PacBio

The power of **high-accuracy sequencing**

Game-changing sequencing capabilities

AGBT workshop

February 9, 2023

Forward-looking statements

All statements in this presentation (and any accompanying oral presentation) that are not historical of fact are “forward-looking statements” within the meaning of Section 21E of the Securities Exchange Act of 1934, as amended, and the U.S. Private Securities Litigation Reform Act of 1995, including statements relating to future availability, uses, accuracy, advantages, quality or performance of, or benefits or expected benefits of using, PacBio products or technologies, including the Revio and Onso systems; customers’ ability to scale HiFi sequencing through the use of the Revio system; throughput, affordability, coverage, run times, data, density, cost per genome, pricing, number of genomes that can be sequenced per year, and the areas of study that can be explored using the Revio system and SMRT Cells; improvements in yield and accuracy for Revio and Onso; the use of fewer consumables required when using Revio; schedule flexibility and downtime; expected delivery timeframes; expected levels of accuracy, sensitivity, and specificity; expected scalability, flexibility and cost optimization of the Onso and Revio systems; expected compatibility of the Onso system with products currently available for short-read sequencers and ability to support diverse library preparation types, single cell analysis solutions, whole-genome sequencing and other targeted methods; anticipated number of reads per run and cycle flow cells; anticipated pricing; expected ability to process more samples with the same output as other commercially available platforms; expectations regarding data quality and efficiency in connection with genome interrogation; expected use in oncology research and agricultural biotechnology applications; expectations with respect to continue development and delivery timeframes, and other future events. Readers are cautioned not to place undue reliance on these forward-looking statements and any such forward-looking statements are qualified in their entirety by reference to the following cautionary statements. All forward-looking statements speak only as of the date of this press release and are based on current expectations and involve a number of assumptions, risks and uncertainties that could cause the actual results to differ materially from such forward-looking statements, including, among others, challenges inherent in developing, manufacturing, launching, marketing and selling new products, and achieving anticipated new sales; the Onso and Revio systems are entering beta testing, are not yet commercially available, and remain subject to additional development and validation; potential product performance and quality issues and potential delays in development and delivery timelines; assumptions, risks and uncertainties related to the ability to attract new customers and retain and grow sales from existing customers; rapidly changing technologies and extensive competition in genomic sequencing that could make the products PacBio is developing obsolete or non-competitive; supply chain risks; successfully completing development of a product that is not yet commercially available; customers and prospective customers curtailing or suspending activities utilizing our products; the impact of U.S. export restrictions on the shipment of PacBio products to certain countries; and third-party claims alleging infringement of patents and proprietary rights or seeking to invalidate PacBio's patents or proprietary rights. Readers are strongly encouraged to read the full cautionary statements contained in the Company’s filings with the Securities and Exchange Commission, including the risks set forth in the company’s Forms 8-K, 10-K, and 10-Q. The Company disclaims any obligation to update or revise any forward-looking statements.

PacBio

Mark Van Oene

Chief Operating Officer

Our mission

Enabling the promise of genomics to better human health

A year of extraordinary innovation

1Q22

HiFi improvements
in efficiency + usability

2Q22

Methylation calling
on instrument

High-throughput sample setup

Gene editing QC workflow

3Q22

Tandem repeat
genotyping tool (TRGT)

Twist Targeted
Sequencing Panels

4Q22

Revio

Onso

MAS-Seq kitted solution

PacBio

What do long reads enable?

Not all genome sequence data is created equally

Short-read WGS	HiFi WGS
	Structural variation
	Methylation
	Segmental duplications
	Phasing/haplotype
SNPs / small indels	SNPs / small indels
✕ Needs a reference genome	✓ Reference quality
✕ Miss 100s of millions of base pairs	✓ Complete T2T assemblies
✕ Blind to ~400 medically relevant genes in dark regions	✓ All known variant classes

PacBio HiFi delivers a new class of WGS

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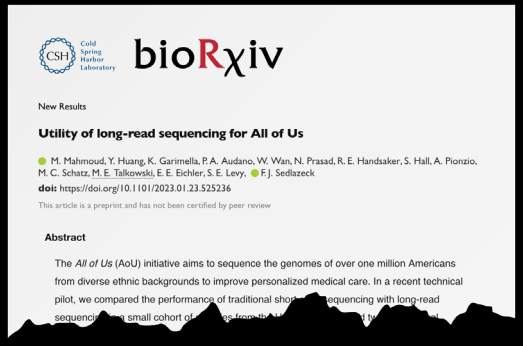
Key findings from the **All of Us initiative**



bioRχiv

New Results

Utility of long-read sequencing for All of Us

M. Mahmoud, Y. Huang, K. Garimella, P. A. Audano, W. Wan, N. Prasad, R. E. Handsaker, S. Hall, A. Pionzio, M. C. Schatz, M. E. Talkowski, E. E. Eichler, S. E. Levy, F. J. Sedlazeck

doi: https://doi.org/10.1101/2023.01.23.525236

This article is a preprint and has not been certified by peer review

Abstract

The *All of Us* (AoU) initiative aims to sequence the genomes of over one million Americans from diverse ethnic backgrounds to improve personalized medical care. In a recent technical pilot, we compared the performance of traditional short-read sequencing with long-read sequencing in a small cohort of

Utility of long-read sequencing for All of Us
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https://www.biorxiv.org/content/10.1101/2023.01.23.525236v1

Compared **variant calling** in HapMap samples

Small variants: "HiFi data achieved the best F-score (99.87)," ONT (98.74), Illumina (99.47)

Structural variants: HiFi(0.93), ONT(0.91), Illumina(0.45)

Analyzed **386 challenging medically relevant genes** in AoU samples

"HiFi outperformed the other technologies in both precision and recall."

> *We should continue developing population-scale cohorts sequenced with long reads only."*

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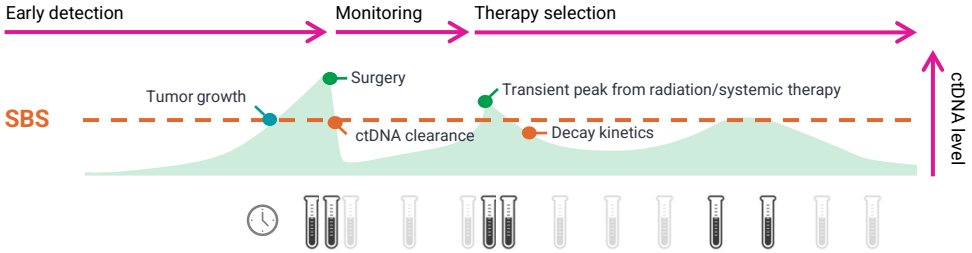
Revio Early Access

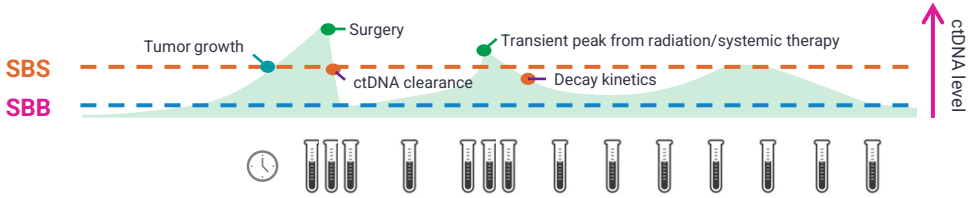
Why does accuracy matter?

- Sequence less, reduce costs
- Sequence more, achieve greater resolution
- Characterize highly complex regions of the genome
- Higher accuracy gives you more confidence in your discoveries

Current sequencing-based liquid biopsy approaches have limited sensitivity



15× increase in accuracy of Onso can mean earlier detection and improved monitoring of cancer



Based on expected specifications for Onso at launch

Onso:
Setting the bar for short-read accuracy

Young Kim

Senior Staff Product Manager, SBB platforms

Onso™

SBB designed to enable extraordinary accuracy for short-read sequencing

Key platform specifications*

400–500 M reads

At launch:

- 200 cycle kit – 1×200 and 2×100
- 300 cycle kit – 2×150

≥90% bases Q40+

Conversion kits for existing short-read ecosystem

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** Expected specs for Onso at launch*

Onso™

Platform on track for H1 shipment

Initial beta feedback demonstrating the value of highly accurate reads

1

BROAD INSTITUTE

"We believe that more accurate reads will be transformative for many genomic applications, including oncology. We are excited to be evaluating the Onso platform with this in mind."

Niall Lennon, PhD,
Senior Director of Translational Genomics, Institute Scientist, Broad Institute

2

CORTEVA™ agriscience

"We've been extremely impressed by Onso's levels of accuracy. This accuracy can open exciting new opportunities to transform agricultural biotechnology, specifically in areas like gene editing specificity analysis."

Gina Zastrow-Hayes,
Biotechnology and GT-Genomics Technology Manager, Corteva Agriscience

3

Weill Cornell Medicine

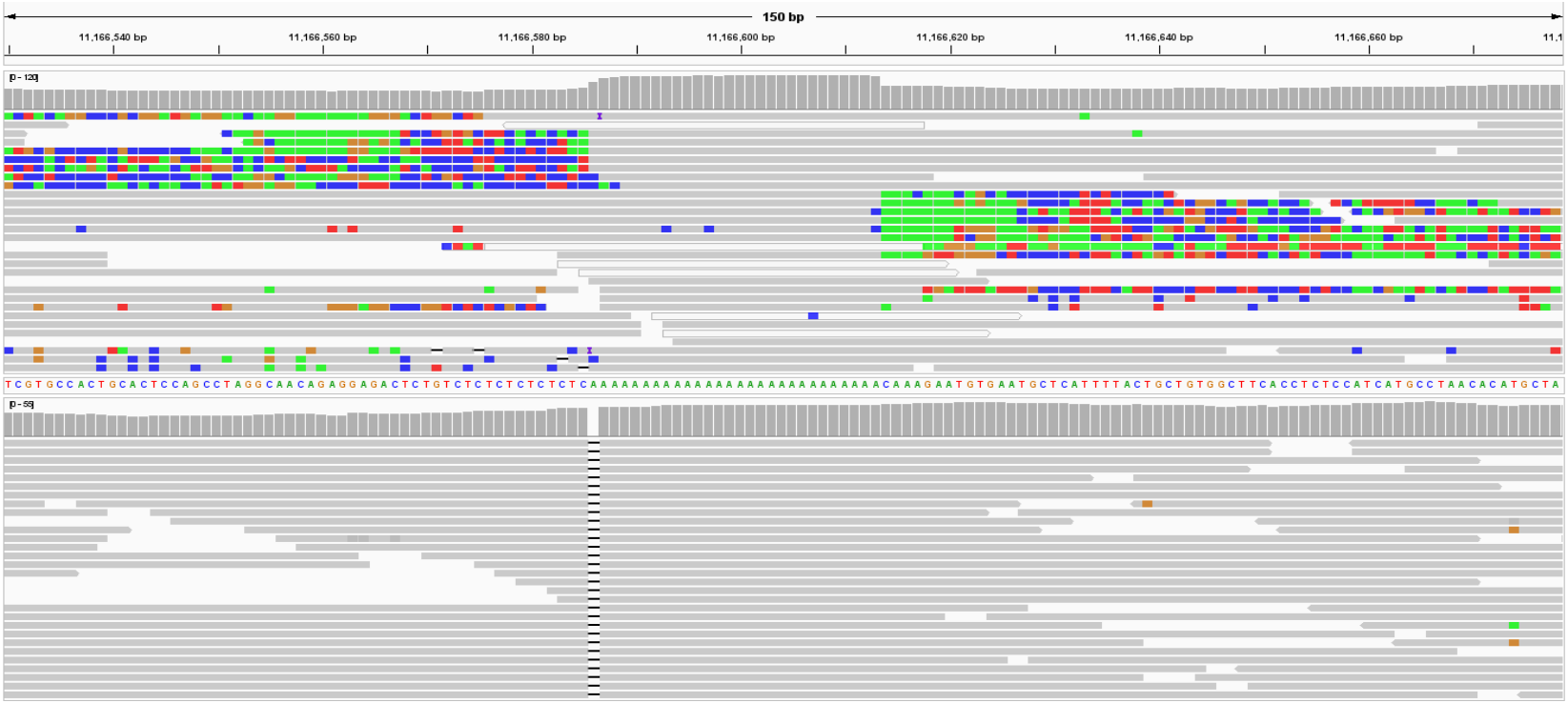
"Some of the first data coming off our new @PacBio Onso sequencer! Starting with a @ZymoResearch titrated microbial mixture and then some @GenomeInABottle controls from @NIST and @US_FDA @maqcsociety data comparisons."

Tweet from Christopher Mason, PhD (@mason_lab)
Professor, Department of Physiology and Biophysics, Weill Cornell Medicine

Sequencing by binding (SBB) chemistry enables highly accurate reads

SBS

SBB



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Data based on internal testing

Onso is designed to deliver a “plug-and-play” experience for short-read NGS customers

Comprehensive library prep and sequencing solutions + connection into existing NGS ecosystem

“Upstream” ecosystem

Sample prep

Automation

Library prep

Sequencing

1°/on-prem analysis

“Downstream” ecosystem

2° analysis

3° analysis

Partnership/license /supply agreements

Core competency

Partnership/license /subscription model

PacBio library prep solutions provide opportunities for higher levels of accuracy

Higher quality achieved with improved library prep kit

Improved raw accuracy performance (Q40 → Q50+)

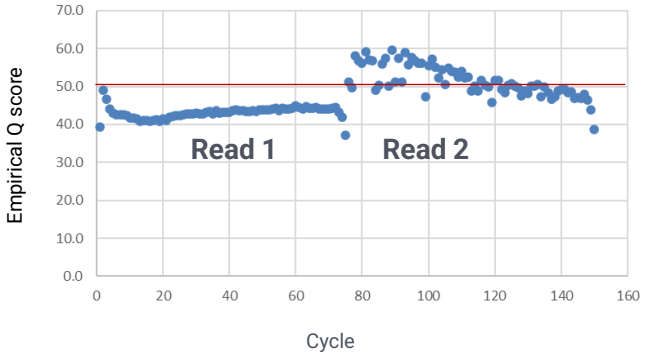
Indexed adapter kit

Negligible index hopping rates (<0.3%)

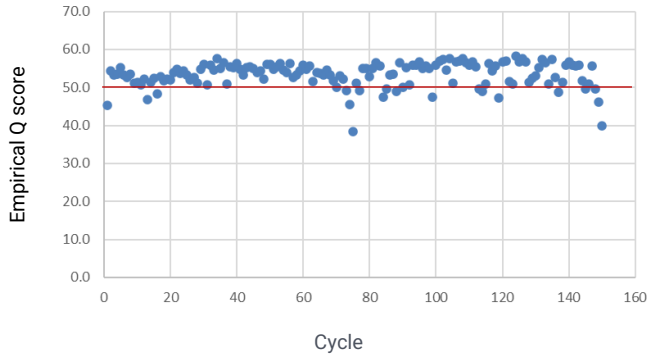
Library conversion kit

Any P5/P7 library conversion | Facilitates direct comparison of SBB vs SBS data

Original library prep



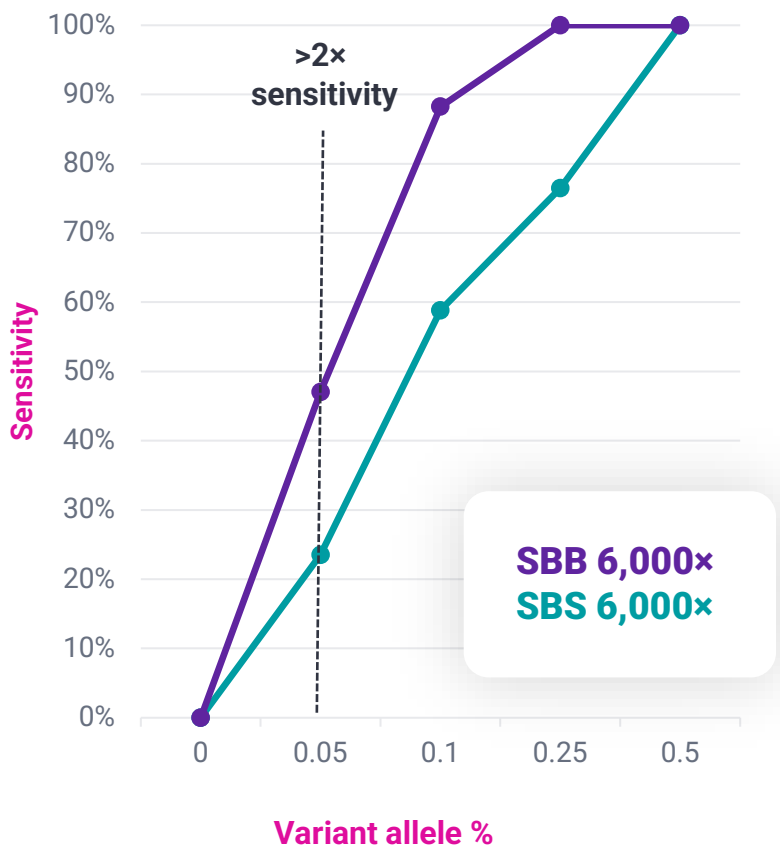
PacBio improved library prep



How does SBB perform with needle-in-a-haystack applications like liquid biopsy and ctDNA?

Superior rare variant detection at equivalent sequence depth

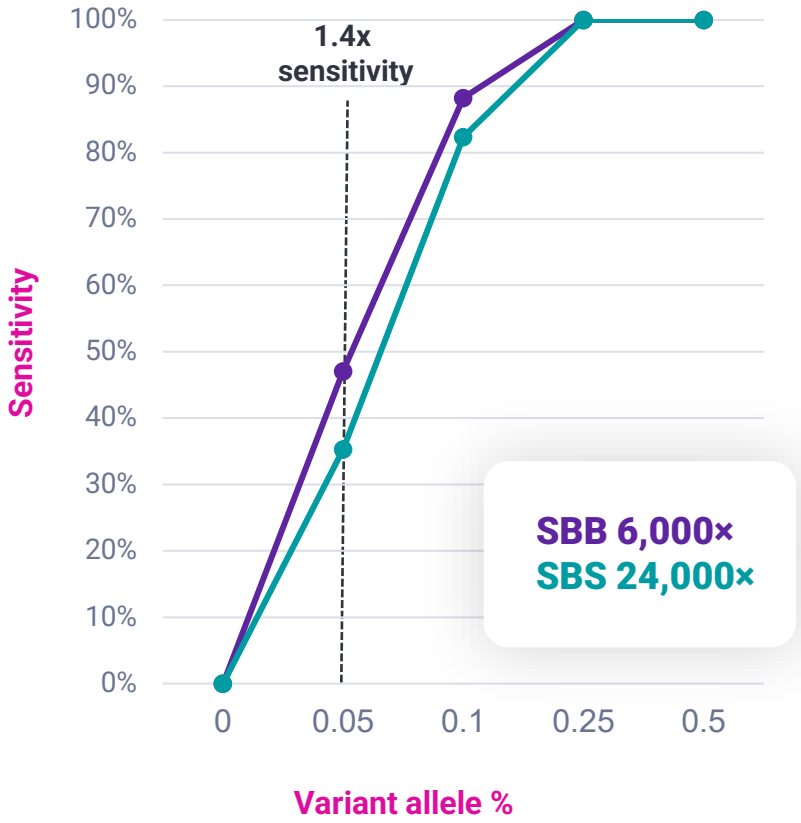
- **Input:** Seraseq® ctDNA *Mutation Mix* v2
- **Library prep:** Agilent XTHS2
- **Enrichment panel:** Agilent *Comprehensive Cancer Panel* (CCP)
- **SBB conversion:** Onso library conversion kit



Data based on internal testing

SBB demonstrates >4× improvement in sequencing efficiency

6,000× non-UMI SBB sequencing **exceeds >24,000× SBS UMI sequencing** at 0.05% and 0.1%



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

SBB demonstrates feasibility for other applications besides needle-in-a-haystack

Initial data from 10k PBMC 3’ single cell RNA-Seq in collaboration with 10x Genomics.

Achieved 99.8% bases Q30 for RNA read!

10k cells

11,419
Estimated Number of Cells

25,926
Mean Reads per Cell

2,865
Median Genes per Cell

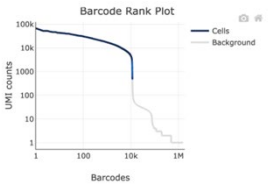
Sequencing

Number of Reads	296,854,227
Number of Short Reads Skipped	0
Valid Barcodes	97.8%
Valid UMIs	99.9%
Sequencing Saturation	50.8%
Q30 Bases in Barcode	99.7%
Q30 Bases in RNA Read	99.8%
Q30 Bases in UMI	99.7%

Mapping

Reads Mapped to Genome	98.6%
Reads Mapped Confidently to Genome	95.8%
Reads Mapped Confidently to Intergenic Regions	3.9%
Reads Mapped Confidently to Intronic Regions	37.6%
Reads Mapped Confidently to Exonic Regions	54.4%
Reads Mapped Confidently to Transcriptome	79.4%
Reads Mapped Antisense to Gene	11.9%

Cells



Estimated Number of Cells	11,419
Fraction Reads in Cells	96.7%
Mean Reads per Cell	25,926
Median UMI Counts per Cell	8,071
Median Genes per Cell	2,865
Total Genes Detected	29,204

Sample

Sample ID	cellranger_FB0032883
Sample Description	
Chemistry	Single Cell 3' v3
Include introns	True
Reference Path	…s/cellranger/refdata-gex-GRCh38-2020-A
Transcriptome	GRCh38-2020-A
Pipeline Version	cellranger-7.1.0

Cell Ranger summary stats and % of cell types identified in line with existing 10k PBMC dataset[1]

1. *10k dataset (reprocessed with Cell Ranger v7.1.0): https://www.10xgenomics.com/resources/datasets/10k-human-pbmcs-3-v3-1-chromium-x-with-intronic-reads-3-1-high.*

Internal beta runs consistently exceed base quality specifications + meet read length targets

Optimization of density in progress and on track to meet launch targets

Metrics	Beta run 1	Beta run 2
Reads	737M	617M
Q score (90%)	46	44
Read length	2×150	2×150
Yield (GB)	111	93
Sequencing run time	48 hrs	48 hrs

Beta run data based on internal QC library used for sequencing

Onso consumable pricing

300 cycle sequencing kit

$ 1,995 / $15 per Gb[1]

120–150 Gb/kit

200 cycle sequencing kit

$ 1,695 / $19 per Gb[1]

80–100 Gb/kit

Sequencing kit contains:

- ☑ 200 or 300 cycle reagent pack
- ☑ Clustering reagent plate
- ☑ Flow cell kit

Onso available for pre-order now!

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1. Expected pricing, subject to change. Consumables available to pre-order end of Q1. Based on 2×150 PE run at yields of 120–150 Gb for 300 cycle kit and 2x100 PE run at yields of 80–100 Gb for 200 cycle kit. Pricing in AMR region only, may vary in different regions. Pricing only includes sequencing consumables and does not include cost of instrument amortization and other operating costs.

How does ~4× less sequencing translate into savings for labs?

Commercial liquid biopsy assay	SBS target seq depth / sample	Onso target seq depth / sample	Gb/sample (SBS)	Gb/sample (Onso)
Vendor panel A	25,000×	6,250×	~75.0 (at max output/sample)	18.8
Vendor panel B	35,000×	8,750×		

SBS cost / sample = $699

Onso cost / sample = $300

~57%

reduction in per sample costs[1]

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1. *Illustrative example for reduced sequencing depth. Actual reduction may vary. Does not include cost of library prep kit / hyb capture reagents / compute costs / labor. Includes instrument amortization costs (4 yr).*

Onso system

- **Designed to enable extraordinary accuracy**
- **Compatible with existing short-read ecosystem**
- **Excels in *needle-in-a-haystack* applications**
- **Reduced sequencing needs vs existing solutions**

Learn more:

List price, USD

$259,000

Revio:
HiFi sequencing at scale

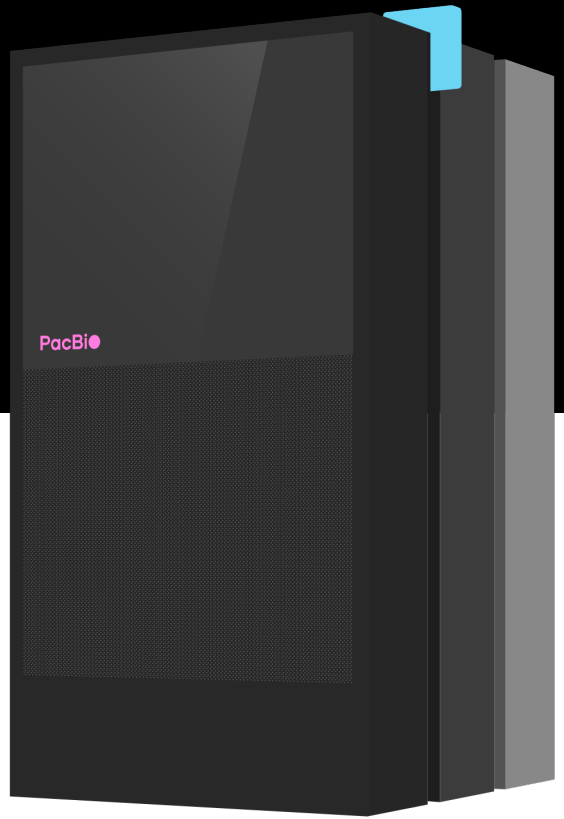
Aaron Wenger

Director, Product Marketing

HiFi sequencing on Revio system

Benefits of HiFi

- **Long reads**
- **High accuracy**
- **Uniform coverage**
- **Native molecules**
- **Epigenetics**



Benefits of Revio

Scale

1,300 human HiFi genomes per year[1]

Ease of use

Simplified consumables and flexible run setup

Compute power

Google DeepConsensus and more on board

Affordability

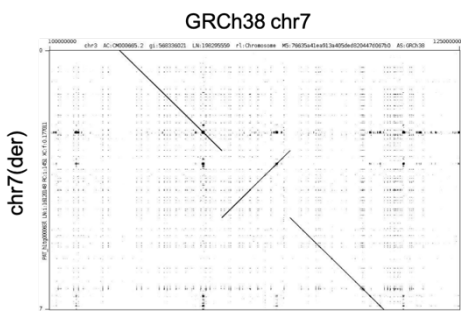
$1,000 per human HiFi genome[2]

1. *Scale based on expected specifications of Revio system at launch.*
2. *USD list price for sequencing consumables for a 30X human genome, assuming 90 Gb yield per SMRT Cell*

“Once impossible becomes possible with HiFi”

Jeremy Schmutz,
HudsonAlpha + Joint Genome Institute

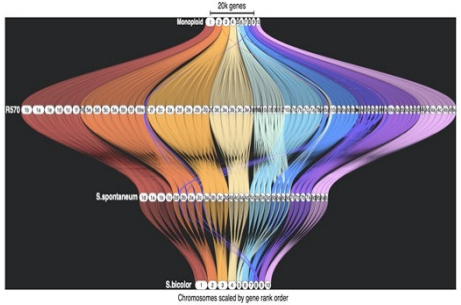
4 Mb pathogenic inversion
Affects gene *ZBTB20*



w/ Greg Cooper,
HudsonAlpha

Sugarcane genome
11,000 more genes with HiFi

Tech	Size	Contig N50
Illumina	5.0 Gb	4.4 kb
PacBio CLR	7.4 Gb	482 kb
PacBio HiFi	9.6 Gb	10 Mb



w/ Karen Aitken,
CSIRO

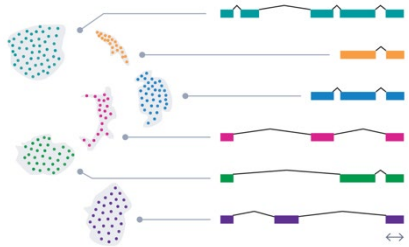
Many scientific firsts have been achieved with HiFi sequencing

Genomes

First telomere-to-telomere assemblies

Nurk et al. (2022) *Science*. 376(6588):44-53.

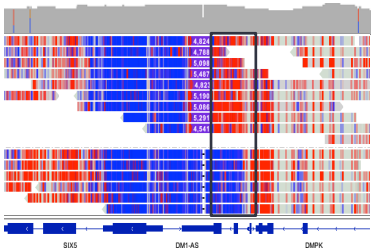
Transcriptomes



First single-cell isoform catalogs

Al'Khafaji et al. (2021). *bioRxiv*.

Epigenomes



First directly phased methylomes

Cheung et al. (2022). *medRxiv*.

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Revio is designed for HiFi sequencing at scale

1,300 human genomes per year[1]

100 M
ZMW / run

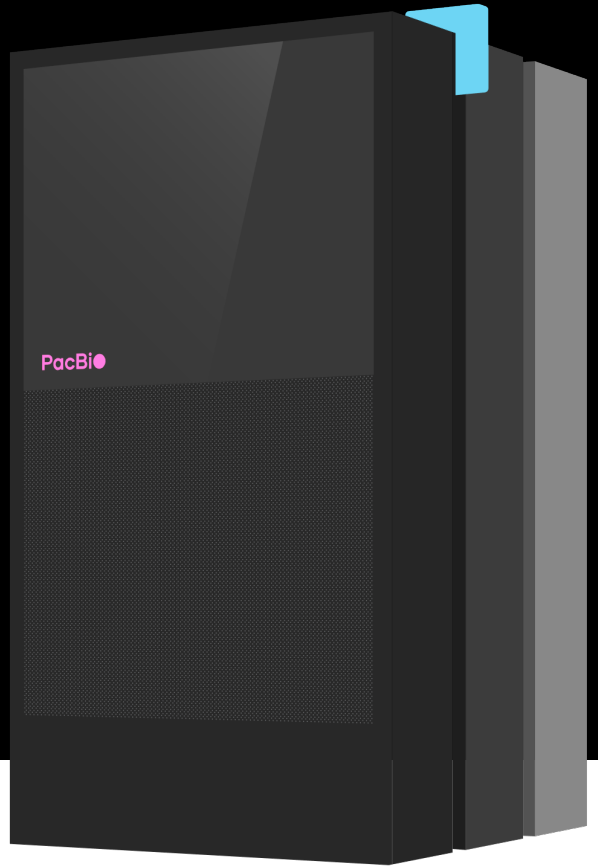
360 Gb
HiFi yield per run

24 hour
Sequencing time

15–18 kb
Read length

5mC
DNA methylation

90% Q30+
Base quality



1. *Throughput based on expected specifications of Revio system at launch.*

Revio: fundamentally powered by the **new 25M SMRT Cell**

Maintained for operational efficiency

- Keeping similar chip size
- Optimized for throughput + manufacturability

Innovated for improved performance

- Increased ZMWs and density
- Improved loading and illumination uniformity
- Capped flowcell design w/ reduced reagent volumes
- Nitrogen-free sequencing

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Four independent stages

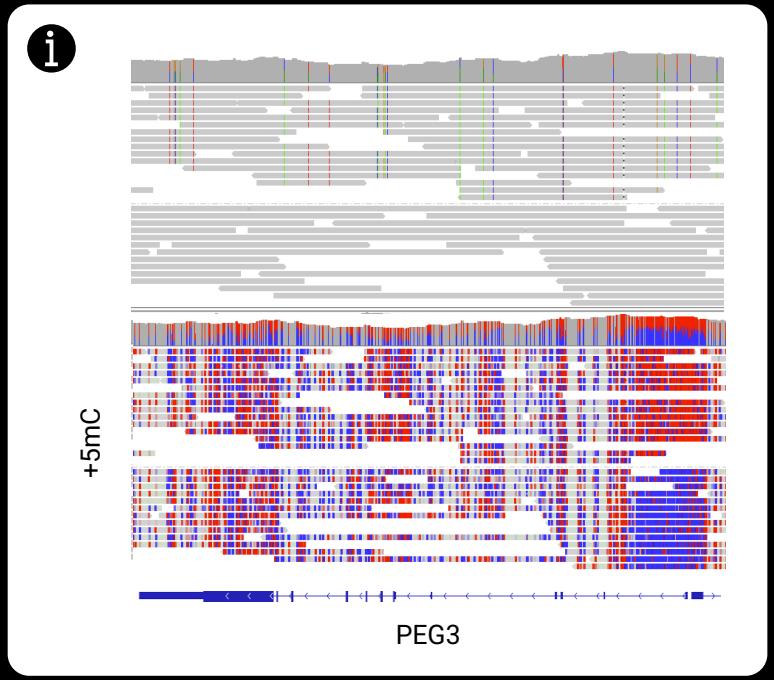
100M ZMW =	25M ZMW	25M ZMW	25M ZMW	25M ZMW
	Normal	Tumor	Tumor	MAS-Seq
360 Gb =	90 Gb	90 Gb	90 Gb	90 Gb

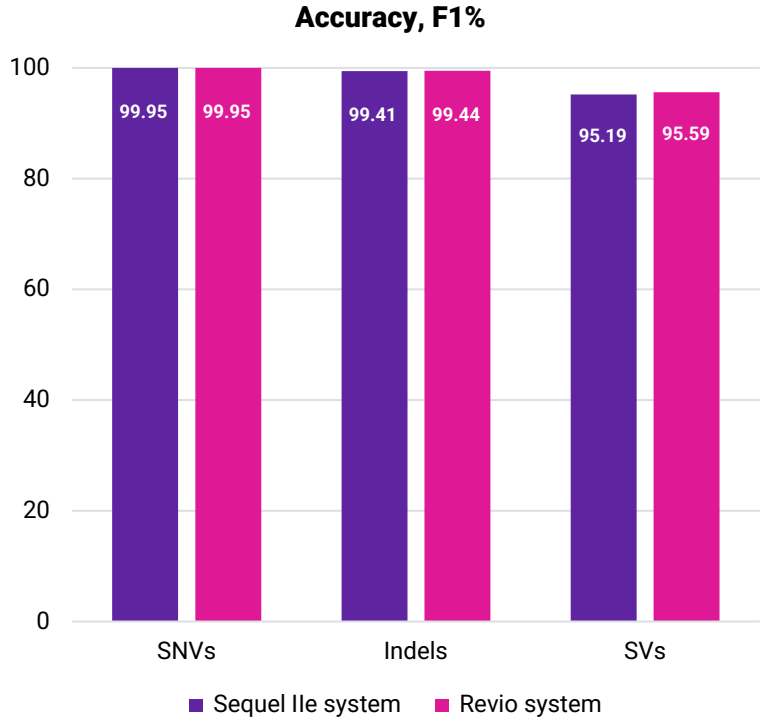
Revio sequencing performance[1,2]

	Sample	Read length	HiFi yield 1 Revio SMRT Cell	Base quality Q30+
	Specification		**90 Gb**	**90%**
	HG002	15.5 kb	101 Gb	93%
	HG002	15.3 kb	92 Gb	91%
	HG002	15.2 kb	87 Gb	90%
	HG003	13.5 kb	99 Gb	92%
	HG003	17.4 kb	94 Gb	92%
	HG004	14.8 kb	103 Gb	92%
	HG004	16.3 kb	96 Gb	92%
	Mouse + ladybug	14.8 kb	95 Gb	90%
	Mouse + ladybug	15.0 kb	94 Gb	91%
	Oak + mistletoe	15.1 kb	100 Gb	92%
	Oak + mistletoe	15.6 kb	95 Gb	92%
	Average	**15.1 kb**	**96 Gb**	**91%**

1. *Performance from internal beta testing*
2. *Example datasets are available at http://pacb.com/data*
3. *Mouse, ladybug, oak, and mistletoe samples provided by Tree of Life Programme at the Sanger Institute*

Revio variant calling and methylation performance





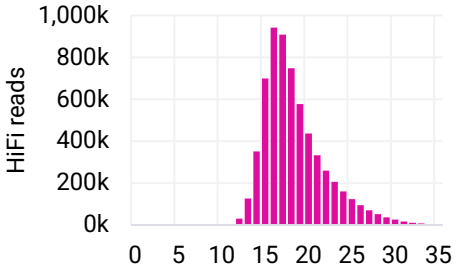
HG002 at maternally imprinted PEG3 locus
Data for internal beta testing https://downloads.pacbcloud.com/public/revio/2022Q4/HG002-rep3/analysis/HG002.m84005_220827_014912_s1.GRCh38.bam
Variant calling measured against Genome in a Bottle benchmarks, HG002

Revio performance: human 20 kb library[1]

118 Gb yield from 1 Revio SMRT Cell

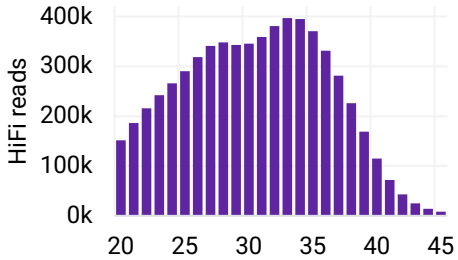
Read length

18.9 kb mean



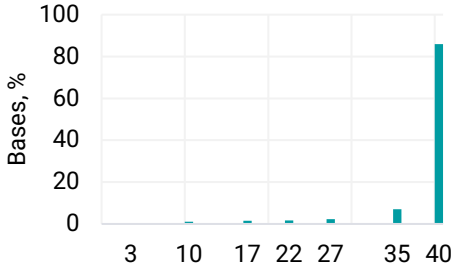
Read quality

Q31 median



Base quality

93% Q30+, 85% Q40+



Genome assembly	Total	N50	N90	Max
HG002 hap1	3.0 Gb	40.1 Mb	4.4 Mb	126.5 Mb
HG002 hap2	3.0 Gb	56.5 Mb	4.6 Mb	168.9 Mb

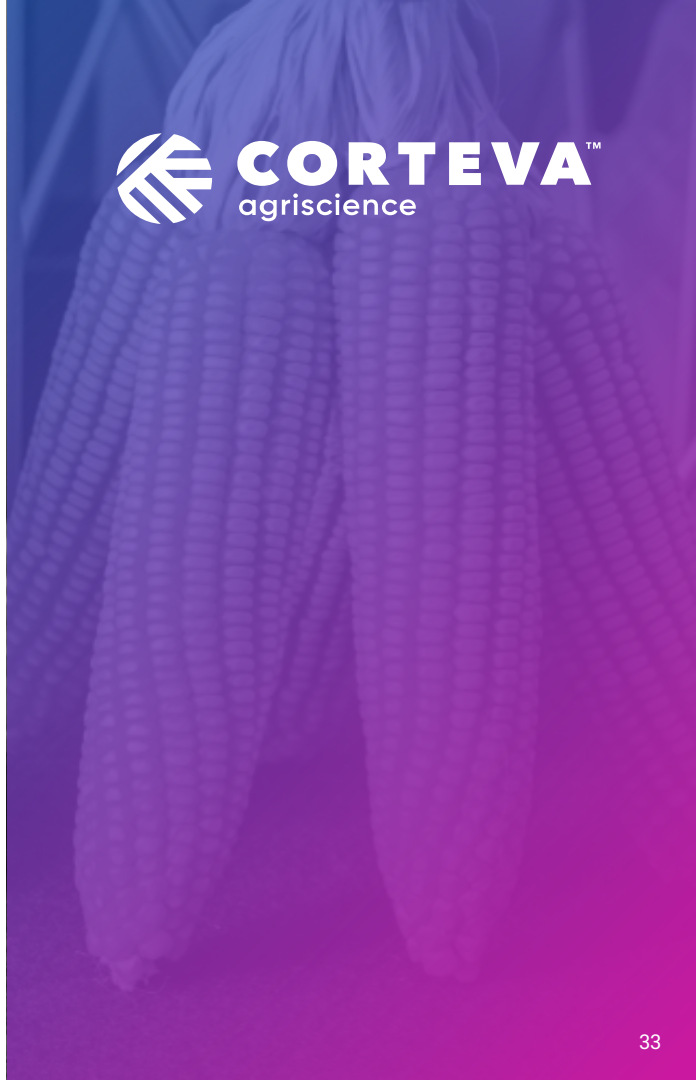
1. Data from internal beta testing. Available at s3://human-pangenomics/submissions/80d00e88-7a92-46d8-88c7-48f1486e11ed--HG002_PACBIO_REVIO/
Sample: HG002, Extraction: Qiagen MagAttract, Shearing: Megaruptor, Library: SMRTbell prep kit 3.0, Size selection: Sage ELF with 1-18 kb cassette
Sequencing: 24-hour movie on Revio system
Analysis: Assembly with hifiasm 0.18.5-r500 with default parameters

Revio performance: Maize B73

71 Gb yield from 1 Revio SMRT Cell[1,2]

> *“This B73 Revio assembly is our best maize assembly to date — that includes over 160 maize genomes.”*

Metric[2]	Value
Total	2.2 Gb
N50	162.5 Mb
Contigs	59



1. *Sequencing from internal beta testing. Sequencing: 24-hour movie on Revio system*
2. *Analysis: Assembly with hifiasm 0.18.5-r500 with default parameters*

Revio performance: American persimmon

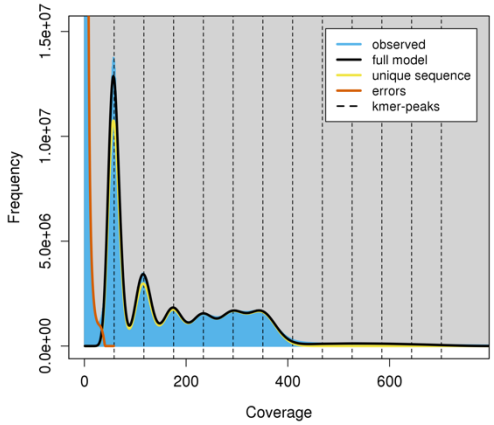
267 Gb yield from 3 Revio SMRT Cells

HUDSONALPHA
INSTITUTE FOR BIOTECHNOLOGY

Jeremy Schmutz, Scott Brainard

Hexaploid

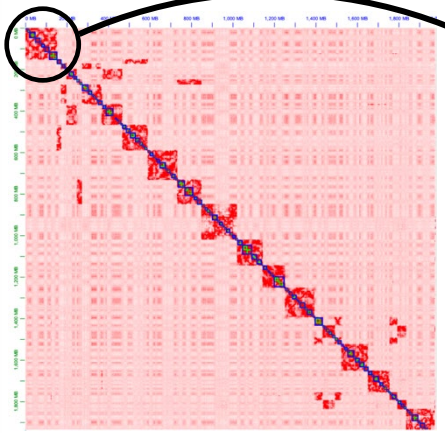
6 distinct kmer peaks

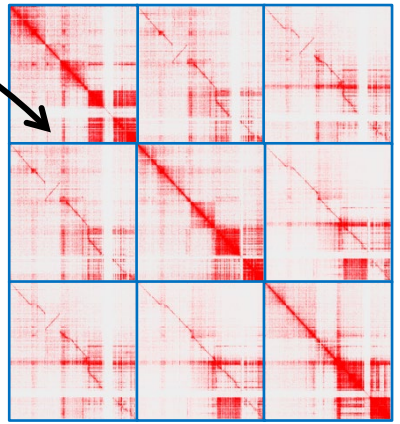


Contiguous + accurate assembly

Haplotype 1: 2.1 Gb with 12.7 Mb contig N50

1 in 200 kb indel error rate





"Early golden" persimmon sample provided by Scott Brainard, Savanna Institute
Sequencing from internal beta testing.
Assembly by Jeremy Schmutz, Jerry Jenkins, and Rachel Walstead of HudsonAlpha Institute of Biotechnology

PacBio

Revio performance: Metagenomics

135 Gb yield from 2 Revio SMRT Cells[1]

	Yield	Reads	Length	Median QV
Run 1	68.3 Gb	8.9 M	7.7 kb	Q46
Run 2	67.4 Gb	8.7 M	7.7 kb	Q46

ZymoBIOMICS
Fecal Reference

1. *Sequencing from internal beta testing. Sequencing: 24-hour movie on Revio system*

Revio is designed to enable genome projects and clinical researchers to **sequence thousands of HiFi genomes**

MBRU

Multi-unit Revio order to **propel genomic medicine discovery in rare disease and cancer** in Dubai

wellcome sanger institute

Multi-unit Revio order to significantly ramp *Darwin Tree of Life* program and **increase long reads in human applications** such as single-cell transcriptomics + variant detection

Radboudumc

The program has already solved several genetic mysteries using PacBio[1] and **with Revio expects to ramp from 100s to 1,000s of genomes**

10.1002/path.5755; 10.1016/j.xhgg.2021.100046; 10.1053/j.gastro.2022.08.041

Revio system

Throughput

360 Gb
HiFi yield per run

24 hour
Sequencing time

1,300 genomes / year
30× human genome equivalent

Learn more:

List price, USD

$779,000

$995* /genome

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

PacBio

2022

1Q22

Improve workflow

2Q22

Methylation calling

HT sample setup

Gene editing QC

3Q22

TRGT

Twist targeted panels

4Q22

Revio

Onso

MAS-Seq kit $475 / sample[1]

PacBio

MAS-Seq for single-cell isoform sequencing

Peripheral blood mononuclear cells (PBMC) MAS-Seq[2]

Learn more at:

	Sequel II	Revio
Reads	40,131,832	**110,127,016**
Reads with valid barcodes	94%	96%
Estimated number of cells	3,966	8,822
Reads per cell, mean	8,708	11,352
UMIs per cell, median	4,821	5,861
Genes per cell, median	704	938
Transcripts per cell, median	818	1,110

1. *USD list price per sample for MAS-Seq kit 102-407-900. 8 reactions per kit.*
2. *https://downloads.pacbcloud.com/public/dataset/MAS-Seq/*

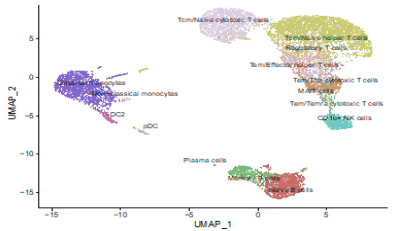
Extending MAS-Seq technology to bulk Iso-Seq and 16S rRNA

New MAS-Seq kits to support bulk Iso-Seq and 16S rRNA amplicons on the Sequel II/IIe and Revio systems

MAS-Seq for 10x Single Cell

- ✓ Launched Oct 2022
- ✓ 16-fold concatenation
- ✓ 40 million reads (Sequel II/IIe)
- ✓ 80 million reads (Revio)

Identify cell type-specific isoform expression

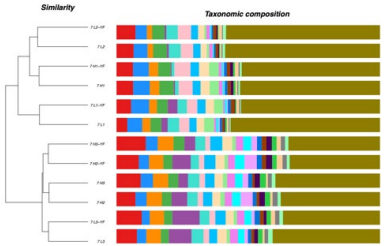


Coming H2 '23

MAS-Seq for 16S rRNA

- → Expected H2 2023
- → 12-fold concatenation
- → Support on Sequel II/IIe and Revio with multiplexing

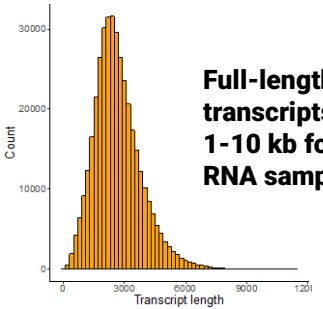
Taxonomic profiling for human, animal, and environmental samples



Coming H2 '23

MAS-Seq for bulk Iso-Seq

- → Expected H2 2023
- → 6-fold concatenation
- → Support on Sequel II/IIe and Revio with multiplexing



Full-length transcripts from 1-10 kb for bulk RNA samples

2022

1Q22

Improve workflow

2Q22

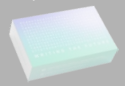
Methylation calling

HT sample setup

Gene editing QC

3Q22

TRGT



Twist targeted panels

4Q22

Revio

Onso

MAS-Seq kit
$475 / sample[1]

PacBio

2023

1Q23

Ship Revio systems

2Q23

Ship Onso systems

2H23

Extended MAS-Seq kits

Expected ship dates

2023 Discoveries Roadshow

Connect. Discover. Be inspired.

We're coming to you:

30+ cities

March – June

Bringing you:

- Scientific talks
- Networking opportunities
- Technology updates

Scan **QR code** and **register** to save your seat!

Thank you!

PacBio

Best of both worlds

$849k for bundle[1] of Revio + Onso

1. *USD list price for instrument bundle*