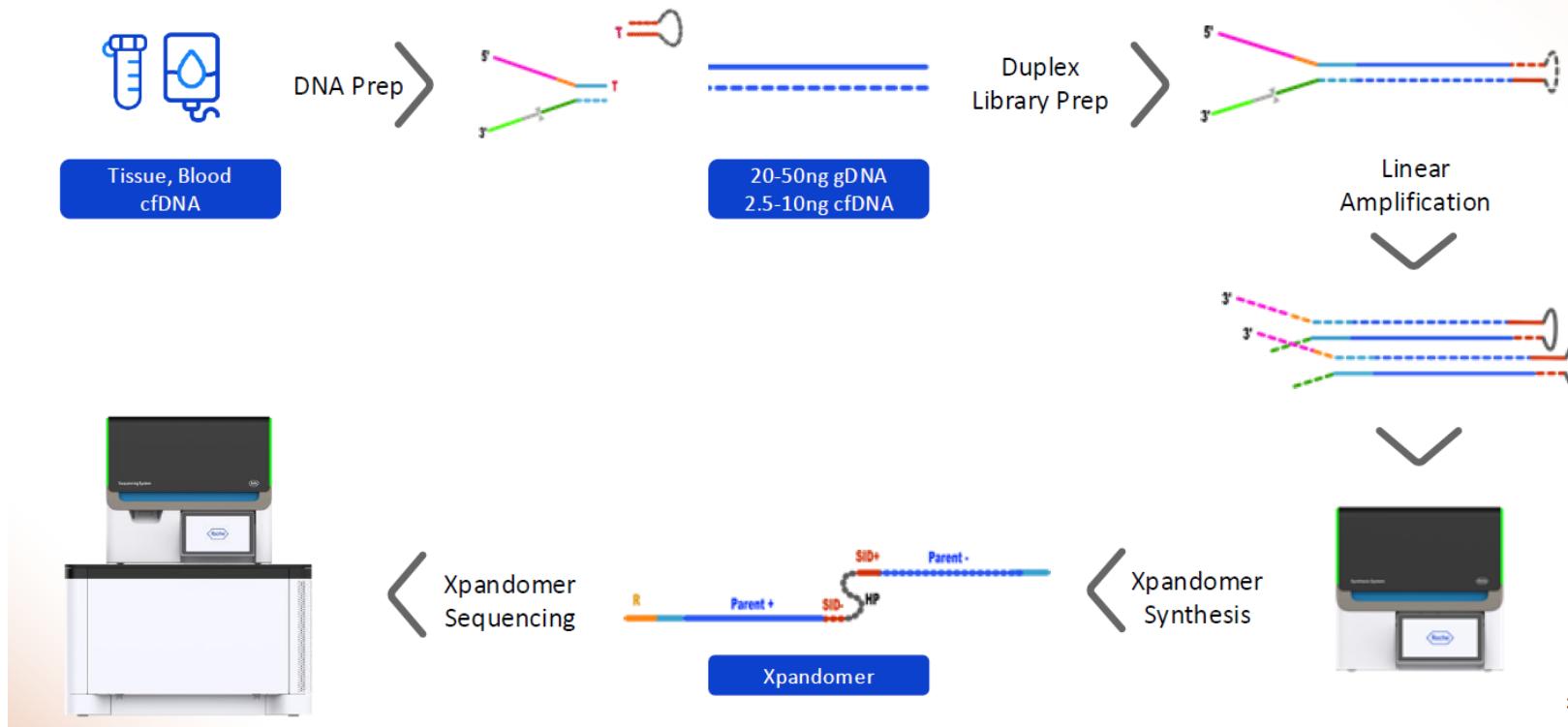


	SBX-Duplex	SBX-Simplex		
Library				
Xpandomer				
	High Accuracy	Ultra-High Throughput		
	200 - 300 bp	< 200 bp      200 - 500 bp      500 - 1500+ bp		
Potential Future Applications	WGS/WES Somatic Oncology MRD Methylation Rapid WGS Panels	WTS Proteomics Spatial Fragment-omics Probe based	WTS Fragment-omics Spatial Panels	DNA Structural Variants RNA Isoforms

24

## SBX-Duplex (SBX-D)

High-accuracy SBX sequencing method



25

# SBX-D GIAB Demonstration

5.3B duplex reads in 1 hour Sequencing run (7-plex)



Sample	Total Duplex Reads (B)	Mean Insert Read Length	Median Coverage	GATK + Roche Machine Learning F1 Score		DeepVariant F1 Score	
				SNV	Indels	SNV	Indels
HG001	0.79	235	<b>38</b>	99.77%	99.54%	99.80%	99.56%
HG002	0.71	233	<b>34</b>	99.69%	99.44%	99.75%	99.46%
HG003	0.76	231	<b>37</b>	99.66%	99.46%	99.70%	99.47%
HG004	0.80	231	<b>38</b>	99.70%	99.56%	99.73%	99.56%
HG005	0.79	230	<b>38</b>	99.69%	99.59%	99.74%	99.60%
HG006	0.71	233	<b>35</b>	99.68%	99.54%	99.74%	99.58%
HG007	0.73	229	<b>34</b>	99.63%	99.45%	99.71%	99.50%

99.7% } % OF GENOME  
AT  $\geq 10X$   
COVERAGE

Q39 } Q-SCORE

Calculated on GIAB 4.2.1 high confidence region

26

# Long Read Sequencers



PacBio Revio



Oxford Nanopore flongle



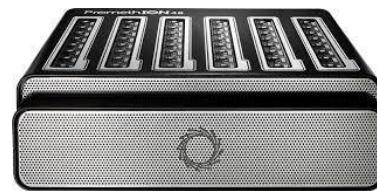
Oxford Nanopore minION



Oxford Nanopore gridION



Oxford Nanopore P2 Solo



Oxford Nanopore P48



# Why Long Read

- » Potential for higher diagnostic rate
- » More complete coverage of genome
- » Better assembly
- » Haplotype linkage
- » Simultaneous modified base detection

# Long Read Platform Comparison

	PacBio Revio	ONT minION	ONT PromethION
Yield/flowcell	120Gb HiFi	10-40Gb	50-200Gb
#Flowcells	4	1	2-48
Output/day	480Gb HiFi	5-20Gb	50-4800
Run time	1 day	2 days	2 days
Cost/Gb	\$5-10	~\$20-50	~\$10
Intrument cost	\$599k	\$1000	\$10k-\$287k
Accuracy	Q30	Q20	Q20
Read length	15-20kb	upto Mb	upto Mb
Modifications	5mC	5mC, 5hmC, 4mC, 6mA	5mC, 5hmC, 4mC, 6mA
Direct RNA	No	Yes	Yes
Time to result	days	minutes	minutes



# Pacific Biosciences



Onso

» Short Read sequencer



Revio

» Long Read sequencer



# PacBio Long Read

- » Long Read Platforms  
RS>RSII>Sequel>SequelII>Revio
- » SMRT. Single Molecule Real Time
- » 3<sup>rd</sup> generation?

# Pacific Biosciences

## Sequel II/IIE (2018-2022)



- » 8 million ZMWs/SMRT
- » \$495K/\$525 instrument



# » Revio system

## Throughput



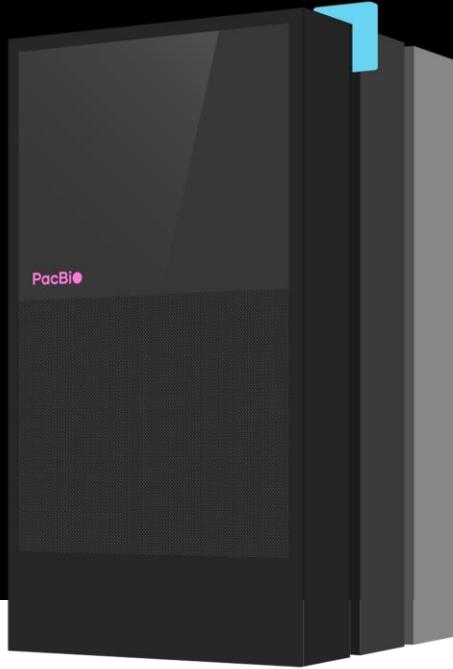
**480 Gb**  
HiFi yield per run with SPRQ



**24 hour**  
Sequencing time



**1,300 30x genomes / year**  
2600 per year to 20x



List price, USD

**\$599,000**

**\$995/genome at 30x\***

**<\$500 at 20x**

\* \$995 for sequencing reagents for one Revio SMRT Cell, which has an expected yield of 90 Gb, equivalent to a 30x human genome. Expected pricing subject to change, your local sales representative can provide detailed pricing in your currency.

PacBio

Learn more:



# Nov' 24 Low Throughput Benchtop

PacBio



## Vega benchtop system HiFi sequencing within reach



Premium HiFi data,  
accessibly priced



Built on proven  
HiFi technology



Sequence  
your way



Diverse  
applications

Vega price  
\$169k system  
\$1,100 per run

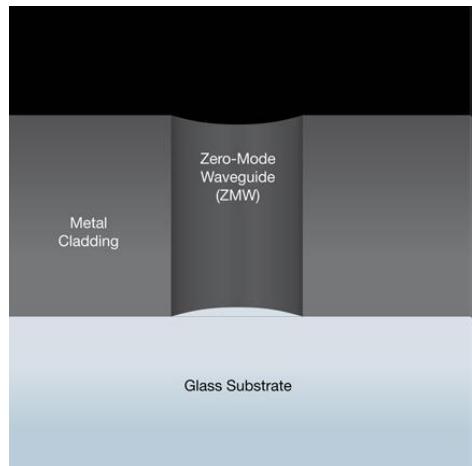
Vega Access  
\$79k system  
\$1,750 per run

Compatible with  
HiFi library prep and  
analysis workflows

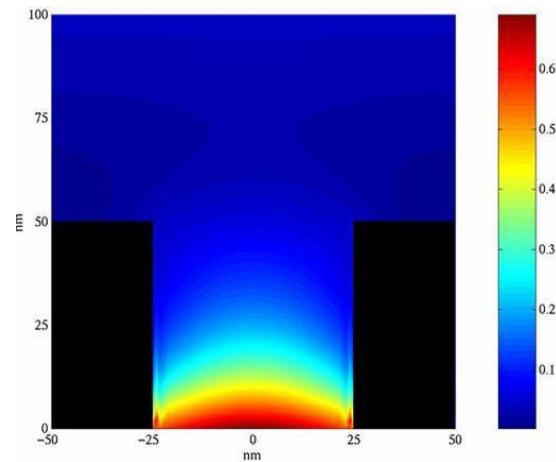
Take control of your  
data and streamline  
project timelines

Full-length RNA, whole  
genomes, targeted  
panels and more

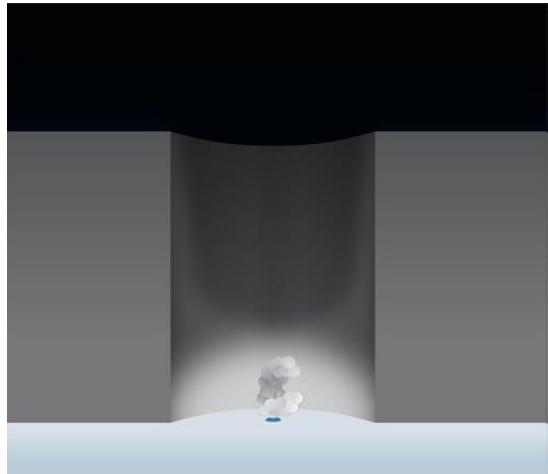
# Pac Bio Technology



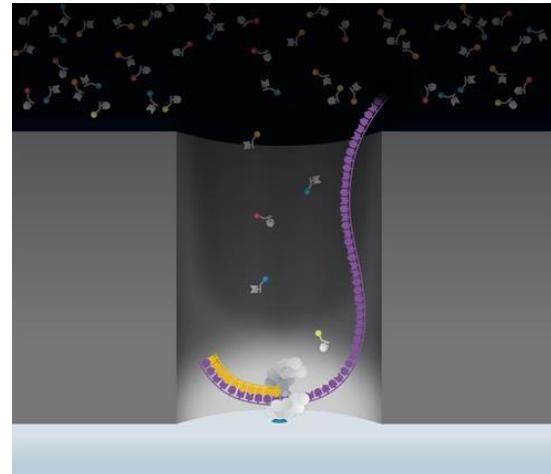
Individual ZMW



Laser light illuminates the ZMW

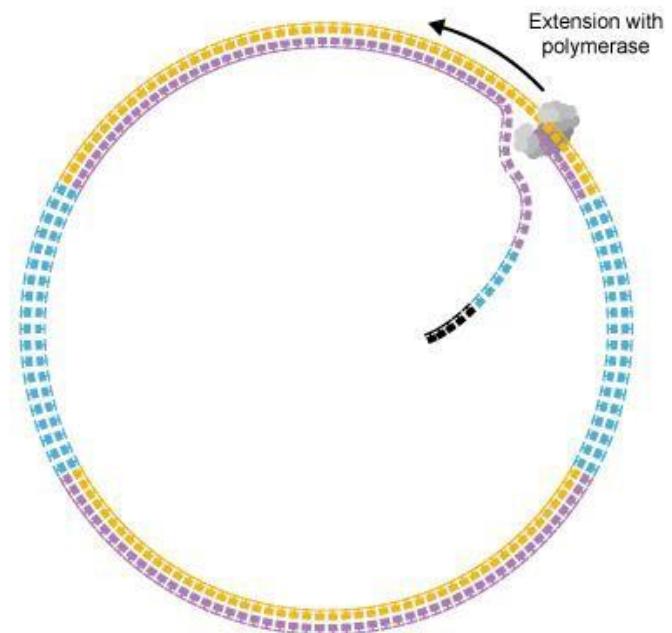
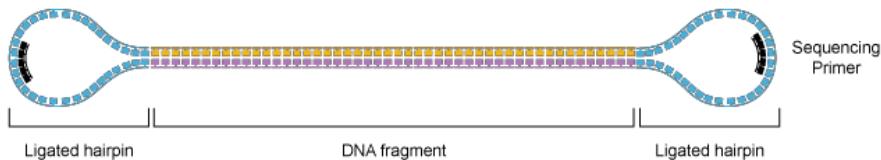


ZMW with DNA polymerase



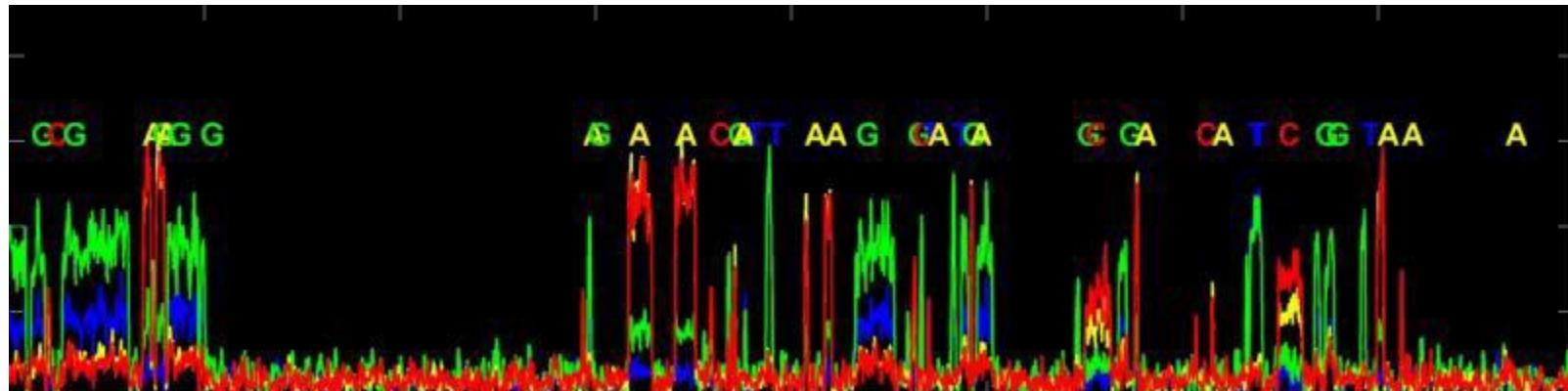
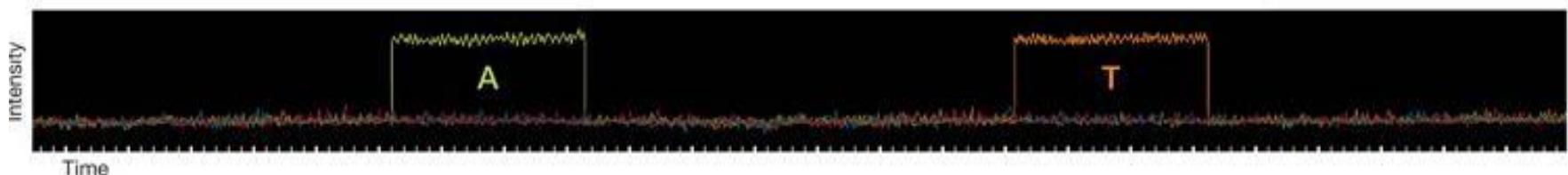
ZMW with polymerase + nucs.

# PacBio Template Preparation

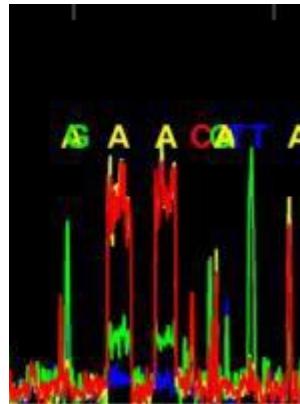


- 1 Anneal primer**
- 2 Bind polymerase**
- 3 Sequence**

# PacBio Sequencing

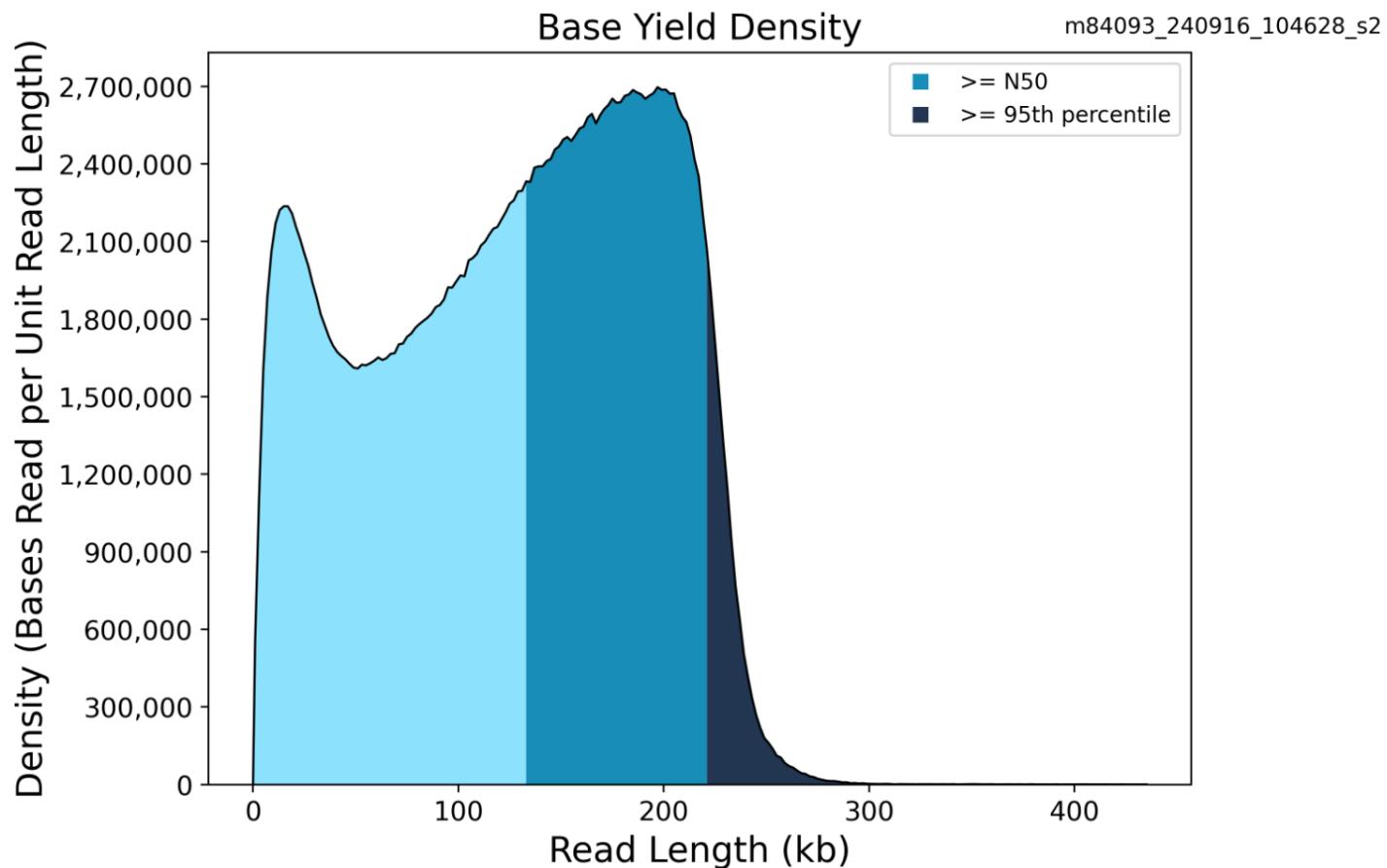


# PacBio Sequencing



- » Some bases added very quickly and missed
- » Some wrong bases flirt with active site and go away
- » SMRT cell has millions of ZMWs (8M Sequel, 25M Revio)

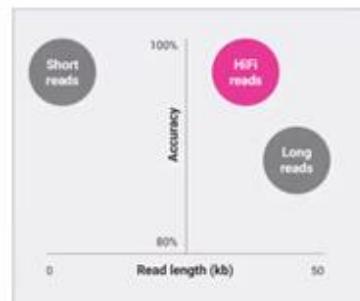
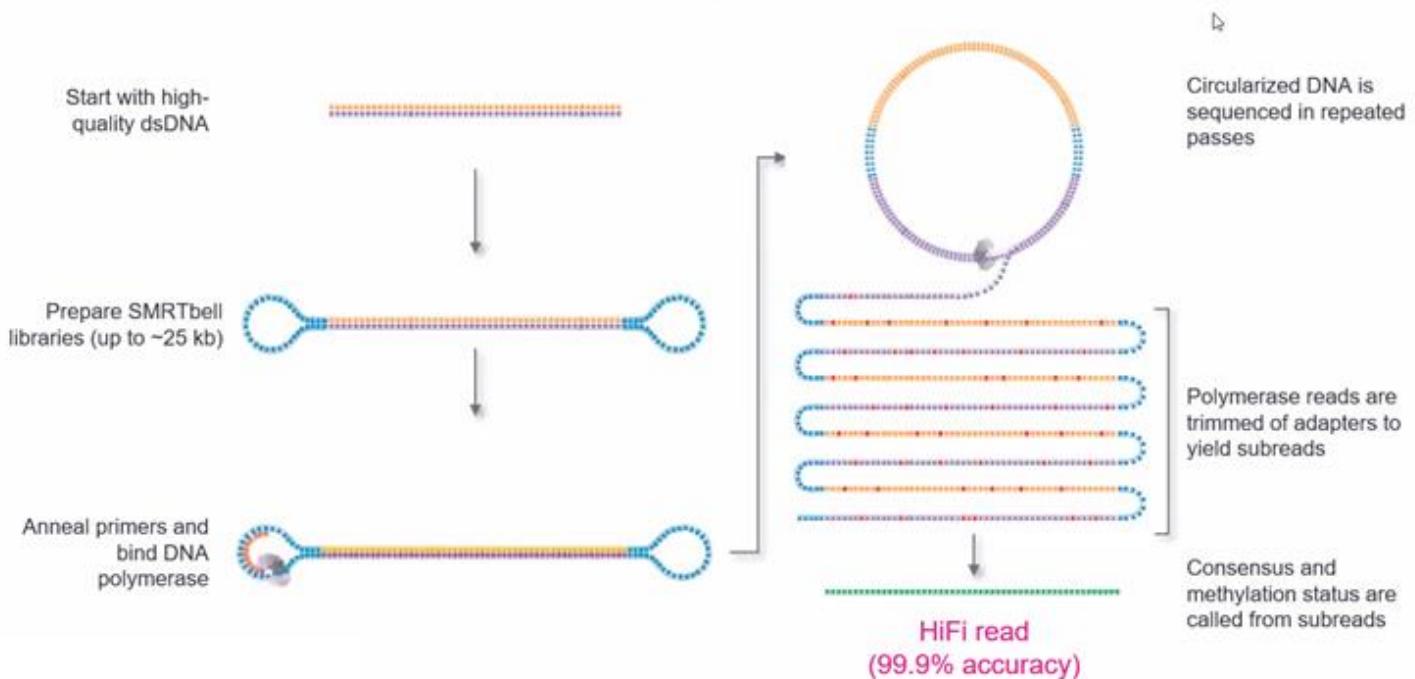
# Revio Polymerase Read Lengths



## What are HiFi reads?

HiFi reads are produced using the circular consensus sequencing (CCS) mode on PacBio long-read systems and provide base-level resolution with 99.9% single-molecule read accuracy

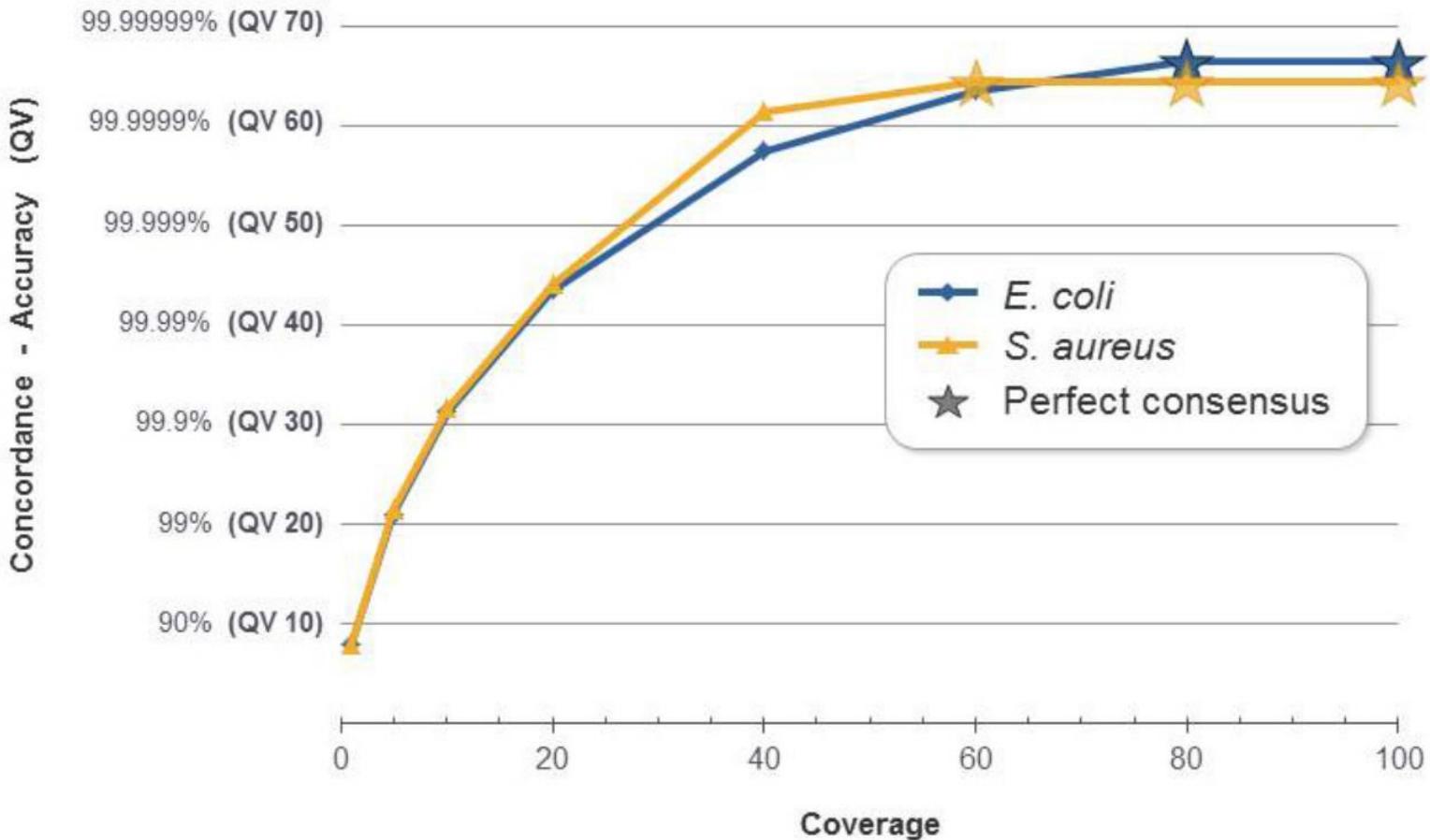
### HiFi read generation



### Benefits of HiFi reads

- Long read lengths up to 25 kb
- High read accuracy 99.9%
- Easy library preparation
- Low coverage requirements
- Small file sizes to minimize compute time
- A single technology solution for a range of applications

# Accuracy



# Long read can access 193 medically relevant genes that short read cannot

## PacBio Claims Differentiation Through Ability to Address Challenging and Medically Relevant Genomic Regions

nature  
biotechnology

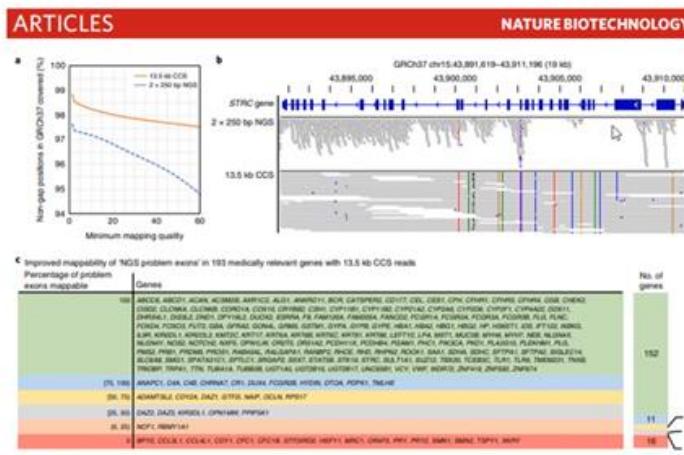
### ARTICLES

<https://doi.org/10.1038/s41587-019-0217-9>

#### Accurate circular consensus long-read sequencing improves variant detection and assembly of a human genome

Aaron M. Wenger<sup>1,2\*</sup>, Paul Peluso<sup>1,3†</sup>, William J. Rowell<sup>2</sup>, Pi-Chuan Chang<sup>2</sup>, Richard J. Hall<sup>2</sup>, Gregory T. Concepcion<sup>2</sup>, Jana Ebler<sup>3,4</sup>, Arkarachai Fungtammasan<sup>1</sup>, Alexey Kolesnikov<sup>2</sup>, Nathan D. Olson<sup>2</sup>, Armin Töpfer<sup>2</sup>, Michael Alonge<sup>2</sup>, Medhat Mahmoud<sup>2</sup>, Yufeng Qian<sup>1</sup>, Chen-Shan Chin<sup>2,5</sup>, Adam M. Phillippy<sup>1,6</sup>, Michael C. Schatz<sup>2</sup>, Gene Myers<sup>1,6</sup>, Mark A. DePristo<sup>2</sup>, Jue Ruan<sup>1,2</sup>, Tobias Marschall<sup>2,3,4</sup>, Fritz J. Sedlazeck<sup>2,3</sup>, Justin M. Zook<sup>2</sup>, Heng Li<sup>2,3</sup>, Sergey Koren<sup>2</sup>, Andrew Carroll<sup>2</sup>, David R. Rank<sup>2</sup> and Michael W. Hunkapiller<sup>2,3\*</sup>

The DNA sequencing technologies in use today produce either highly accurate short reads or less-accurate long reads. We report the optimization of circular consensus sequencing (CCS) to improve the accuracy of single-molecule real-time (SMRT) sequencing (PacBio) and generate highly accurate (99.8%) long high-fidelity (HiFi) reads with an average length of 13.5 kilobases (kb). We apply this approach to sequence the GRCh37 genome and obtain CCS coverage with an N50 of 2,238,500 bases and a concordance rate of at least 99.99% for short-nucleotide variants (SNVs), 95.99% for insertion/deletions (>5 bp) (indels) and 95.99% for structural variants. Our CCS method matches or exceeds the ability of short-read sequencing to detect small variants and structural variants. We estimate that 2,434 discordances are correctable mistakes in the 'genome in a bottle' (GIAB) benchmark set. Nearly all (99.64%) variants can be phased into haplotypes, further improving variant detection. Denovo genome assembly using CCS reads alone produced a contiguous and accurate genome with a contig N50 of >15 megabases (Mb) and concordance of 99.997%, substantially outperforming assembly with less-accurate long reads.



**Fig. 2 | Mappability of the human genome with CCS reads.** **a**, Percentage of the nongap GRCh37 human genome covered by at least ten reads from 28-fold coverage NGS (2 x 250 bp, HiSeq 2500) and CCS (13.5 kb) datasets at different mapping quality thresholds. **b**, Coverage of the congenital deafness gene STRC in HG002 with 2 x 250 bp NGS reads and 13.5 kb CCS reads at a mapping quality threshold of 10. **c**, Improvement in mappability with 13.5 kb CCS reads for 193 human genes previously reported as medically relevant and problematic to map with NGS reads\*.



## INTRONIC INSERTION IN AUTS2 (AUTISM)



PacBio reads  
show insertions

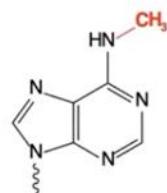
Illumina reads  
do not map

Tandem repeat

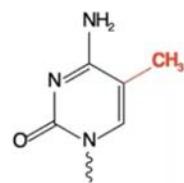
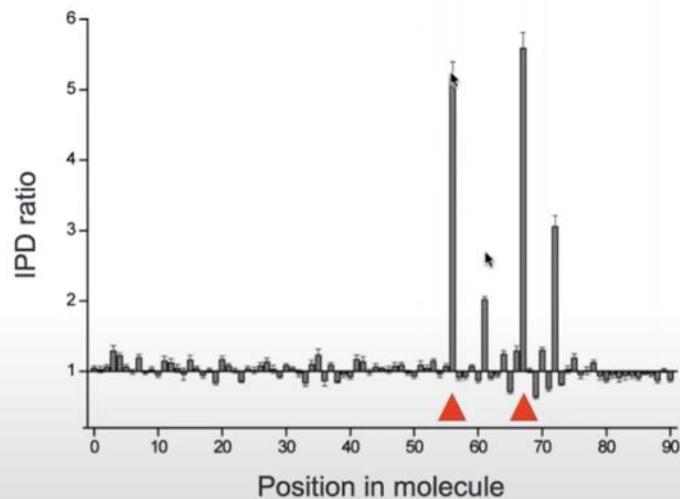


# Epigenetics

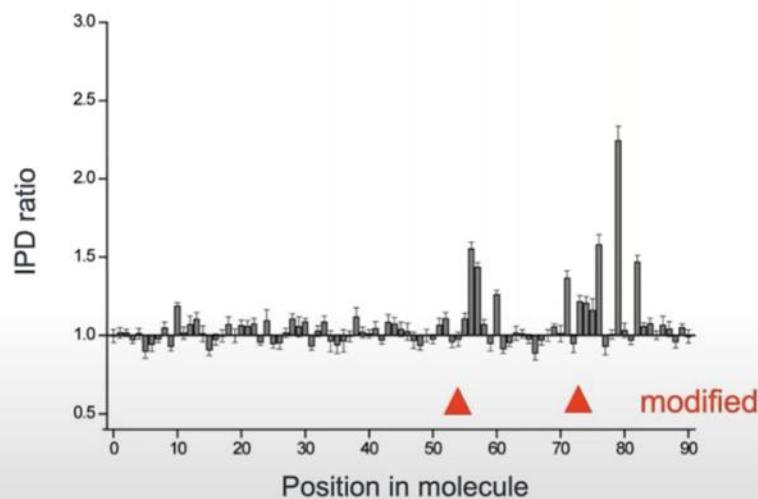
Different modifications have distinct kinetic signatures



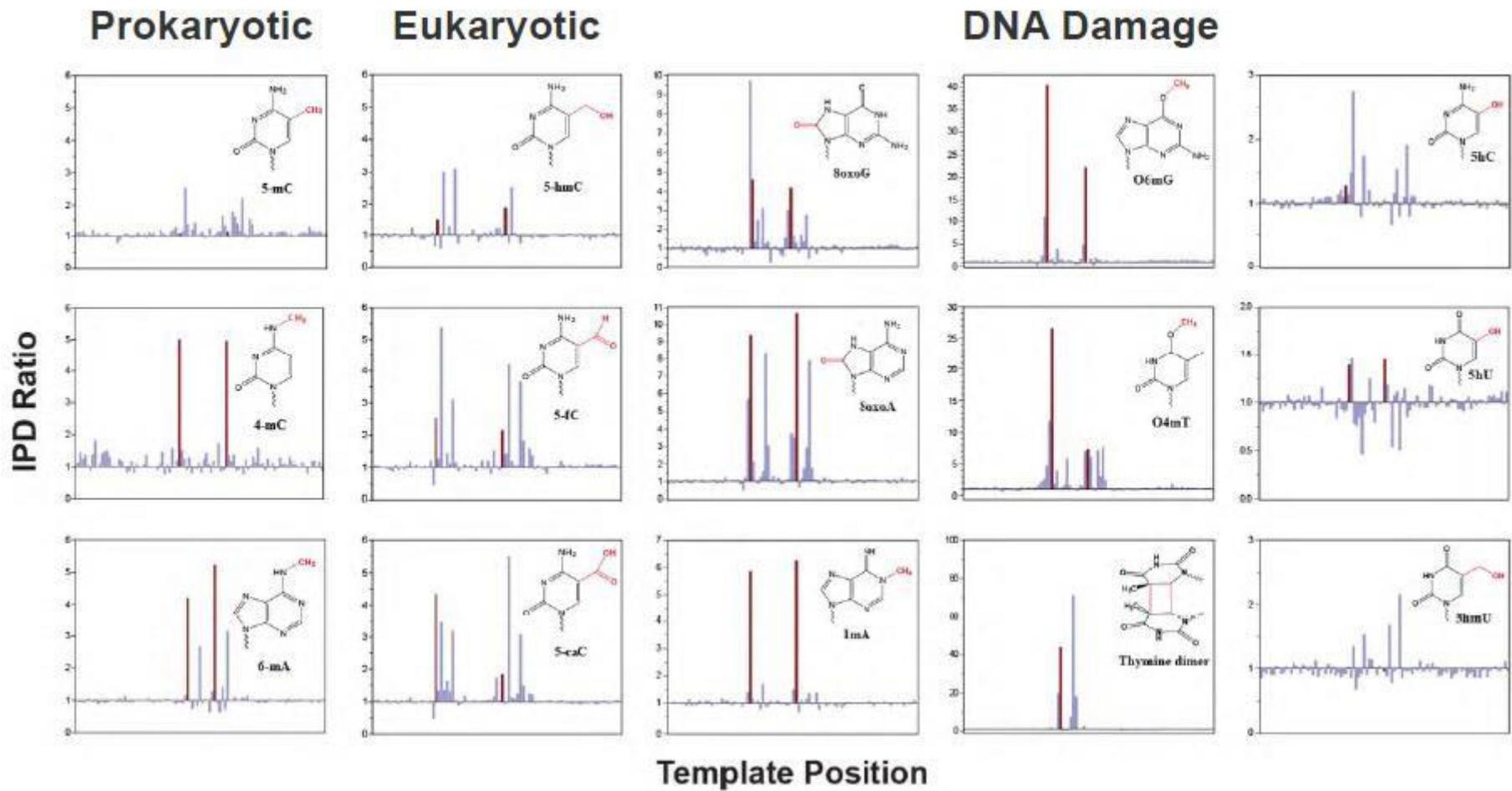
6mA  
strong, localized signal



5mC  
diffuse signal



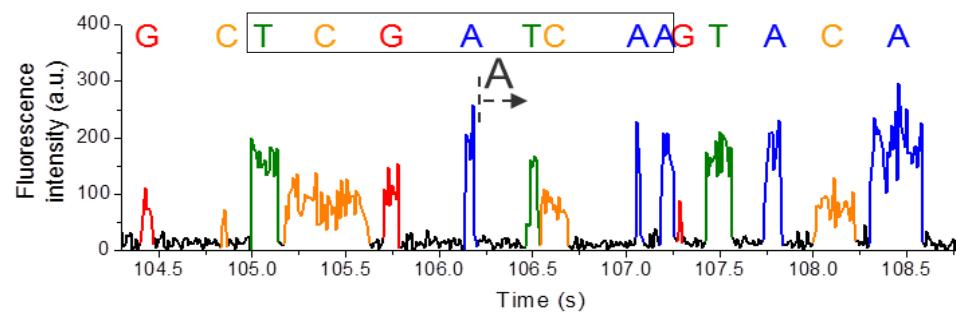
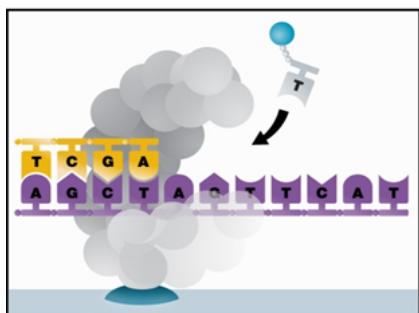
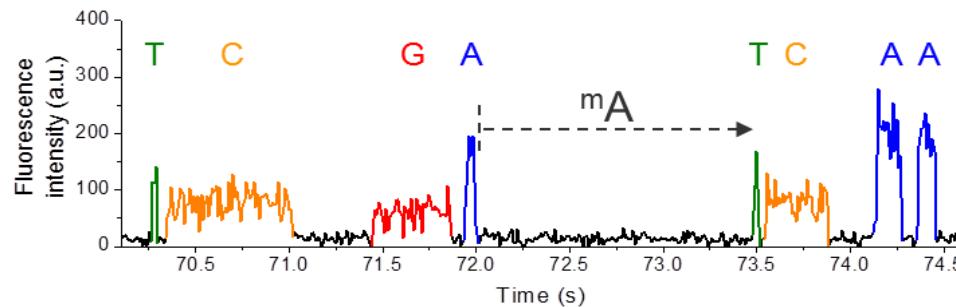
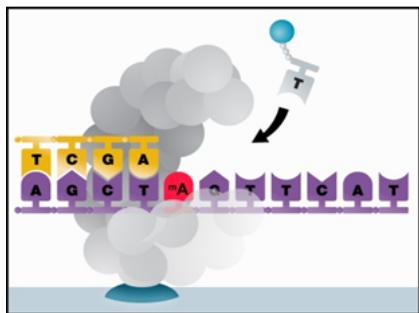
# Base Modifications





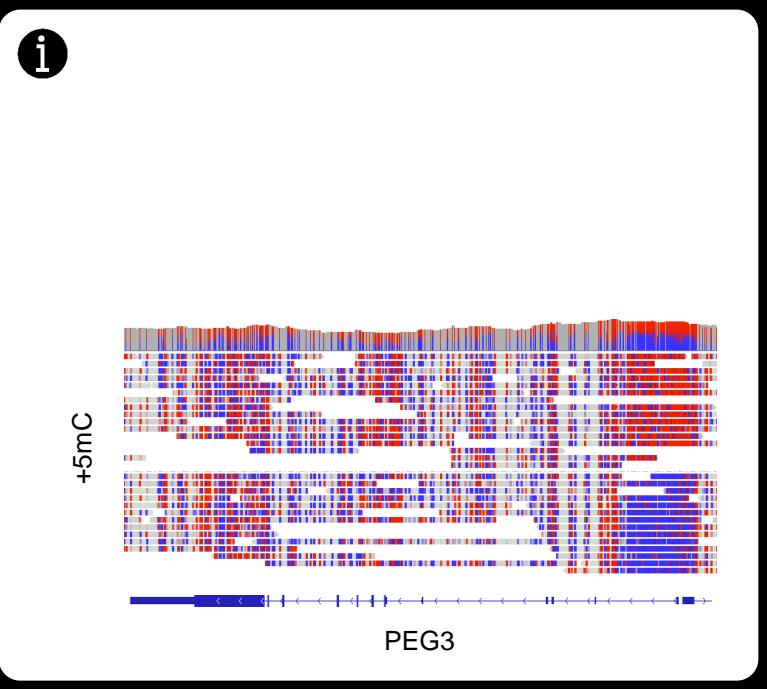
# DETECTION OF DNA BASE MODIFICATIONS USING KINETICS

Example: N<sup>6</sup>-methyladenine

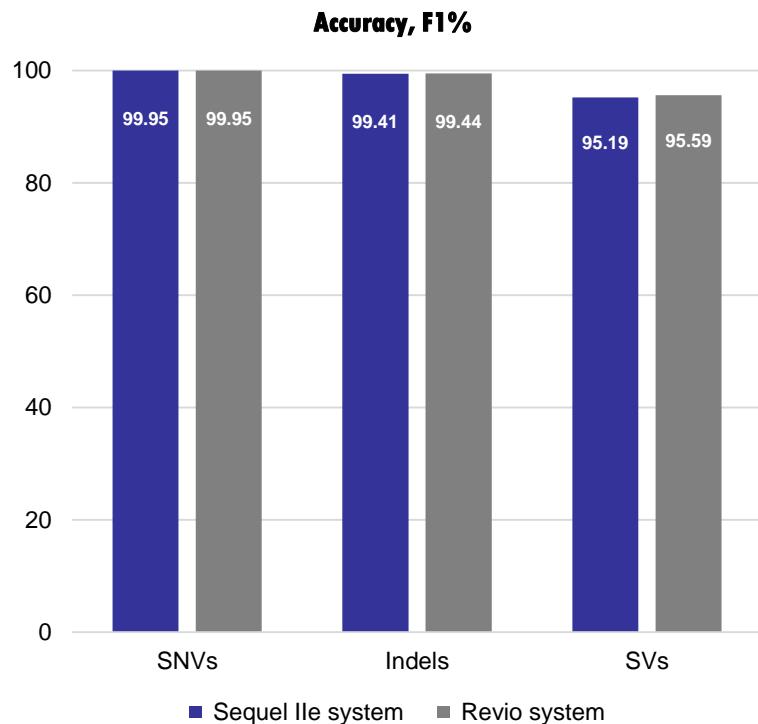


- SMRT Sequencing uses kinetic information from each nucleotide addition to call bases
- This same information can be used to distinguish modified and native bases by comparing results of SMRT Sequencing to an *in silico* kinetic reference for incorporation dynamics without modifications.

# Revio variant calling and methylation performance



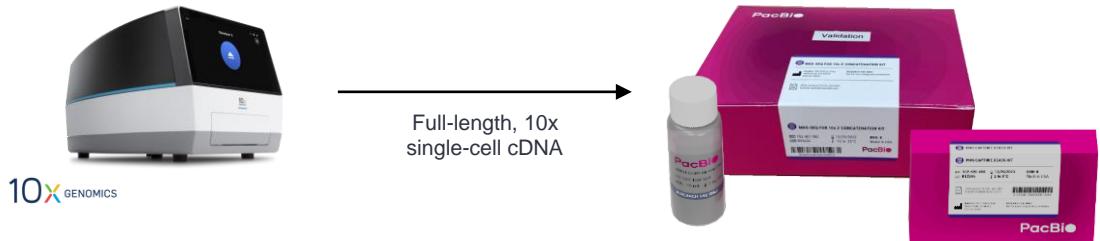
PacBio



HG002 at maternally imprinted PEG3 locus  
Data for internal beta testing [https://downloads.pacbcloud.com/public/revio/2022Q4/HG002rep3/analysis/HG002.m84005\\_220827\\_014912\\_s1.GRCh38.bam](https://downloads.pacbcloud.com/public/revio/2022Q4/HG002rep3/analysis/HG002.m84005_220827_014912_s1.GRCh38.bam)  
Variant calling measured against Genome in a Bottle benchmarks, HG002

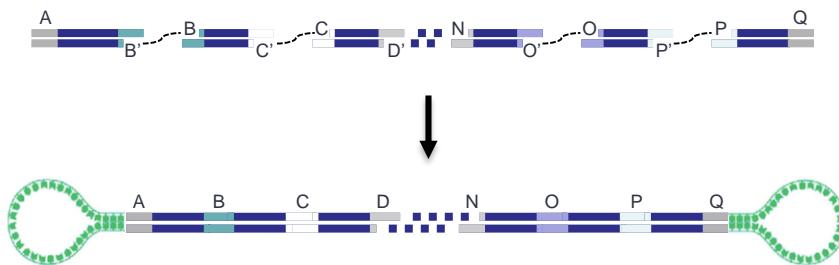
131

# MAS-Seq: high-throughput, full-length single-cell sequencing



## MAS-Seq for 10x Single Cell 3' kit (102-659-600)

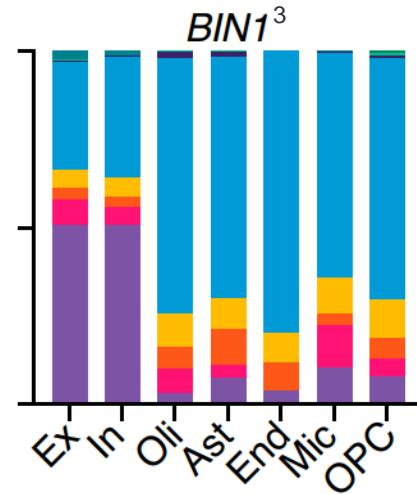
- 16x concatenation



### High-throughput RNA isoform sequencing using programmable cDNA concatenation

✉ Aziz M. Al'Khafaji, ⚭ Jonathan T. Smith, ⚭ Kiran V Garimella, Mehrtash Babadi, ⚭ Moshe Sade-Feldman, Michael Gatzen, Siranush Sarkisova, ⚭ Marc A. Schwartz, Victoria Popic, Emily M. Blaum, Allyson Day, Maura Costello, Tera Bowers, Stacey Gabriel, Eric Banks, Anthony A. Philippakis, Genevieve M. Boland, ⚭ Paul C. Blainey, Nir Hacohen

# Cell atlas, single-cell sequencing



95%

of human multi-exon genes have more than one isoform<sup>1</sup>

>7

isoforms per gene on average<sup>2</sup>

2,900

of brain expressed genes are heritable at isoform level<sup>4</sup>

<sup>1</sup><https://www.sciencedirect.com/science/article/pii/S20001037020305341>

<sup>2</sup><https://www.biorxiv.org/content/10.1101/2022.06.08.495354v1.full>; <sup>3</sup><https://www.pnas.org/doi/full/10.1073/pnas.2114326118>

<sup>4</sup><https://www.medrxiv.org/content/10.1101/2022.10.18.22281204v1>

# Comparison of PacBio with other approaches

	PacBio Revio with SPRQ	SBS sequencing	Nanopore sequencing
Read length	15-20 kb <sup>5</sup>	2x150 bp	10–100 kb
Read accuracy	99.95% (Q33) <sup>1</sup>	99.92% (Q31) <sup>3</sup>	98.90% (Q19) <sup>4</sup>
Run time	24 hours <sup>2</sup>	44 hours	72 hours
Yield	120 - 480 Gb <sup>2</sup>	2,400–3,000 Gb	50–110 Gb
Coverage	Unbiased	Reduced at low and high (GC)	Reduced in low-complexity runs
Genome completeness <sup>6</sup>	✓	✗	✓
Variant calling – SNVs, Indels, SVs <sup>5</sup>	✓	✗	✗
Areas of high homology	✓	✗	✗
Phasing	✓	✗	✓
Methylation - 5mC and 6mA	Every run	Via library prep	Every run



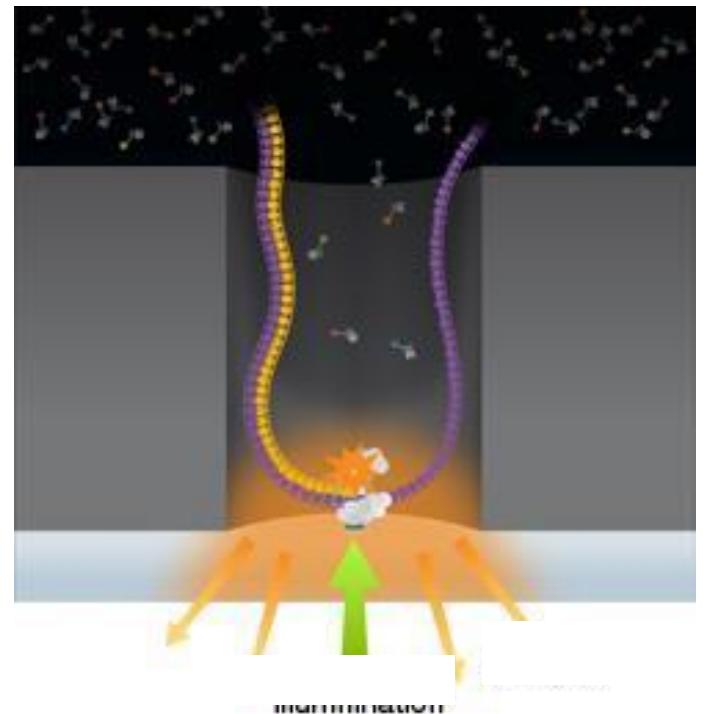
# Pacific BioSciences Sequel/Revio Applications

- » Long read applications
- » *De Novo* sequencing
- » Full length cDNA sequencing
- » Haplotyping
- » Variant calling with access to repetitive regions
  
- » DNA modification studies



# Pacific Biosciences

- » Single polymerase mol. in a 20nm hole
- » Watch incorporation in real time
- » ~2 bases per second
- » Yield 120Gb
- » ~\$5-10/Gb
- » HiFi reads 15-20kb



# PacBio Onso

- Released 2023
- Most accurate short read
- 90% > Q40, Q50 possible
- \$99K current offer price
- \$4-12/Gb
- 100Gb/run
- 1x200 or 2 x 150 in 48hr
- Sequencing by binding
- Accuracy to 0.001% VAF without UMIs



# Sequencing by Binding

SBB advantages: no base modifications, no molecular scarring

Creates optimized steps, incorporates native nucleotides, and produces unmodified DNA

## Sequencing by Synthesis (SBS)

Unblocked  
3' end



Incorporate/  
Interrogate



Cleavage



## Sequencing by Binding (SBB®)

Blocked 3' end



Interrogate



Activate



Incorporate

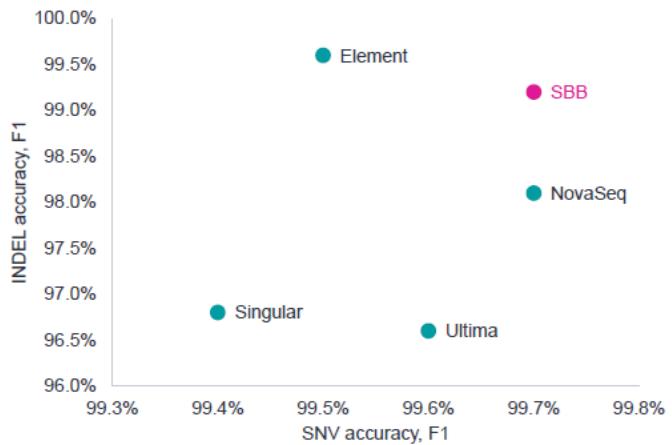




# SNV and Indel accuracy are better than any other platform

## Excellent variant calling performance for SBB

	SBB	NovaSeq	Element	Ultima	Singular	
SNV	Recall	99.6%	99.9%	99.1%	99.6%	99.2%
	Prec.	99.7%	99.5%	99.8%	99.6%	99.7%
	F1	99.7%	99.7%	99.5%	99.6%	99.4%
INDEL	Recall	98.9%	97.9%	99.3%	96.4%	96.4%
	Prec.	99.4%	98.4%	99.8%	96.8%	97.1%
	F1	99.2%	98.1%	99.6%	96.6%	96.8%



- Expecting further improvements through variant caller training

SBB / NVSQ: HG002 GIAB benchmark v4.2, DeepVariant 1.3.0 with the Illumina WGS model  
Element: <https://go.elementbiosciences.com/access-app-note-roche-human-microbial>

PacBio

Ultima: <https://doi.org/10.1101/2022.05.29.493900> HG002 40x coverage

Singular: G4 Human Genome Sequencing Technical Report 1.0. NA12878 40x coverage

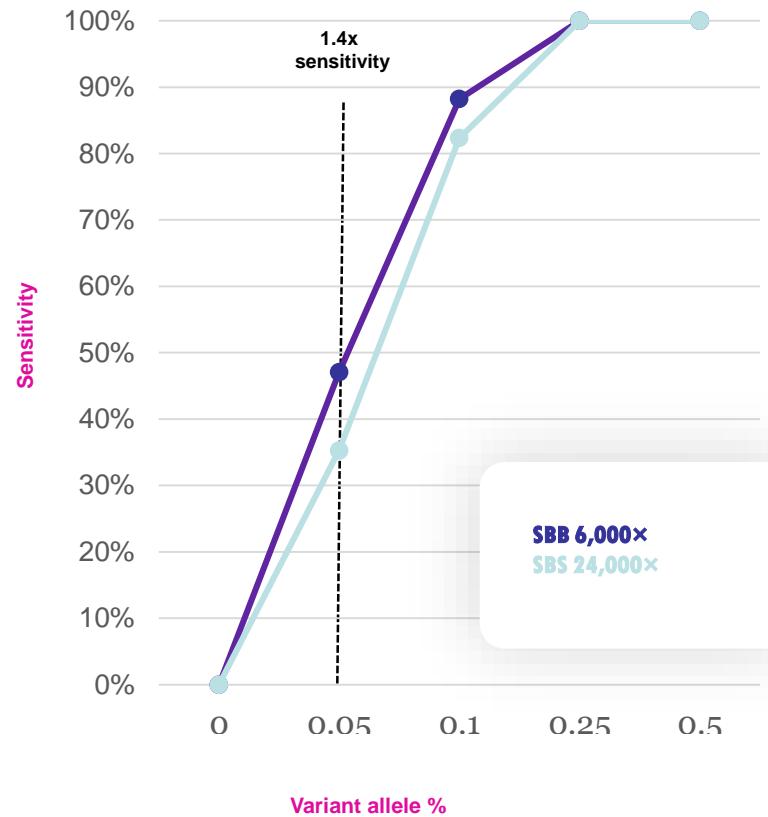
15

## SBB demonstrates >4x improvement in sequencing efficiency



6,000 $\times$  non-UMI SBB sequencing exceeds >24,000 $\times$  SBS UMI sequencing at 0.05% and 0.1%

PacBio



Data based on internal testing. >4x improvement in sequencing efficiency relative to SBS



# Nanopore Sequencing



# Oxford nanopore

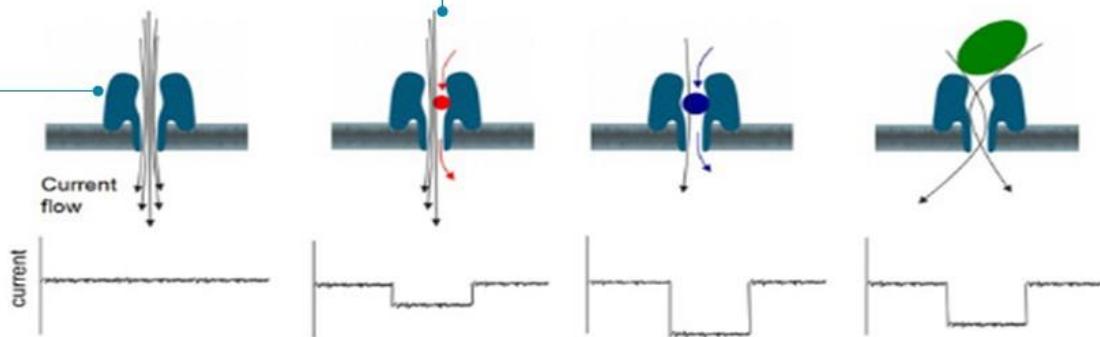
- » Sequences single molecules of DNA as travel through pore
- » Can also identify modified bases and sequence RNA directly
- » Reads typically 5-50kb but some much longer (4Mb record)



## NANOPORE SENSING

1

Nanopore creates hole in membrane  
Current passes through nanopore



2

As analyte passes through or near the nanopore,  
this creates characteristic disruptions in the current

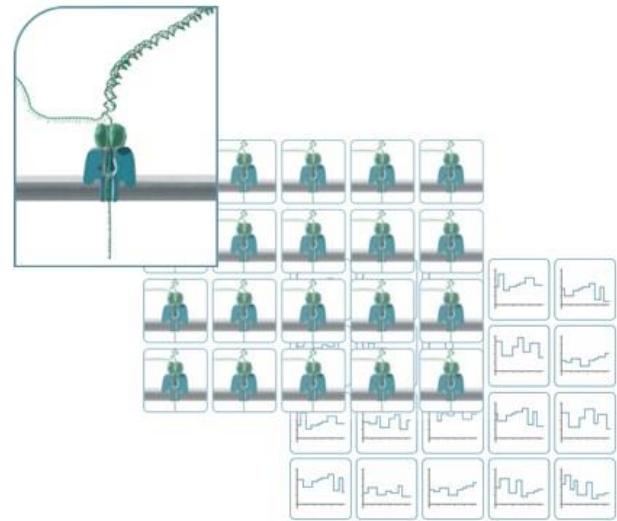
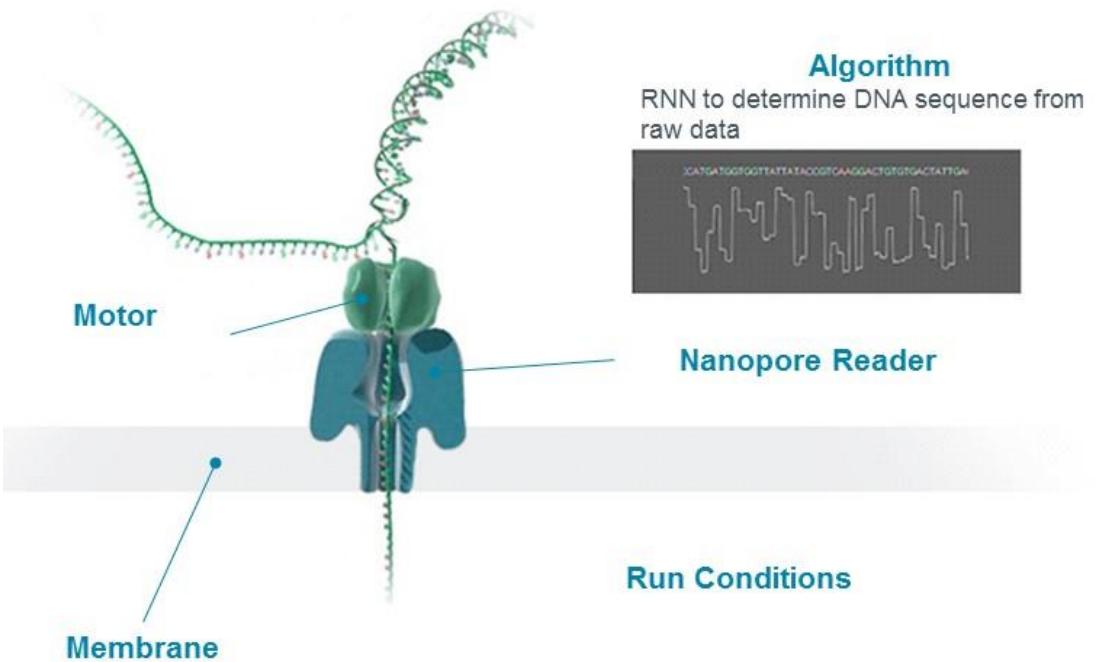
3

Current disruption is interpreted to understand the  
identity of the analyte

An explanation from **Oxford NANOPORE Technologies™**

# NANOPORE DNA SEQUENCING

How does it work?



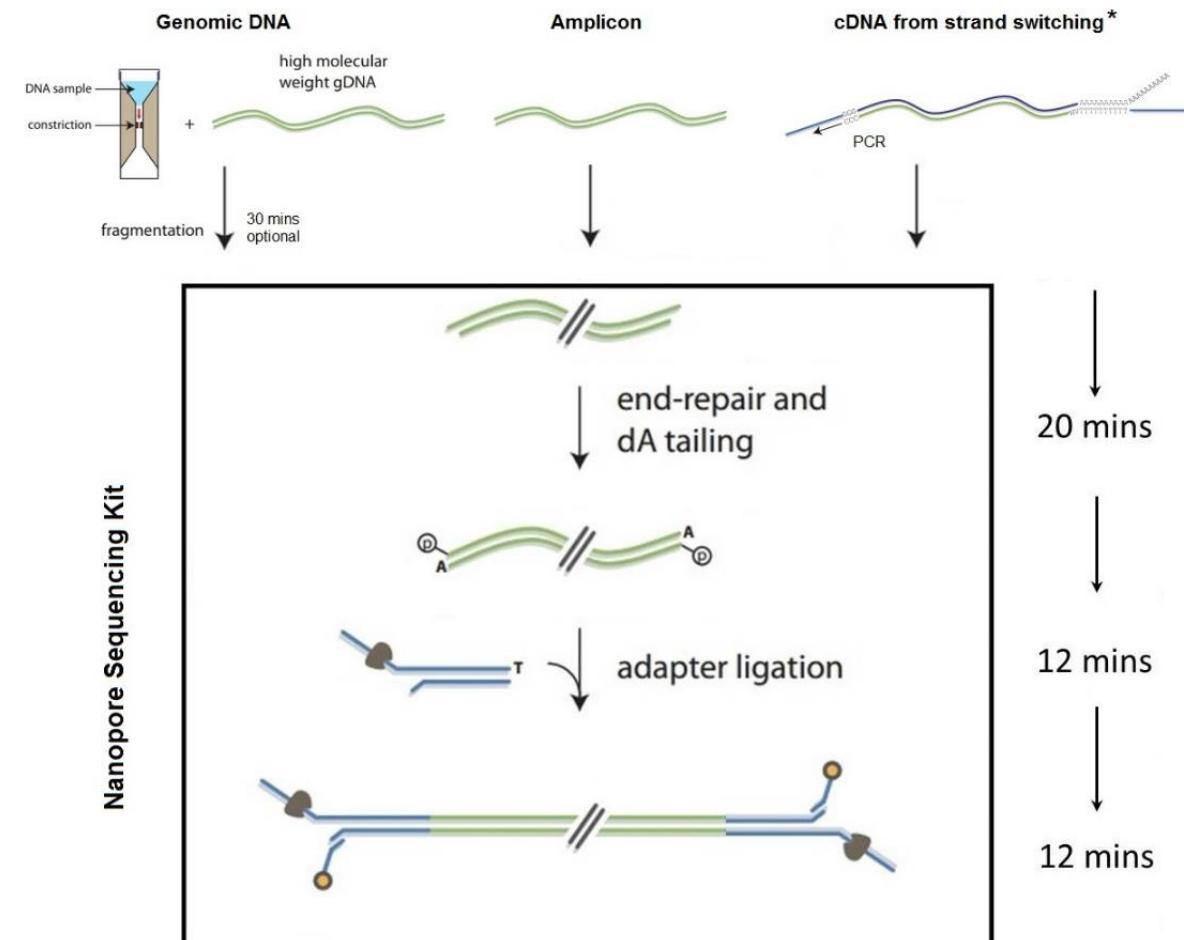
Multiple nanopore sensors arrayed in one device

Operate independently but at the same time

An explanation from  Oxford  
**NANOPORE**  
Technologies™

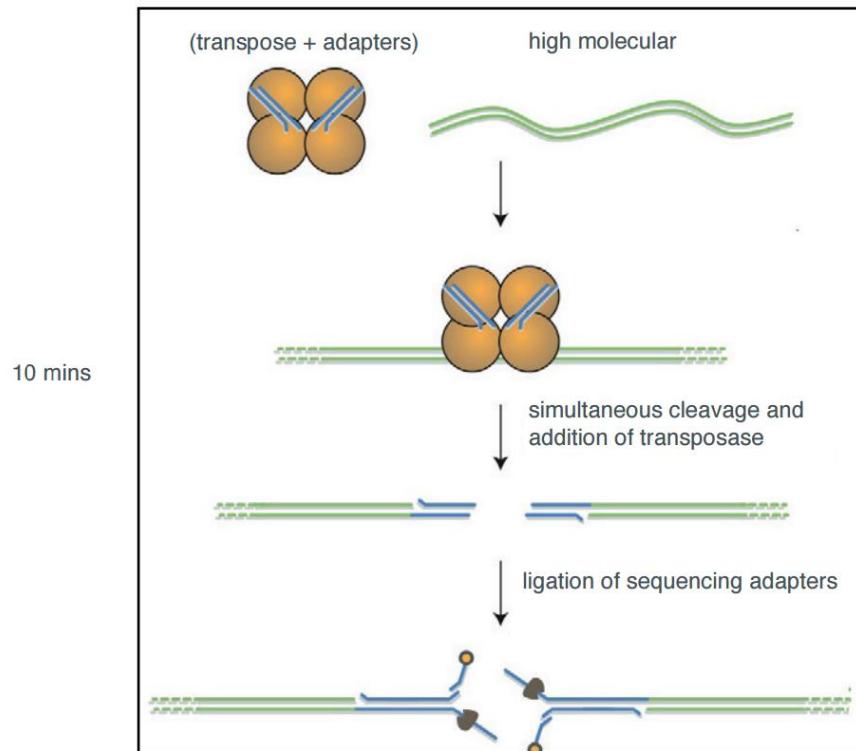
 wellcome  
**sanger**  
institute

# ONT Simplex (1D) Library prep



## RAPID SEQUENCING KIT

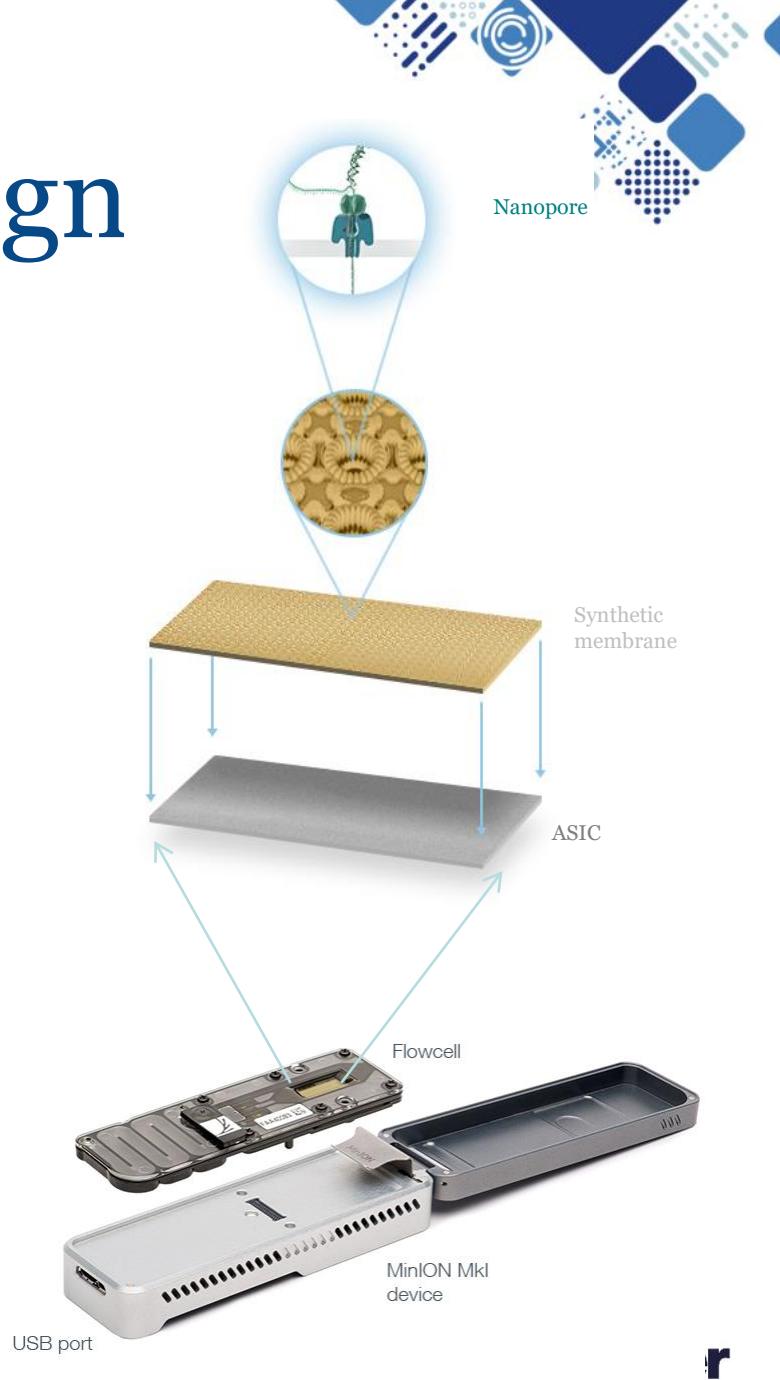
A two-step, 10 minute protocol



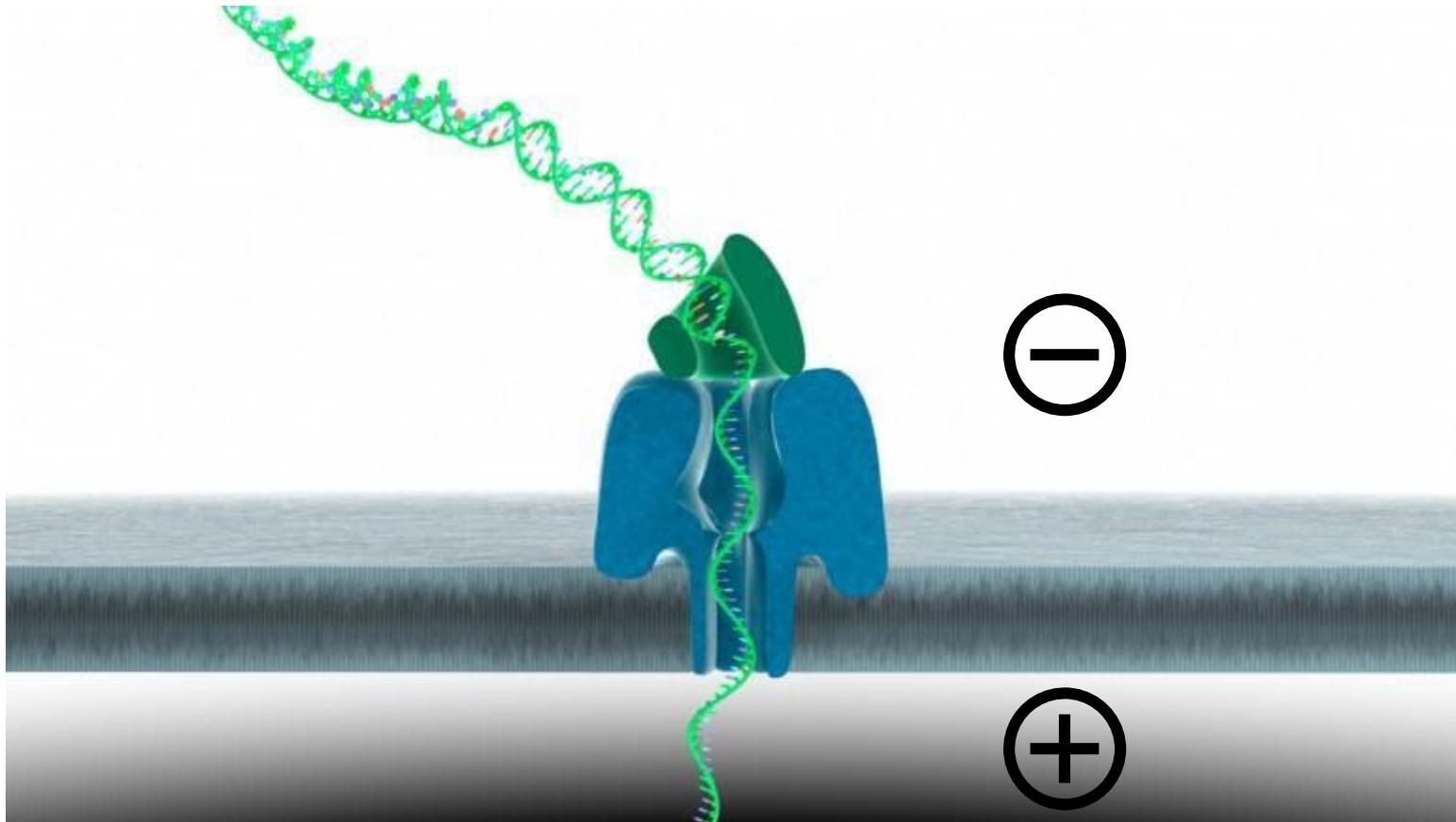
Starting material will be fragmented; recommended starting size >30 kb for genomic DNA

# Flow cell design

- » Application-Specific Integrated Circuits (ASICs), MinION ASIC contains 512 channels
- » Each channel is surrounded by 4 pores & records only 1 at a time
- » 512 pores max recorded at the time



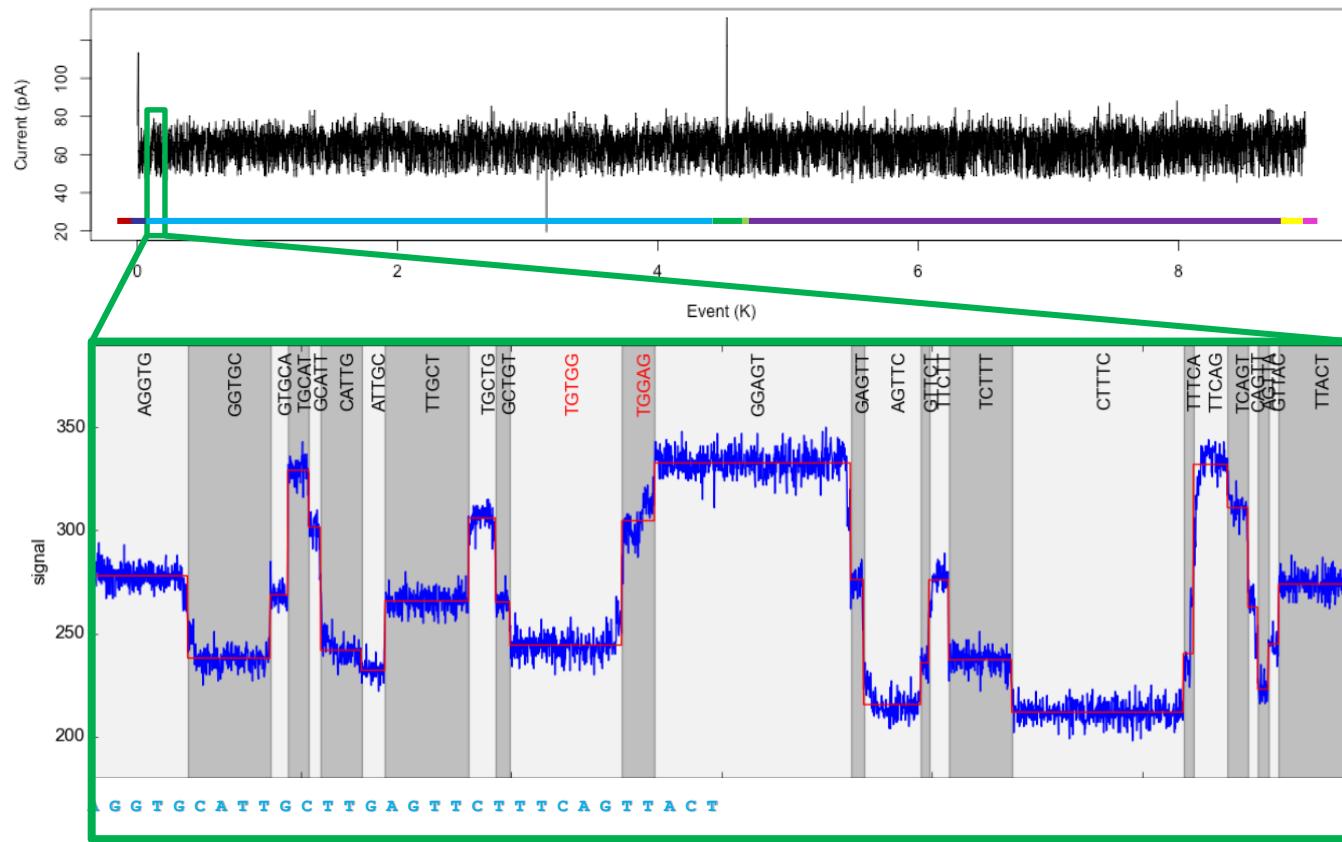
# Oxford Nanopore Technologies



Imagine 5-base words = 1024 current levels

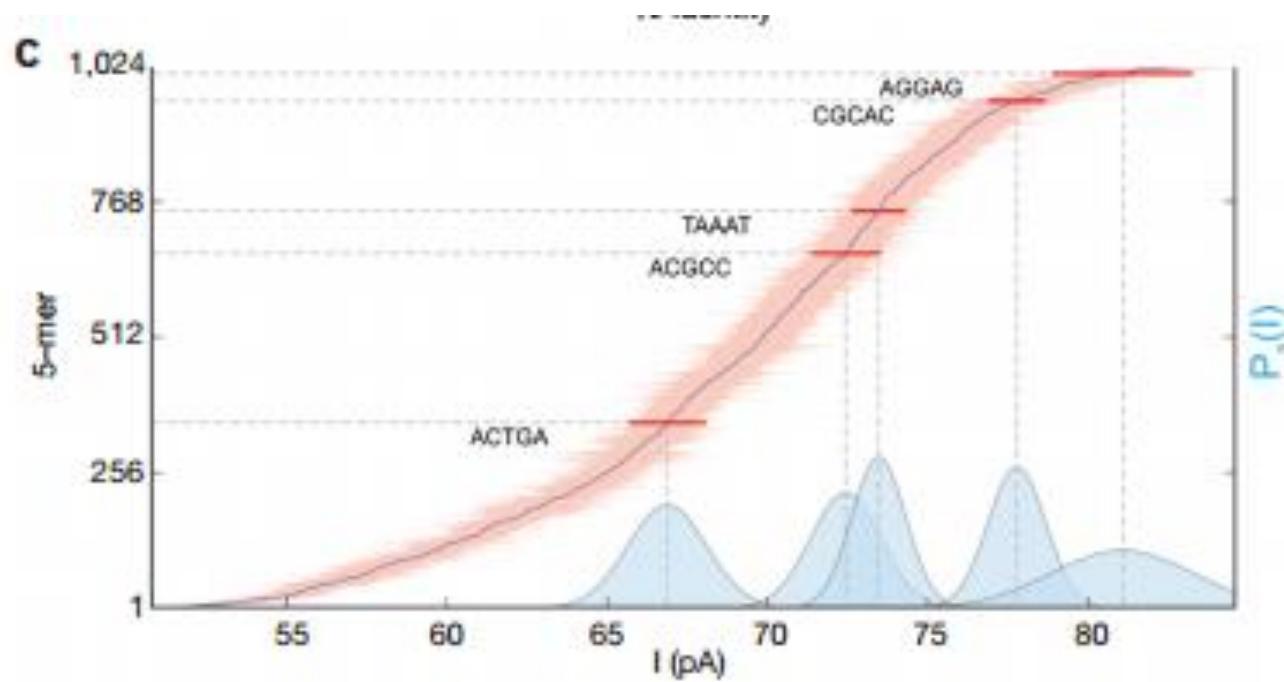


# ONT: The squiggles



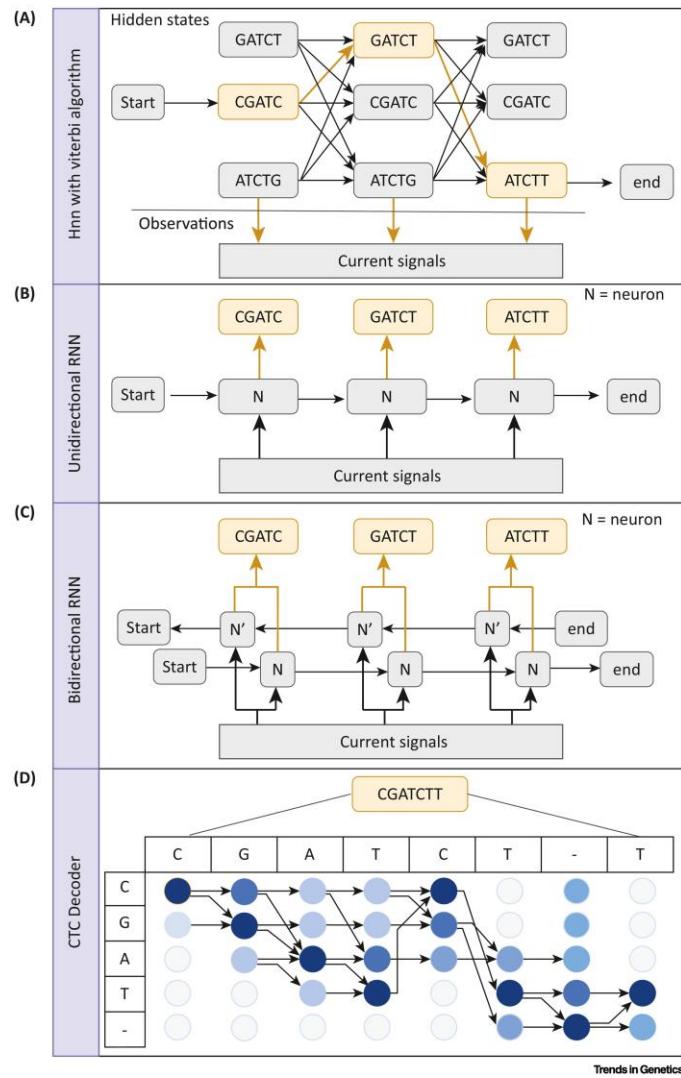
Slide courtesy of David Buck. WTCGH

# Assuming 5mer current signals

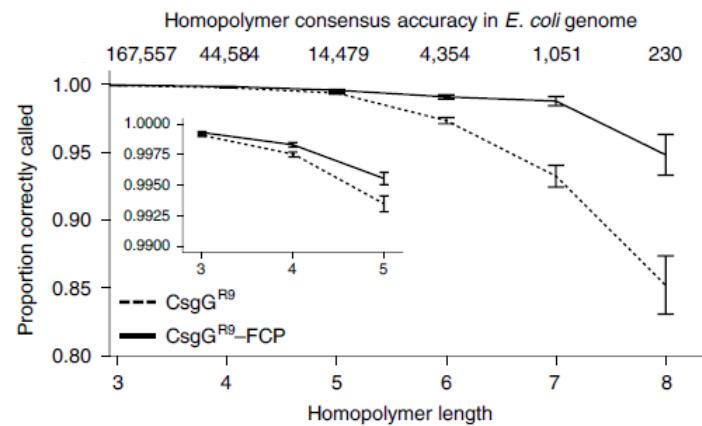
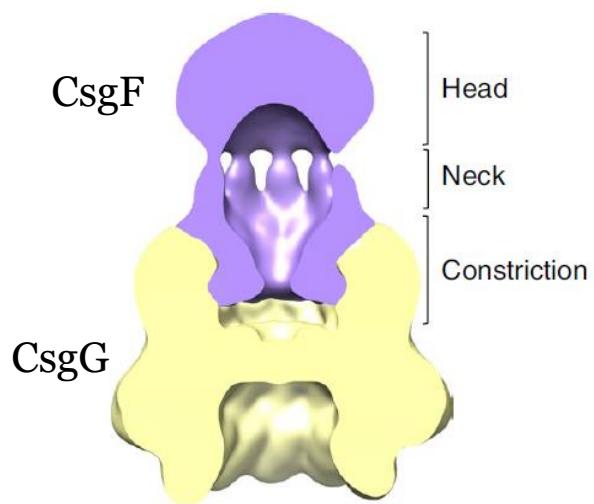


From Szalay & Golovchenko, Nat. Biotech., 2015.

# ONT basecalling with machine learning

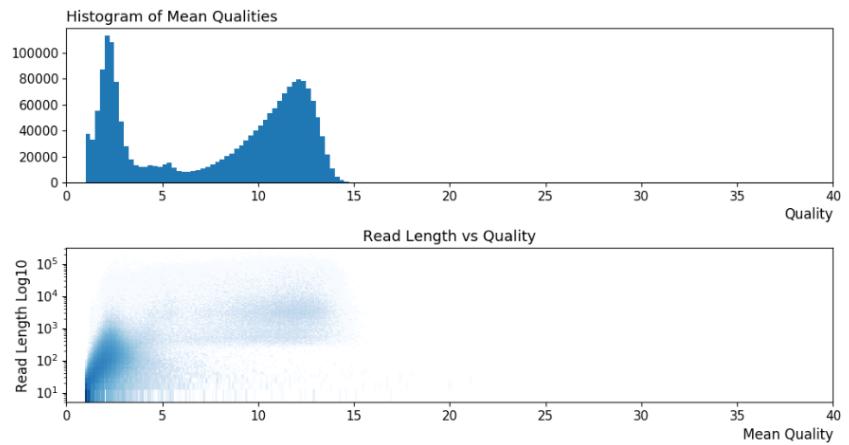
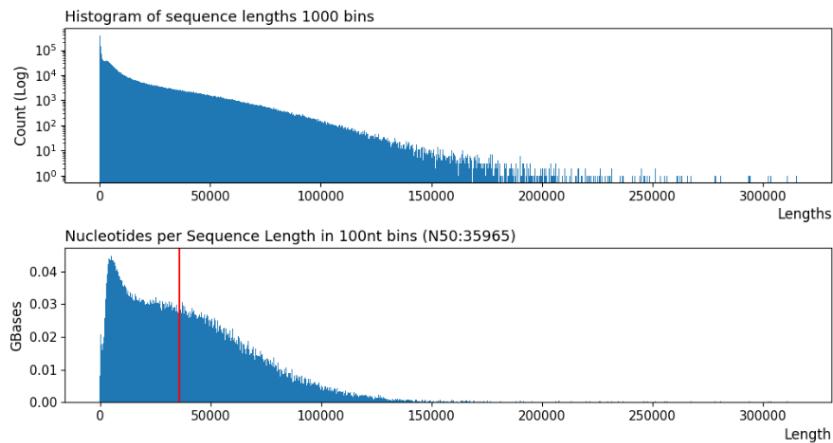


# R10. A dual head nanopore to help resolve homopolymers

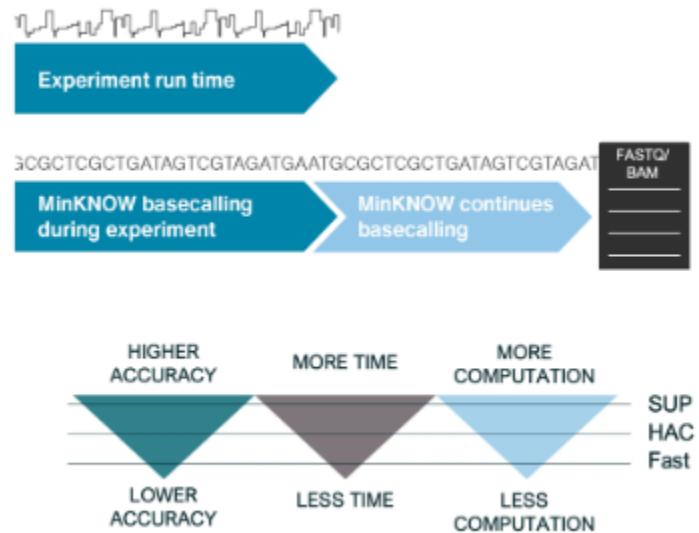


From Van der Verren et al., Nature Biotech 2020

# ONT Sequencing performance graphs



# ONT accuracy





# ONT accuracy

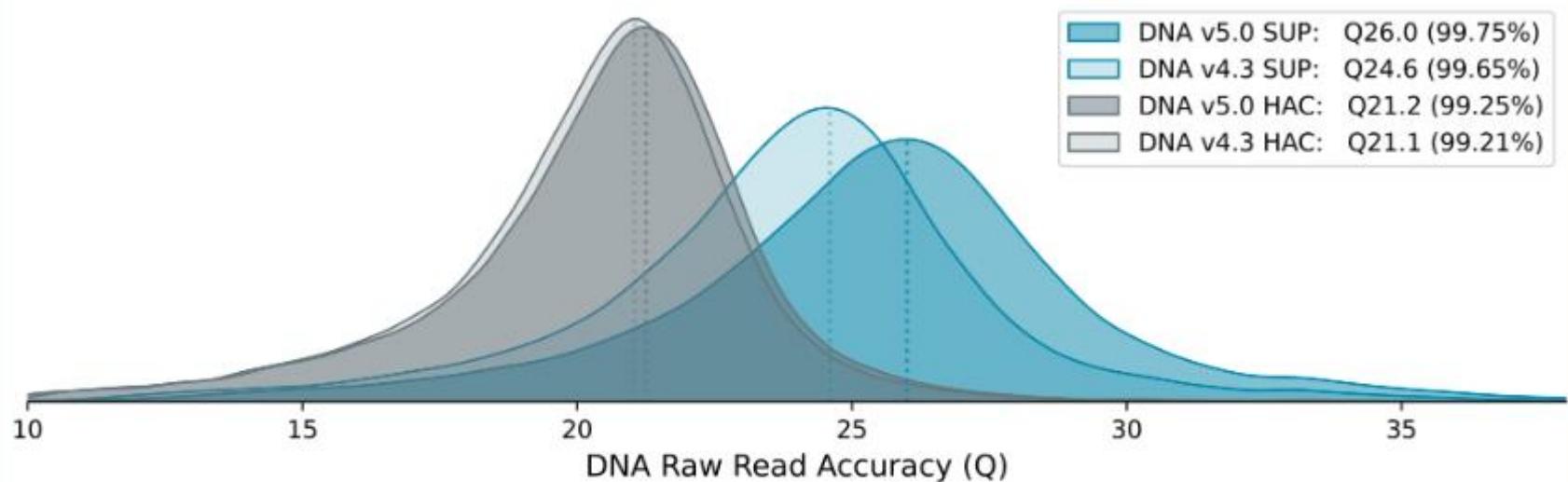
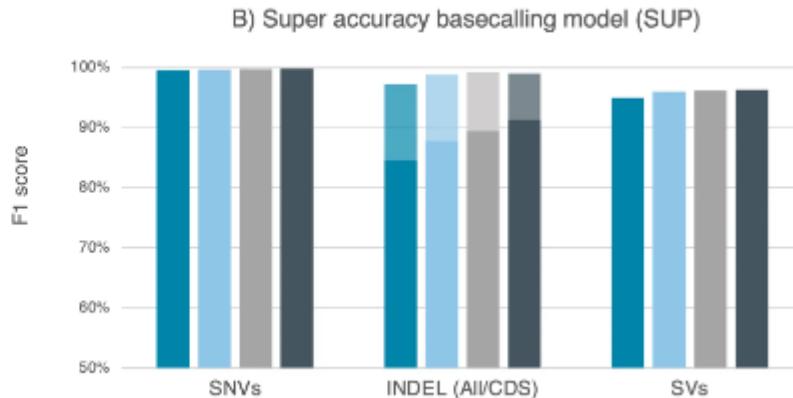
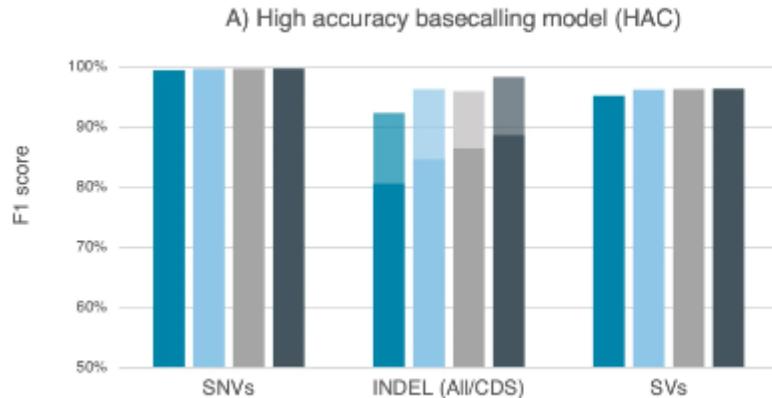


Figure 2. DNA raw read accuracy modal data obtained with Ligation Sequencing Kit V14 (with enzyme E8.2.1) and PromethION R10.4.1 Flow Cells, using nanopore sequencing data for the human genome (HG002 cell lines). Both HAC and SUP models are featured for version 4.3 (currently integrated in MinKNOW) and 5.0

# ONT accuracy



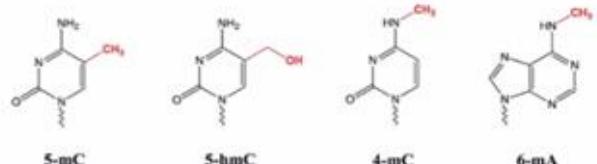
Molecule	Modification	Molecular context	Raw read accuracy (SUP)
DNA	5mC/5hmC	CpG	98.81%
	5mC/5hmC	All	97.87%
	6mA	All	97.52%
	4mC/5mC	All	96.33%
RNA	m6A	DRACH	99.17%
	m6A	All	97.12%
	pseU	All	97.62%

# Direct sequencing of modifications

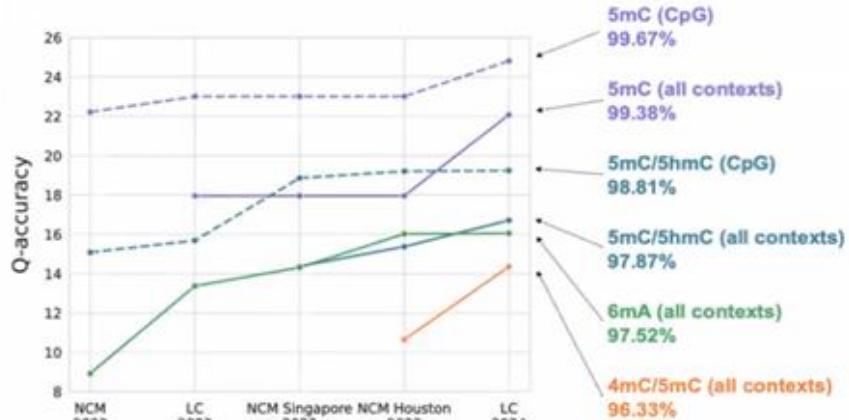
## Broader DNA base modifications offering with improved accuracy



Released



### Evolution of raw read modification accuracy



Fully integrated in MinKNOW

A screenshot of the MinKNOW software interface. It shows 'Basecalling options' and 'Modified bases'. Under 'Modified bases', '5mC & 5hmC CG contexts' is selected, with '5mC & 5hmC CG contexts' and '5mC & 5hmC all contexts' as other options.

Available live or post-run

- 5mC and 5hmC in CpGs
- 5mC and 5hmC in all contexts
- 6mA in all contexts

Offline offering – Dorado v0.7

DNA all contexts: 5mC, 5hmC, 6mA and 4mC (new!)

# minION

## \$1999. Yields upto 48Gb ~\$5/Gb.

### MinION: PORTABLE DNA/RNA SEQUENCING

Sample to scientific insight easily, quickly

Channels\*  
512



An explanation from **Oxford NANOPORE Technologies™**

# Connected minION



Mk Id based on ipad pro



# GridION. For service sector

Nanopore Live LIVE

## GridION X5

Bench top sequencing device

**SEQUENCING**

- 5 individually addressable flow cells
- Based on current MinION flow cell design
- Road map to on board Run Until... and Read Until...

---

**COMPUTE**

- Embedded high performance compute
- Full Real time basecalling and data analysis in the box
- Simple user interface and single ethernet for data transfer

Component	Specification
Size and weight	H200 x W 360x D 360 mm , 10 kg
Power	600 W
Compute spec:	8 TB SSD Storage, 64 GB RAM Latest Gen CPU for OS and orchestration FPGA processor
Pre-loaded software	Linux OS, MinKNOW
Connections	5x USB 3. 1x USB-C 1 x HDMI. 1 x Ethernet



Oxford

\$67000

# Flongle flow cell adapter

\$90 for 2-3Gb



# PromethION

## 24 or 48 flowcells with 12000 pores

### New PromethION P24 / P48 with A100 compute

- Significant performance improvement delivered by A100 technology
- Increased CPU, RAM
- Will ship from May onwards
- Upgrade options available – please contact sales

	P24	P48	A100 tower
2x GV100	4x GV100	4x A100	
CPU	2x Intel 112 cores total	2x Intel 112 cores total	2x Intel 160 cores total
Storage	30 Tbytes	60 Tbytes	60 Tbytes
Connectivity	Dual-port 10 Gbase-T Ethernet Dual-port 10 GB SFP+ fibre		
Memory	384 GB	384 GB	512 GB
Display out	VGA	VGA	MiniDP and VGA



P24



\$225,000

- 192 flow cells
- 42 kits
- 12M SW licence

P48



\$310,000

- 288 flow cells
- 48 kits
- 12M SW licence

### New PromethION P24 / P48 pricing

- Includes the new compute tower
- Brings pricing in line with flow cell price reductions from Jan

~\$7-16/Gb

# PromethION P2 Solo

## PromethION “P2 solo”

Powerful flow cells, small device – up to 550-600Gb/run\*

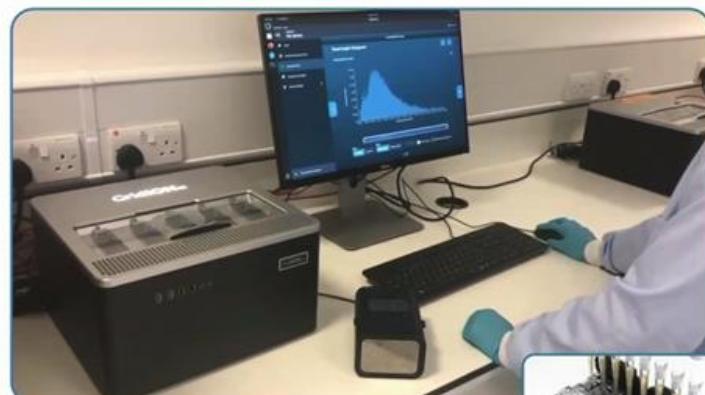
### Two PromethION flowcells, connects to existing compute

- Based on existing flow cell design
- Two individually addressable prom flow cells
- Small versatile instrument to couple to existing GPUs/storage
- Ideal for studying large genomes or transcriptomics
- Easier to place in automation solutions
- Available as a starter pack or CapEx

Pre-order in store today

P2 Solo: \$10,455

Consumable pricing will enable human WGS for under \$1,000



Four chamber flowcells in development



~\$7-16/Gb

# ONT unique applications

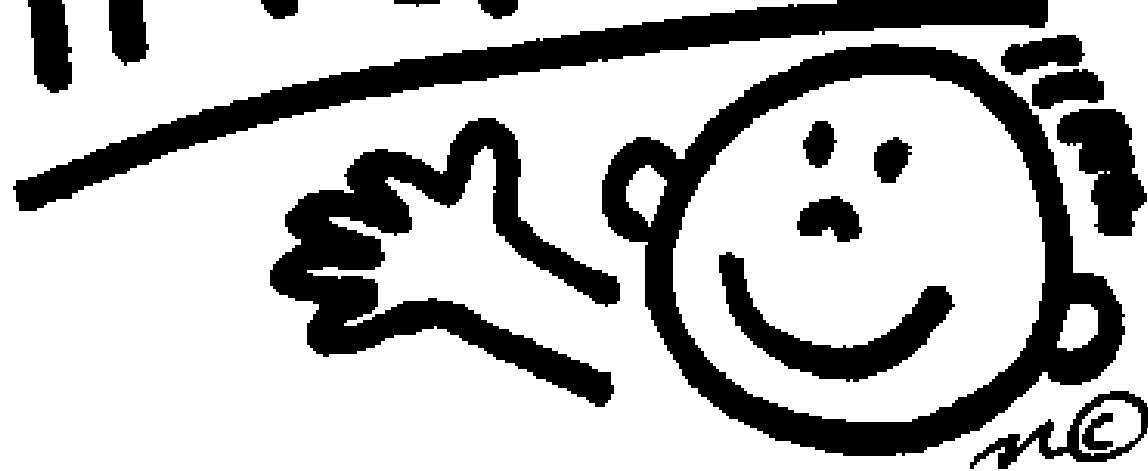
- » Ultra-long reads (>100kb reads possible)
- » “Run until” done. W.I.M.P
- » Selective reads
- » Mobile sequencing
- » Direct RNA sequencing
- » Direct methylation detection over wide range of DNA and RNA modifications

# So. Which to choose?

- » Budget
- » Technical fit
- » Applications
- » Throughput
- » Peers
- » Space



Thanks!





# Any Questions ?

**[mq1@sanger.ac.uk](mailto:mq1@sanger.ac.uk)**