Nabsys

OhmX Analyzer
High Resolution Structural Variant Detection

Sal Mazza, Director of Sales

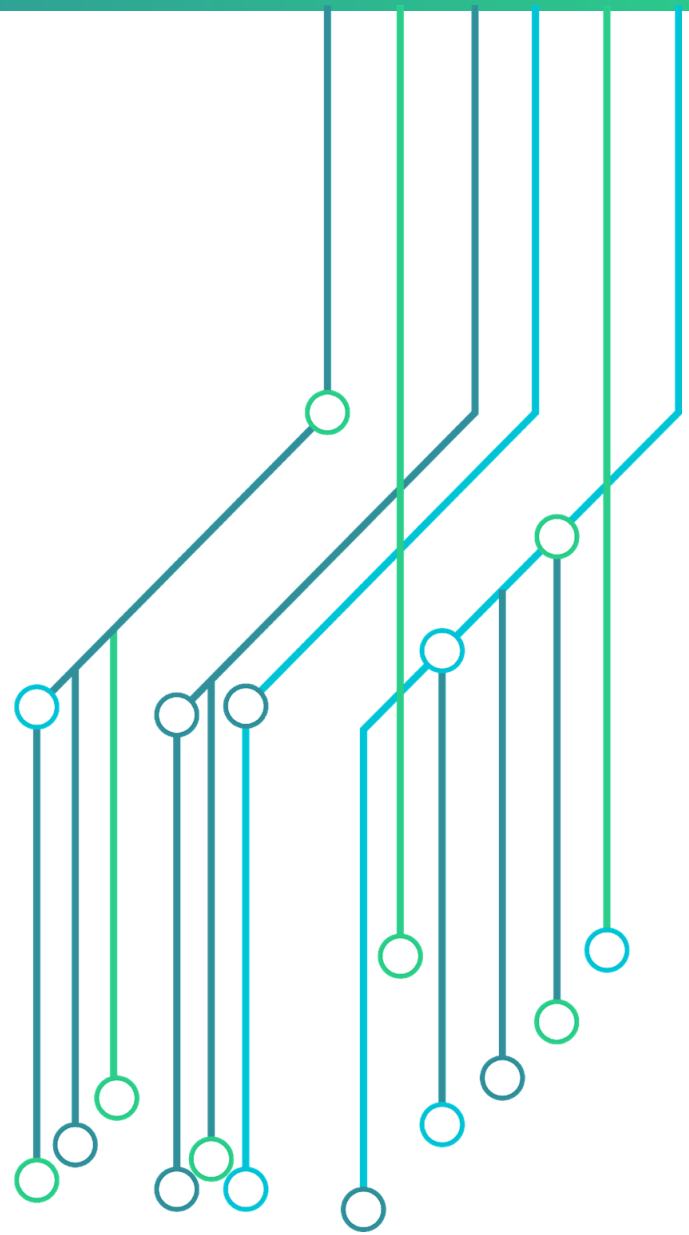
Nabsys Corporate

Company overview and mission

Nabsys

Our Mission

Advance the understanding of disease, increase diagnostic yield, and improve patient outcomes by enabling routine, accurate, cost-effective analysis of **genomic structural variation**



Nabsys: The Pioneer in Electronic Genome Mapping

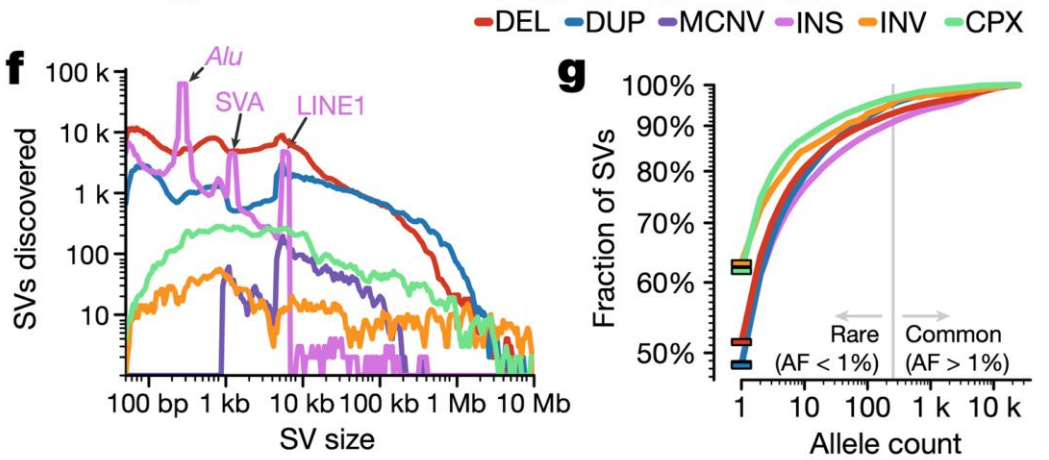
Business Summary

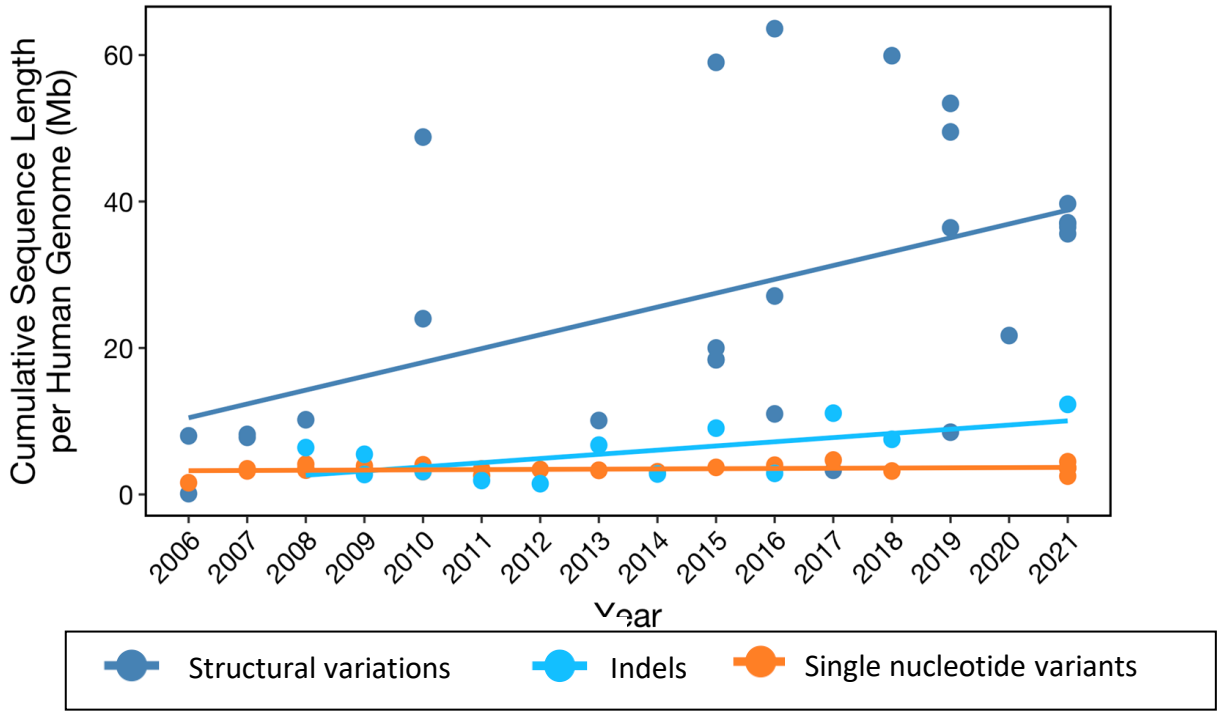
- $150M investment to date
- Patented HD-Mapping™ technology provides high resolution, long range genomic information
- OhmX™ product line in Early Access for human whole genome analysis
- Complementary to NGS
- Hitachi strategic partnership

Addressing significant unmet needs in structural variation analytics with a proprietary solid-state semiconductor-based nanodetector platform

Significant unmet need in Structural Variant analysis

- Structural variants (SVs) are the dominant form of genetic variation
- Larger SVs (>20kb) are 50-fold more likely to affect gene expression than SNVs[1]
- >300,000 SVs in the human genome[2]
- SVs are historically under analyzed due to cost and technology constraints

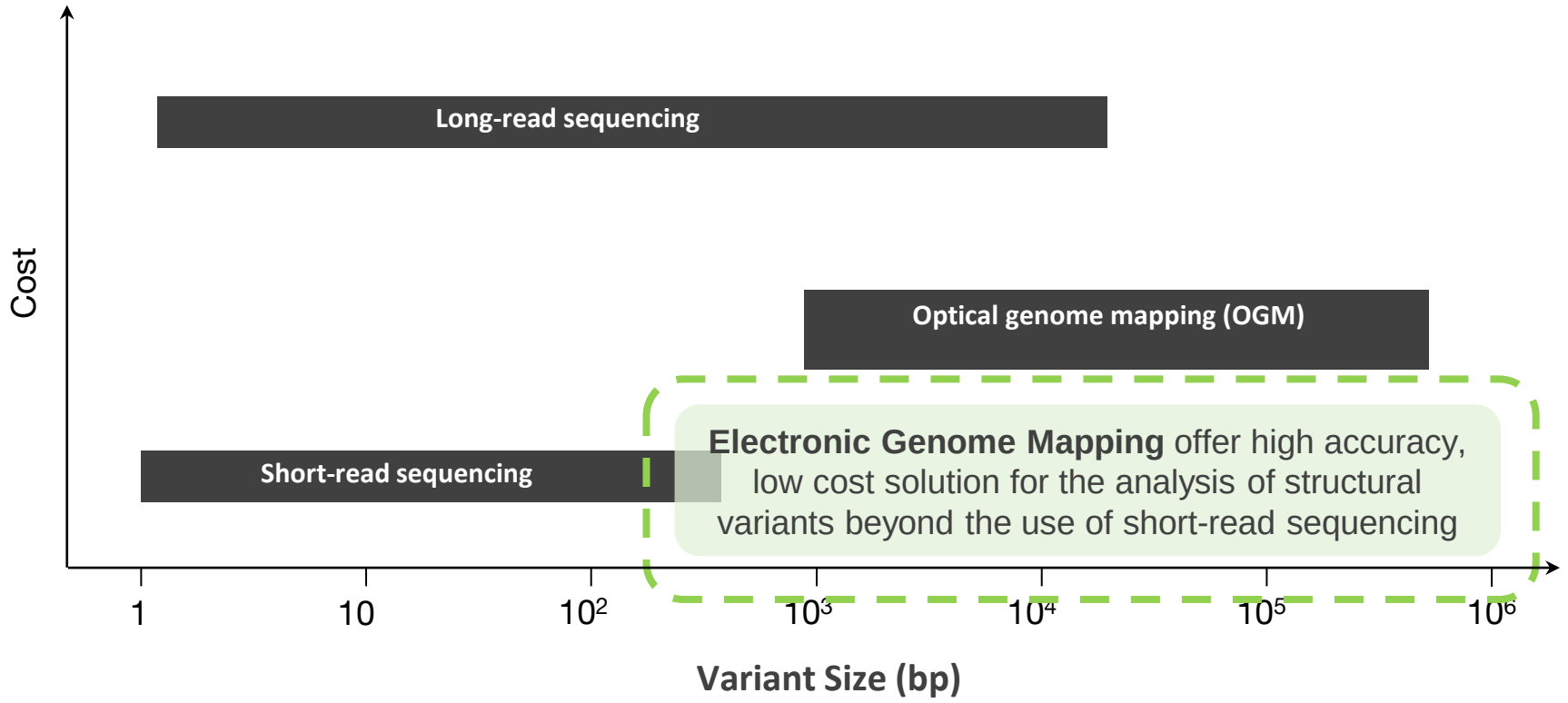




1. Chaisson, M.J.P., Sanders, A.D., Zhao, X. *et al.* Multi-platform discovery of haplotype-resolved structural variation in human genomes. *Nat Commun* **10**, 1784 (2019).
2. Collins, R. L., Brand, H, Karczewski, K. J. *et al.* A structural variation reference for medical and population genetics. *Nature* **581**, 444 (2020).

Genomics tools space

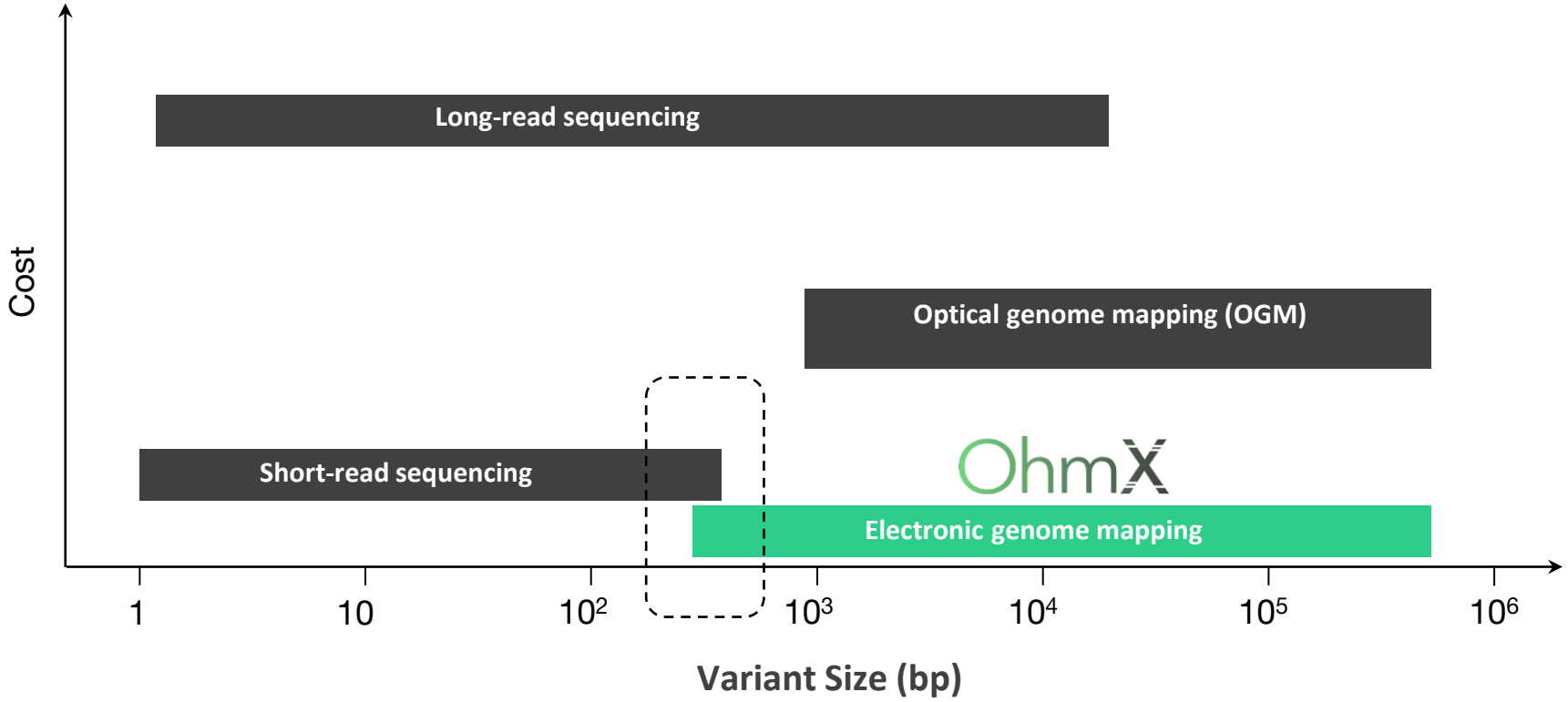
There has been a concerted effort to address variant size within the genomics tools landscape



- Short read sequencing cannot be used to analyze most SVs with confidence
- Long read sequencing is high quality, however, inherently expensive
- OGM will never reach the cost or resolution thresholds for a scalable technology

EGM positioning in the genomics tools landscape

Nabsys addresses variant size and cost



The Nabsys platform is the only platform that extends the high accuracy, low-cost genomic information obtained by short-read sequencing to longer read lengths

OhmX Analyzer

Introduction to the platform and benefits of electronic genome mapping

Nabsys

OhmX addresses limitations in structural variant detection

Nabsys platform is designed for high resolution whole genome structural variant analysis at cost that will support wider adoption of SV analysis in research and clinical markets

Integrated ecosystem
Detection down to 300 bp optimal for use alongside NGS data

High Resolution
Better diagnostic yield for cytogenetics

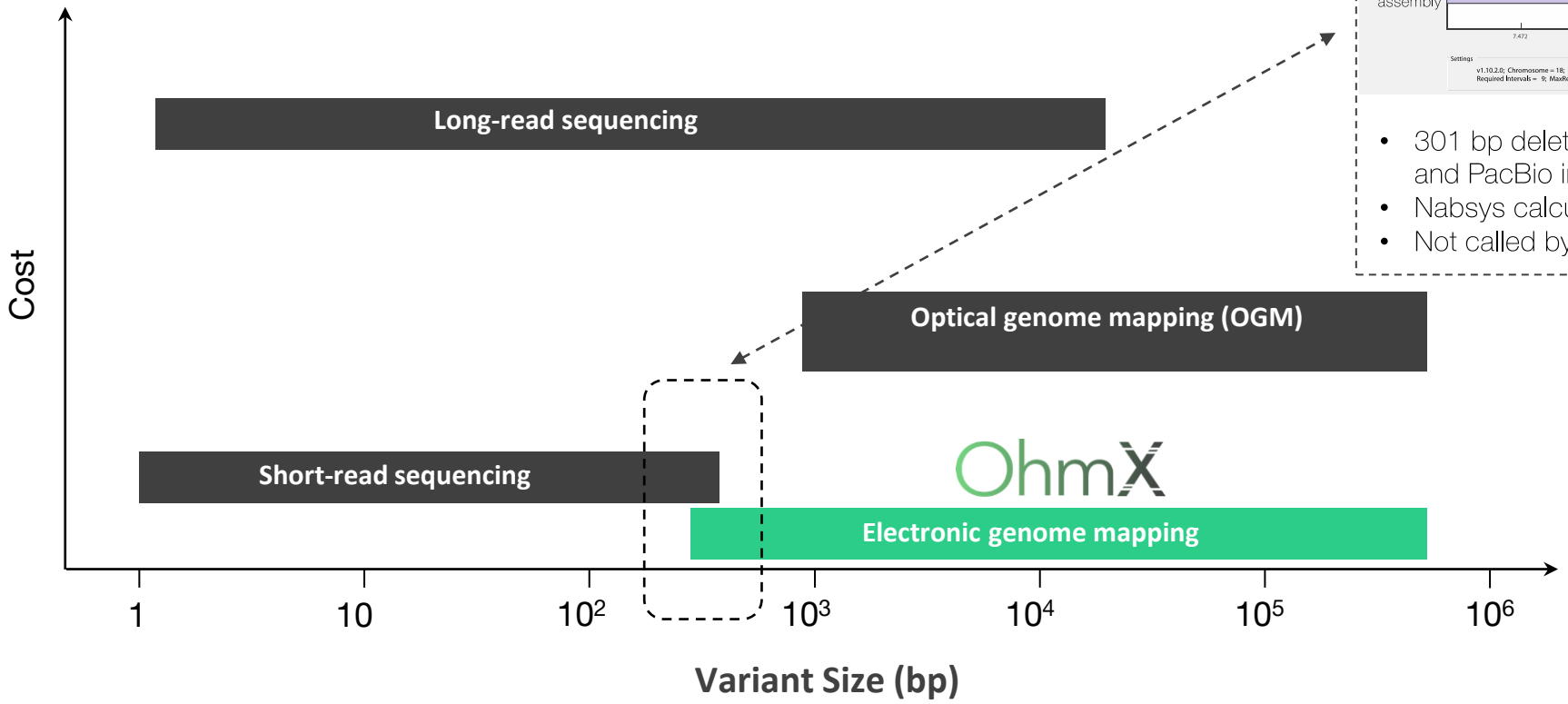
Low Cost
Low instrument and consumable compared to long-read & OGM

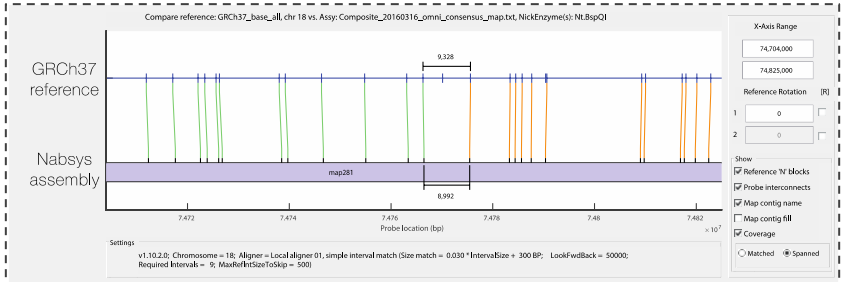
OhmX

Nabsys

Extends genomic information at a low cost

Nabsys addresses a gap in resolution

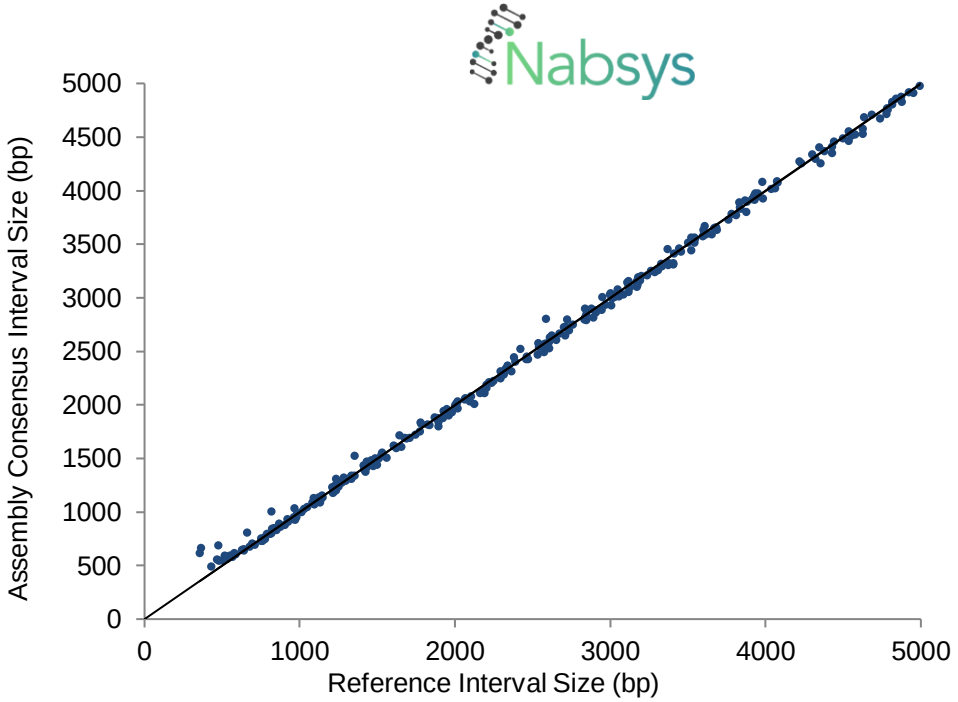




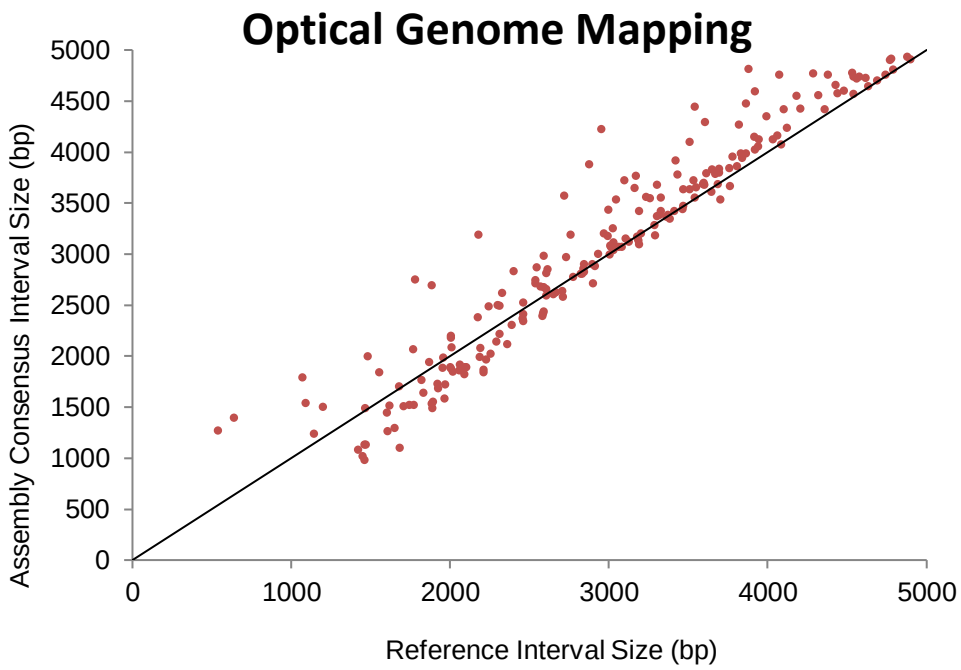
- 301 bp deletion in chromosome 18 called by Illumina and PacBio in NA24385 relative to GRCh37
- Nabsys calculated deletion size = 336 bp
- Not called by Bionano

Advantages of electronic detection

Comparison of assemblies using the same recognition sequences demonstrates the advantage of the Nabsys approach with interval accuracy and the benefit of eliminating optics for improved resolution



All intervals from 500-1,500 bp are detected in the Nabsys map assembly



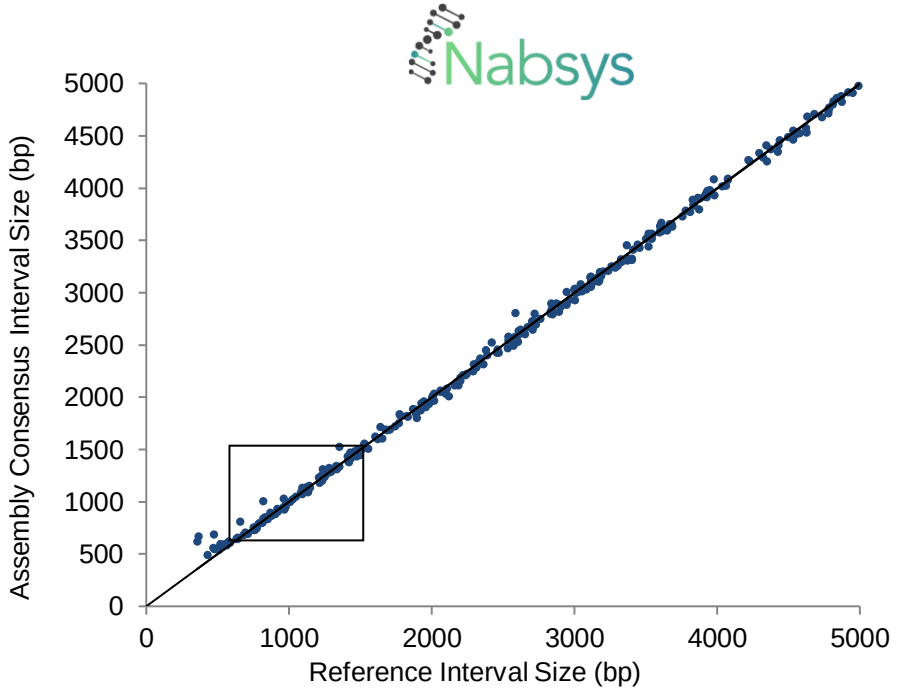
89% of intervals from 500-1,500 bp are missing in the optical map assembly

Nabsys

Source: Nabsys and BioNano assemblies of E. coli MG1655 provided by the respective companies.

Accuracy and resolution advantages of electronic detection

Nabsys electronic nano-detection provides superior accuracy over optical imaging for improved resolution



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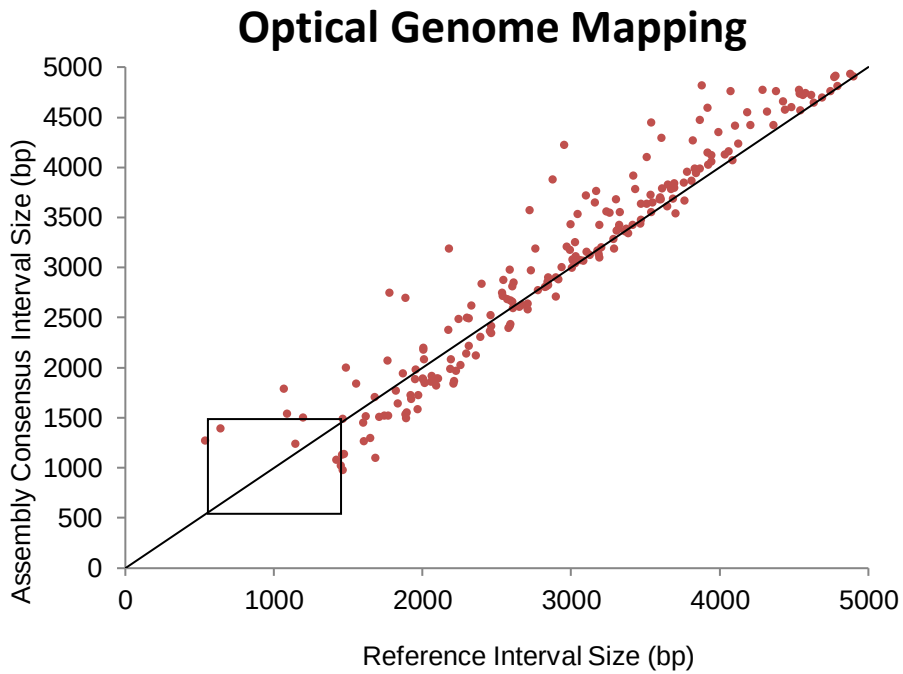
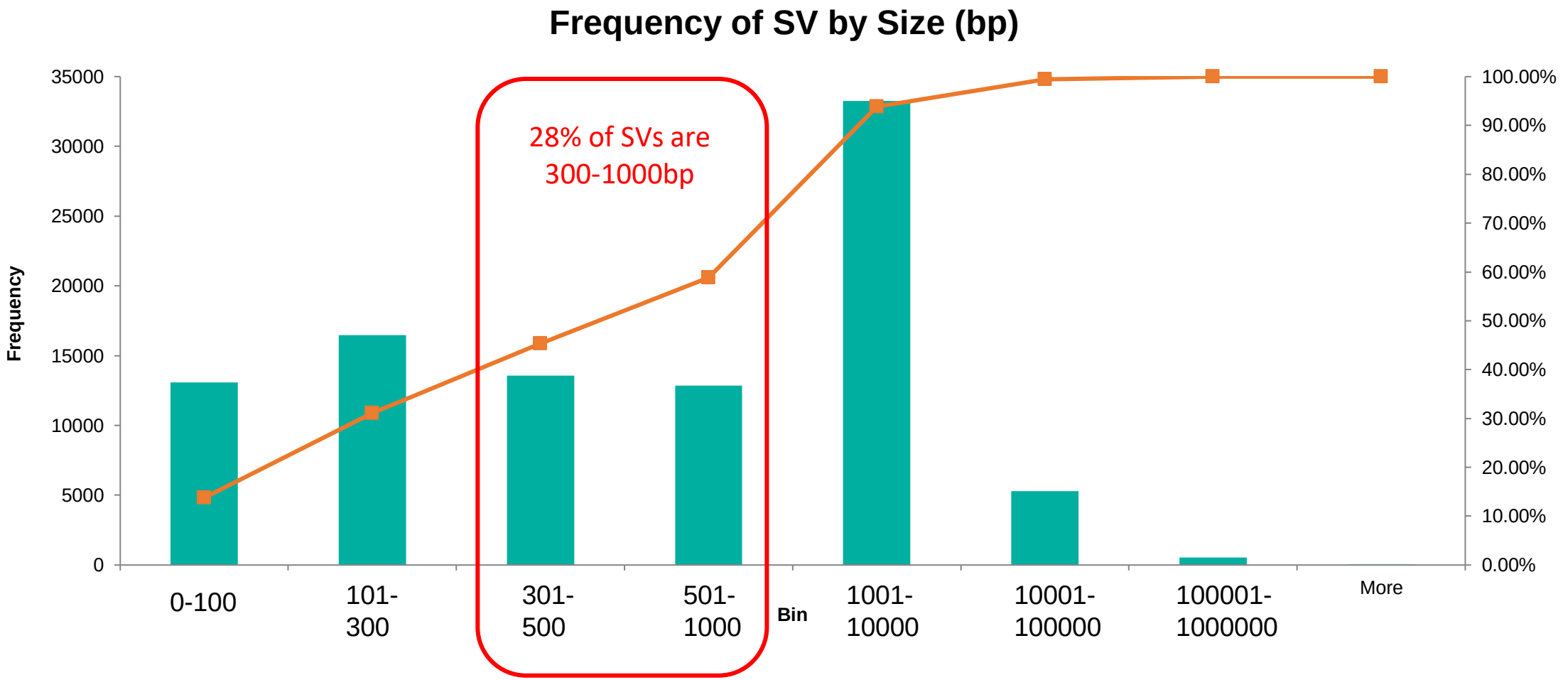


89% of intervals from 500-1,500 bp are missing in the optical map assembly

Nabsys

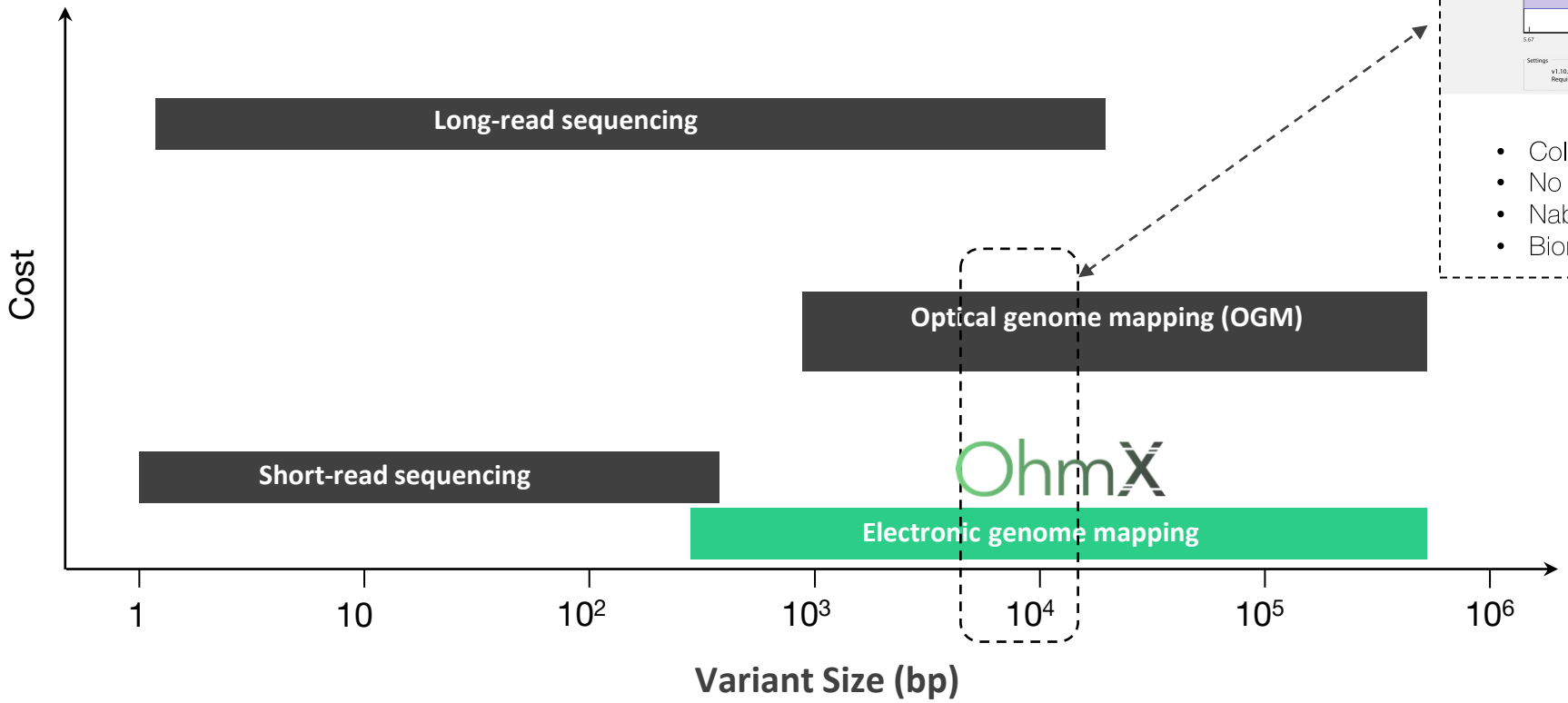
Source: Nabsys and BioNano assemblies of E. coli MG1655 provided by the respective companies.

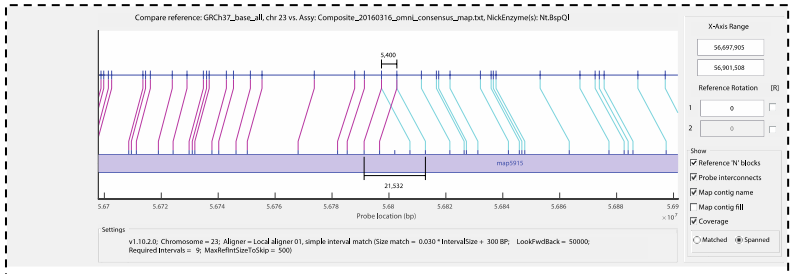
SV Size Range of Grch37 Remapped to Grch38



Lower cost and better resolution than OGM

Nabsys offers a lower cost solution to OGM

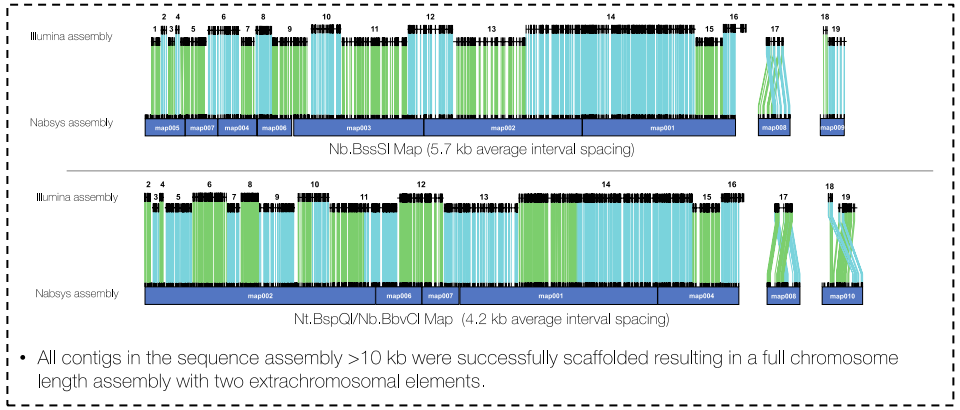




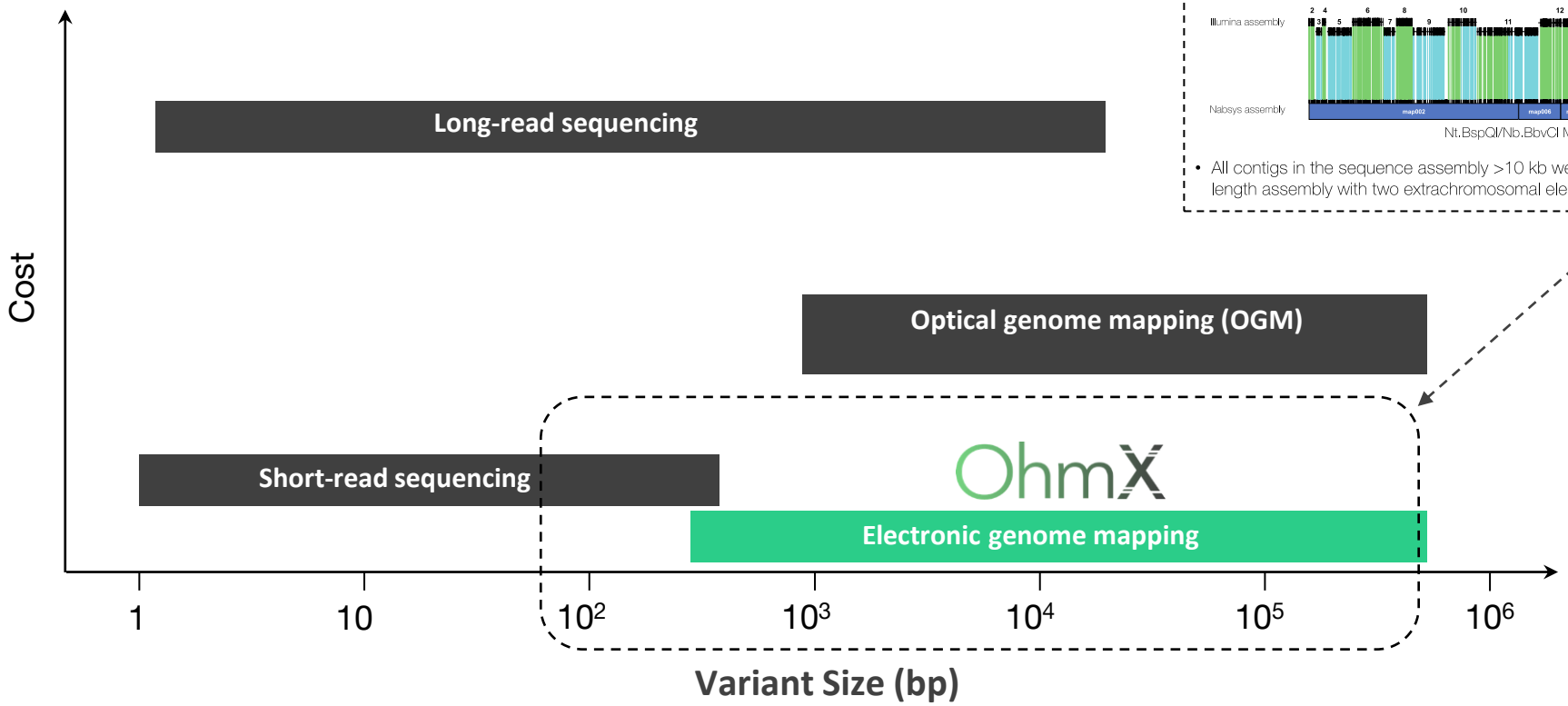
- Collapsed repeat region on X chromosome
- No call made by GiaB
- Nabsys calculated insertion size = 16,132 bp
- Bionano calculated insertion size = 16,774 bp

Provide scaffolds for short read sequencing

Nabsys improves short read sequencing



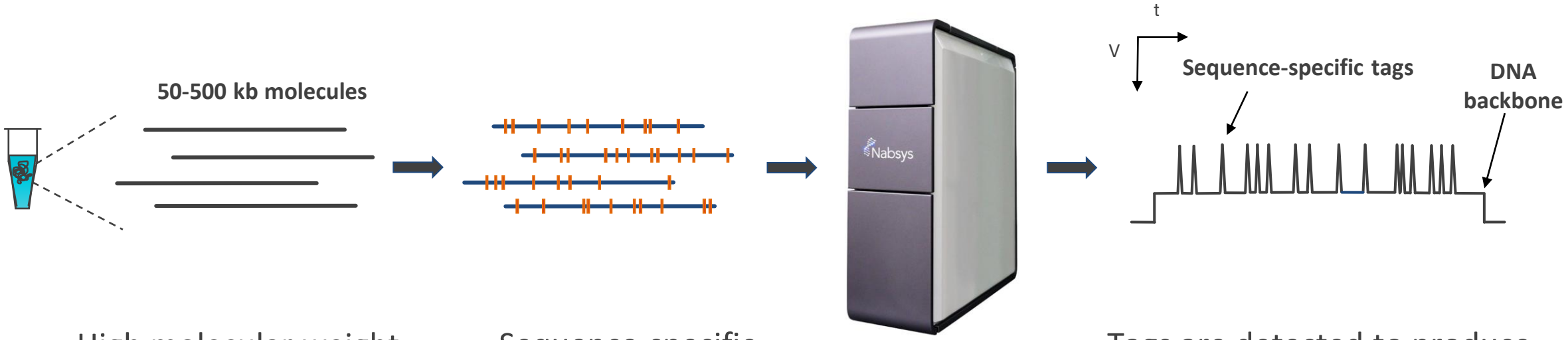
- All contigs in the sequence assembly >10 kb were successfully scaffolded resulting in a full chromosome length assembly with two extrachromosomal elements.



Work flow

Easy to use workflow and technology

EGM offers a simple workflow



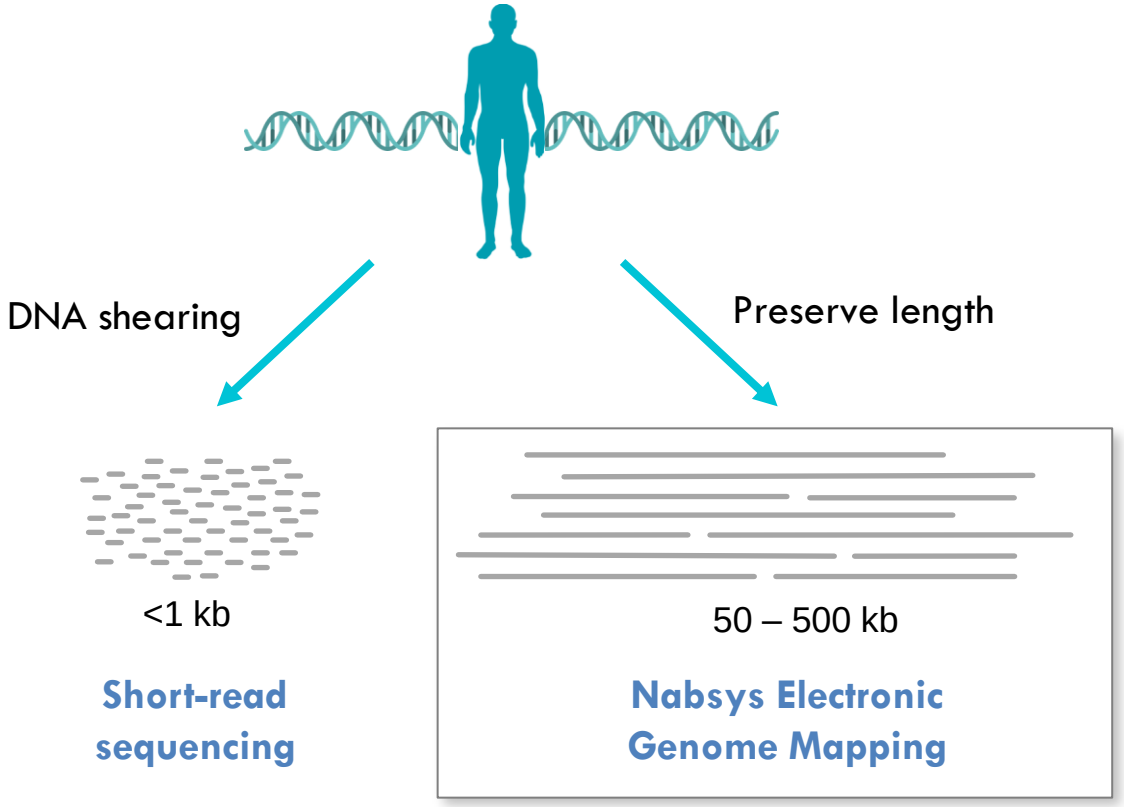
High molecular weight DNA isolation

Sequence-specific tag attachment

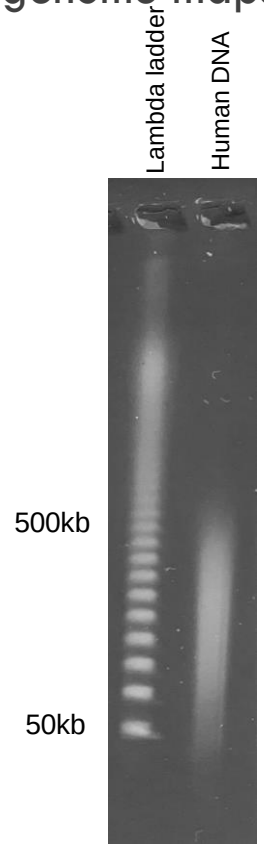
Tags are detected to produce single-molecule reads covering the entire genome

High molecular weight DNA for long-range structural information

The isolation of high molecular weight (HMW) DNA is important for generating high resolution genome maps to access long-range structural information of the genome



- Isolation of HMW DNA using commercially-available kits
- Target DNA size range of 50-500 kb



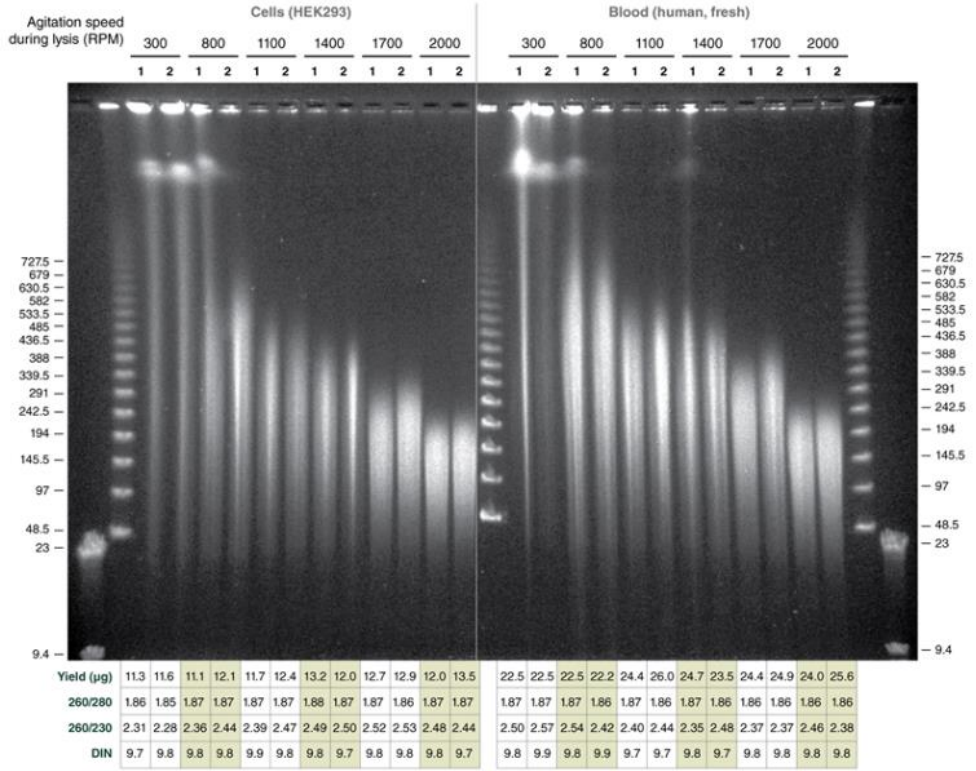
Nabsys

High molecular weight DNA isolation

Recommended workflow for isolation of HMW DNA takes ~30m

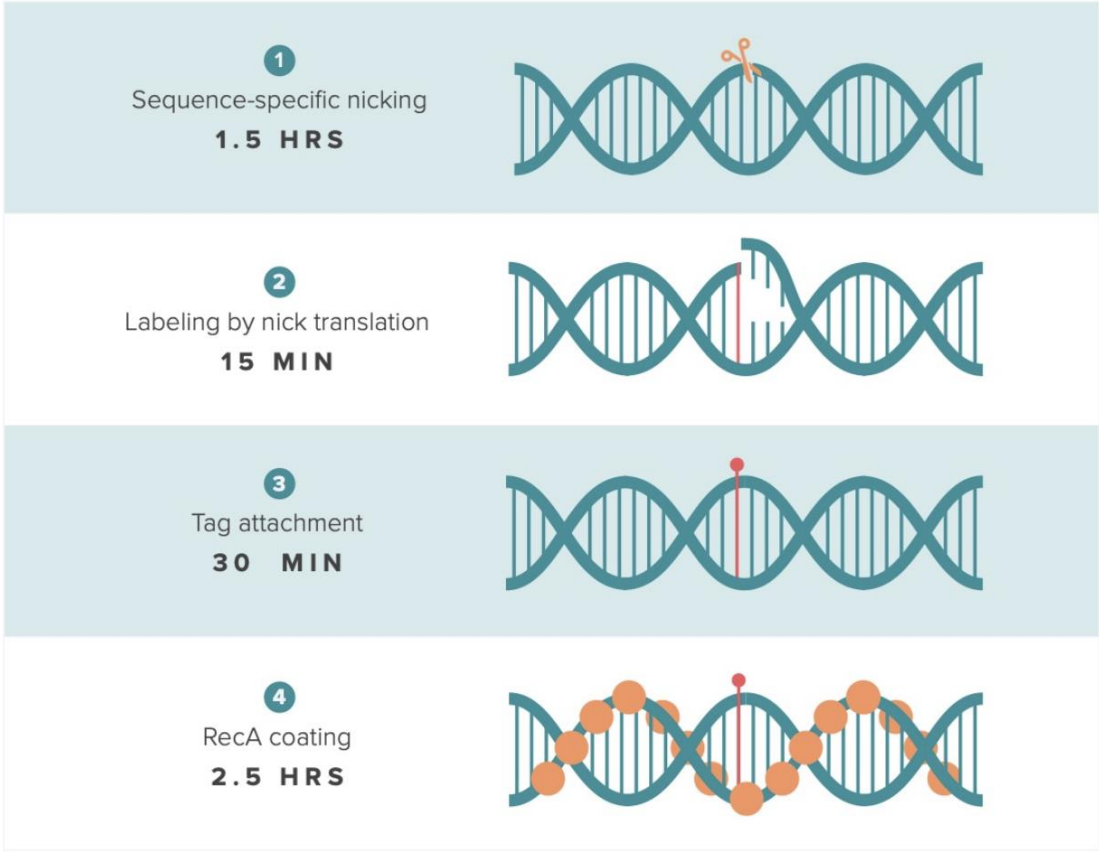
Monarch® HMW DNA Extraction Kit for Cells & Blood

Optimized agitation speed during lysis produces correct fragment lengths for EGM workflow



Sample preparation for sequence specific tagging

- Requires 4.5ug of purified HMW DNA
- ~6hrs of protocol time with minimal hands-on-time
- Sequence-specific labeling using dual nicking enzymes
  - Nickases selected for optimal density that enables for whole genome coverage
- Tagged DNA is coated with RecA protein to enable proper linear structure through the nanochannel
- Produces enough material for 4 injections into OhmX (1-2 injections is required for most applications)



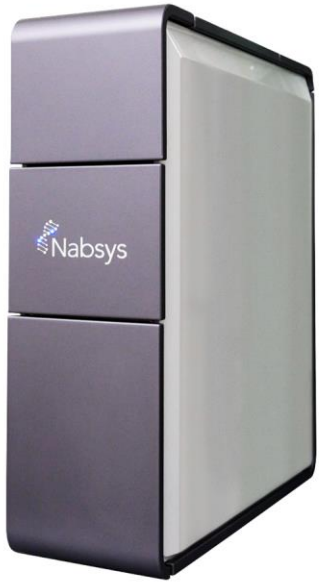
Sample introduction and data collection

Sample Injection

- System operation includes automated protocols for cleaning and running Nabsys detectors
- After sample injection, single DNA molecules translocate through the detector and are electronically analyzed to determine the distance in base-pairs between sequence-specific tags on each molecule.

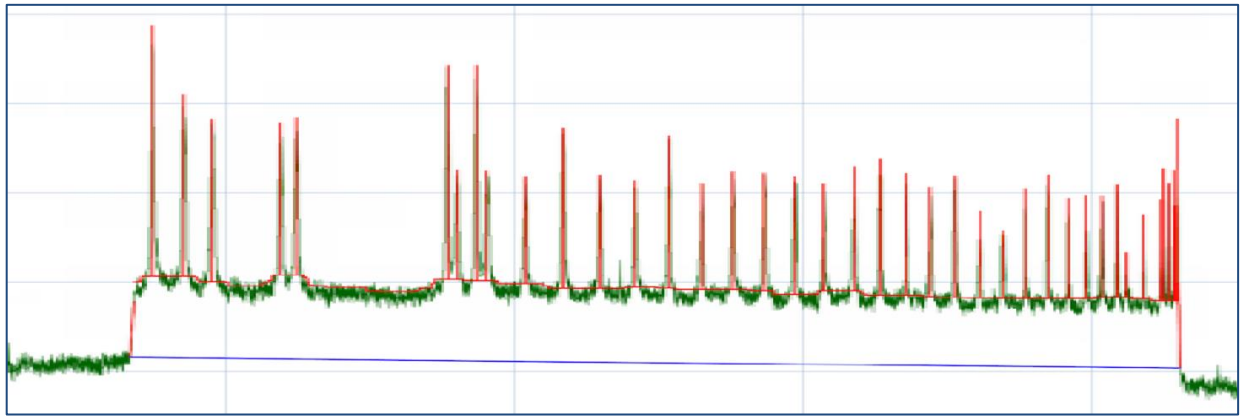
Detectors

Labels on DNA molecules are detected using electronic sensors



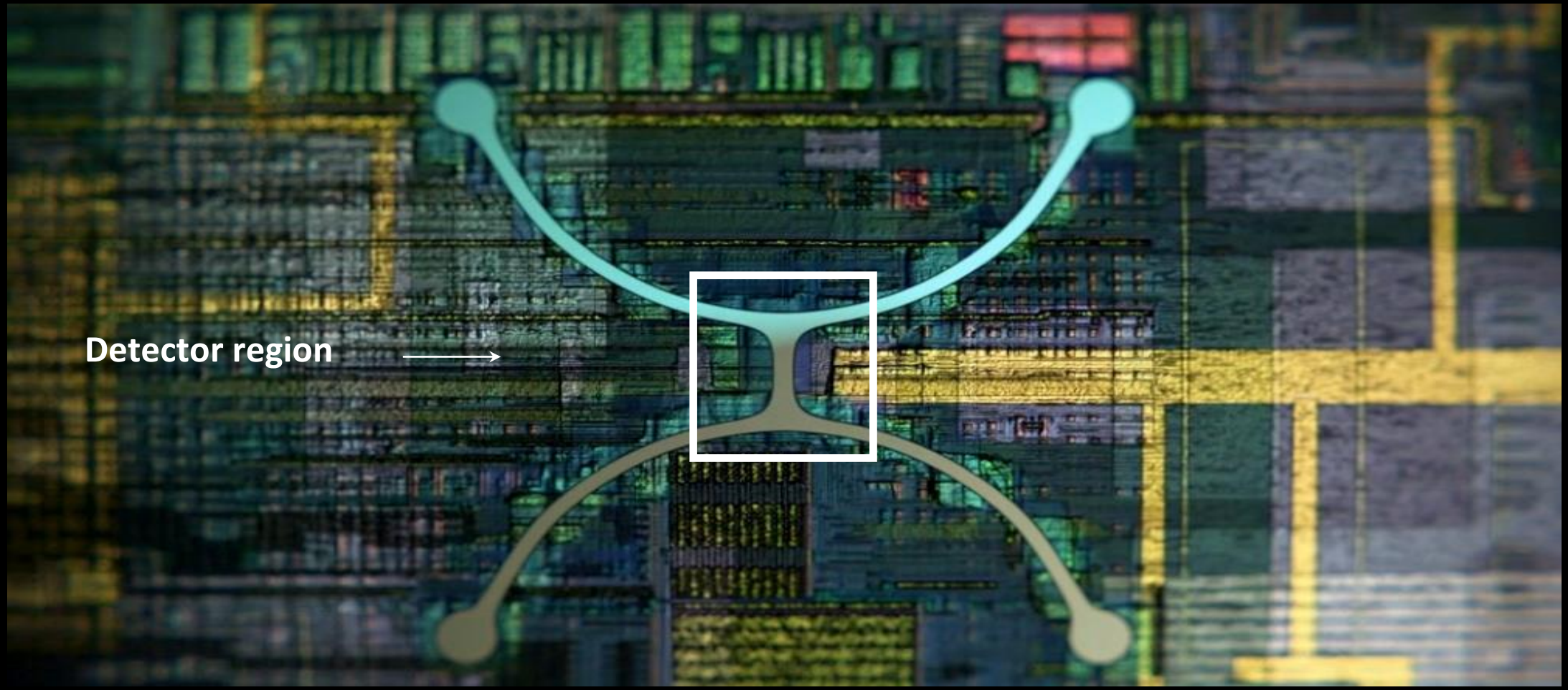
OhmX Detectors

Labeled linear DNA flows through nanochannel detectors allowing for data capture

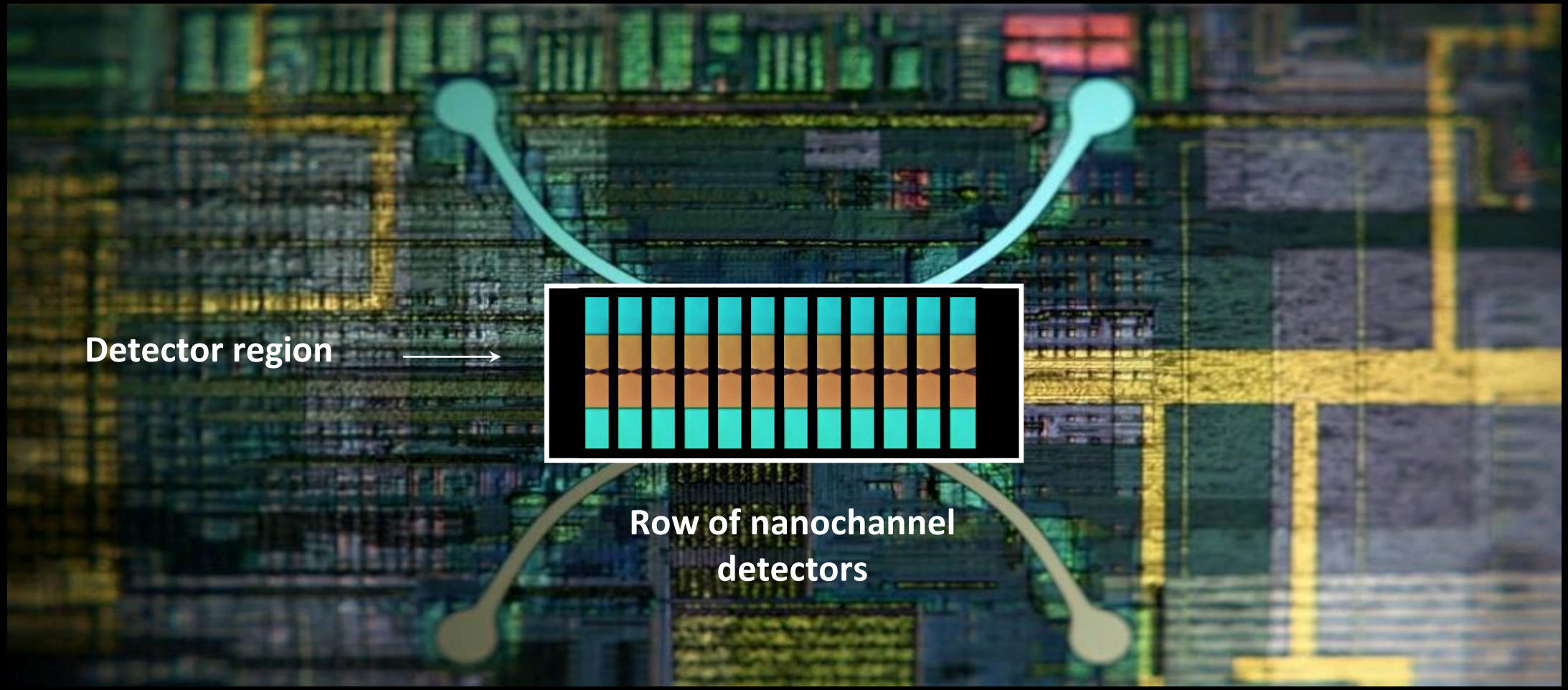


Real-time signal processing registers a voltage change over time for every tag on every DNA molecule

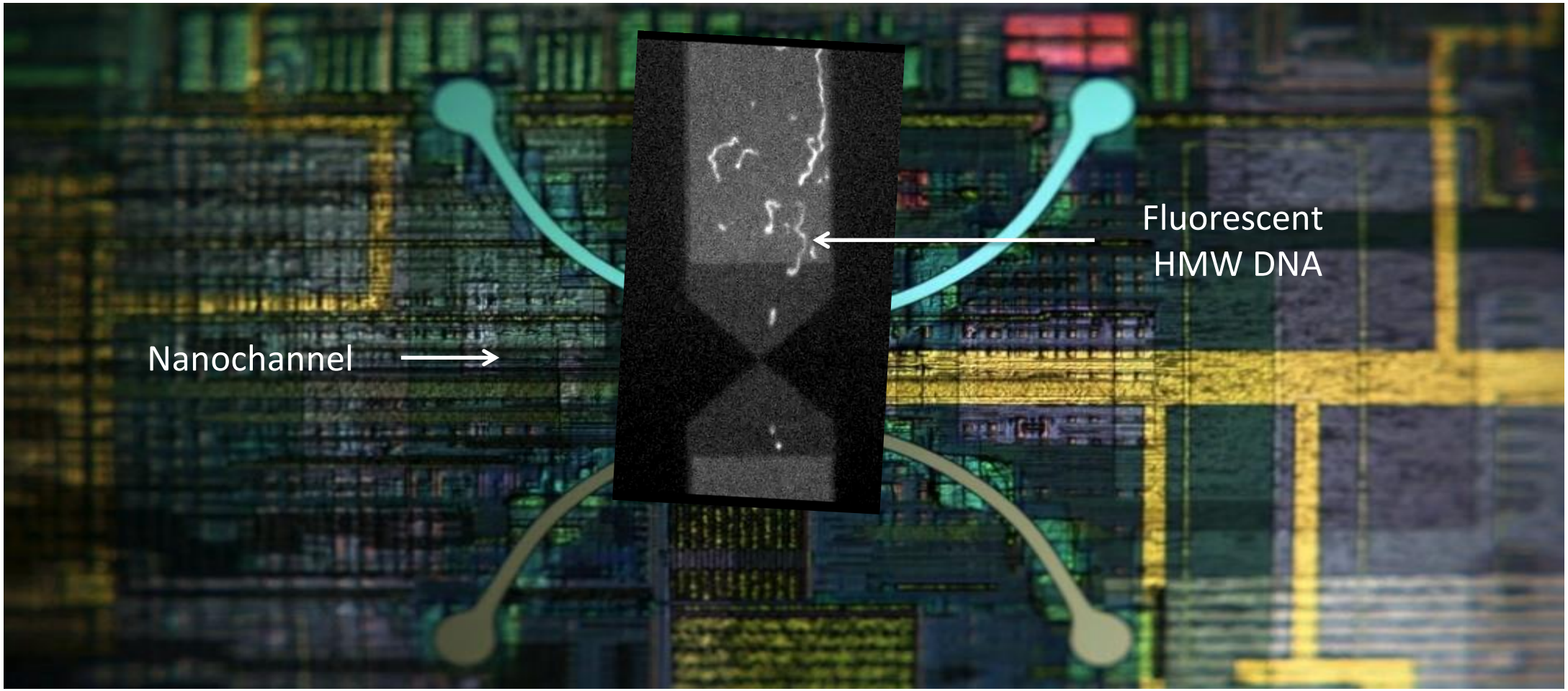
Detector schematic



Detector schematic

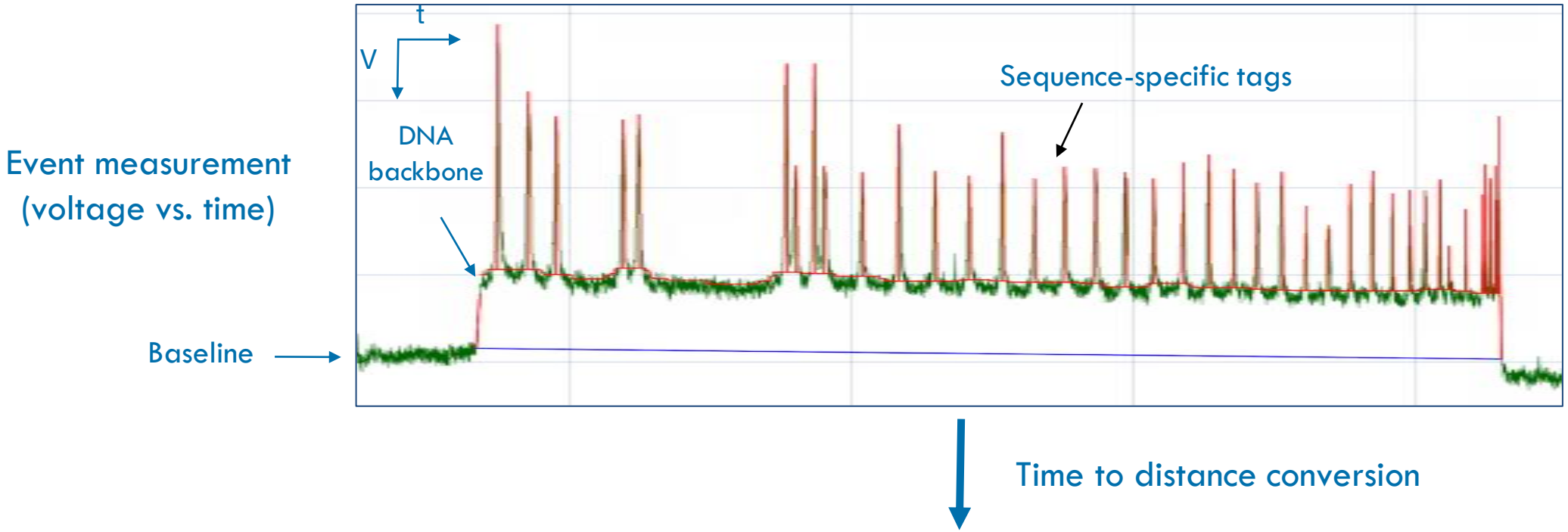


Detector schematic



Electronic detection of DNA

The time to distance between sequence specific tags is converted to base pairs to create sequence specific molecules or “maps”



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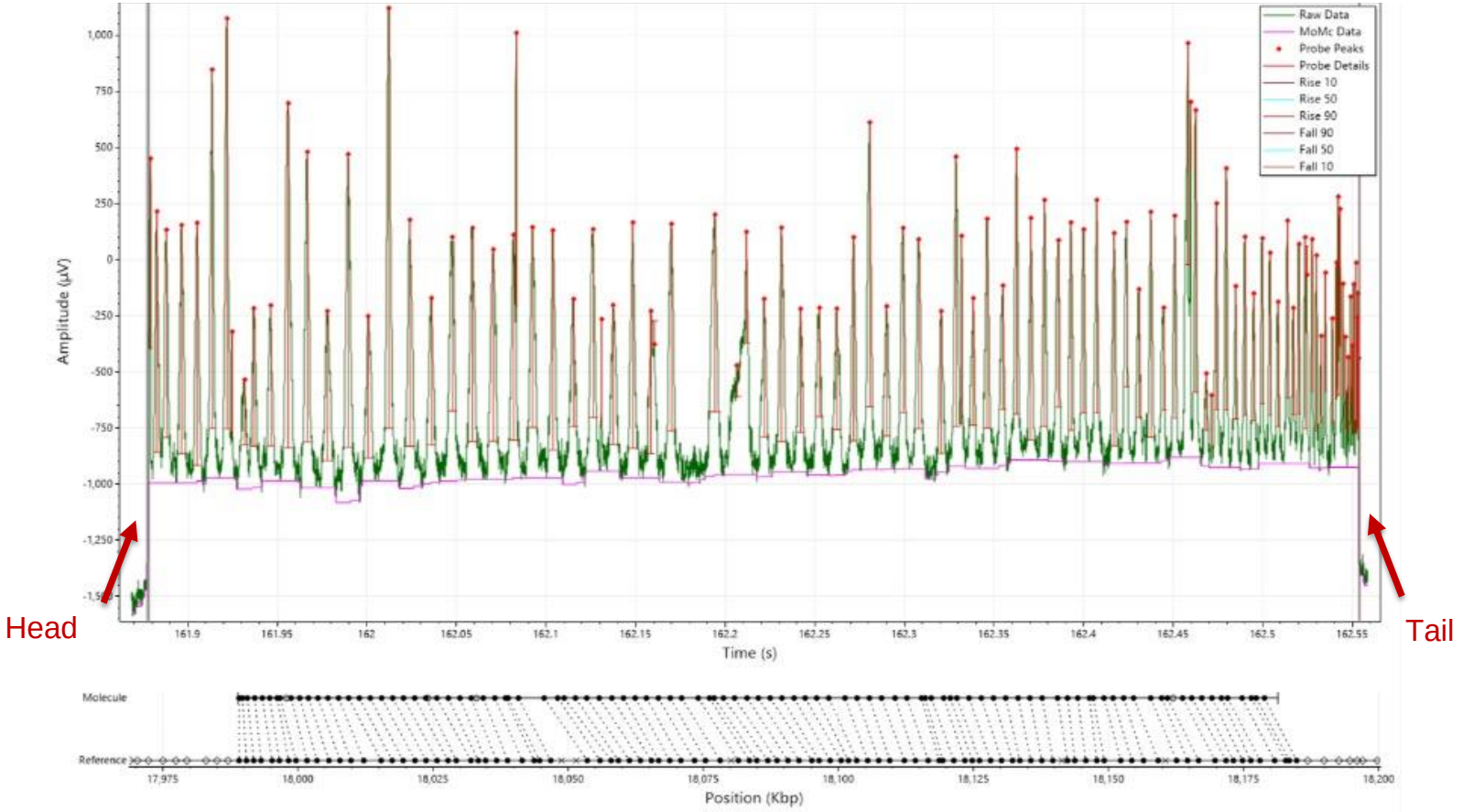
Human DNA Molecule >200 kb, 102 tags



- Dynamic measurements of HMW DNA molecules (molecules never stop moving up to 1Mb/sec through the system)
- Tremendous information content per molecule collected over time
- Machine learning will continue to improve analysis of information

Advantages of electronic detection

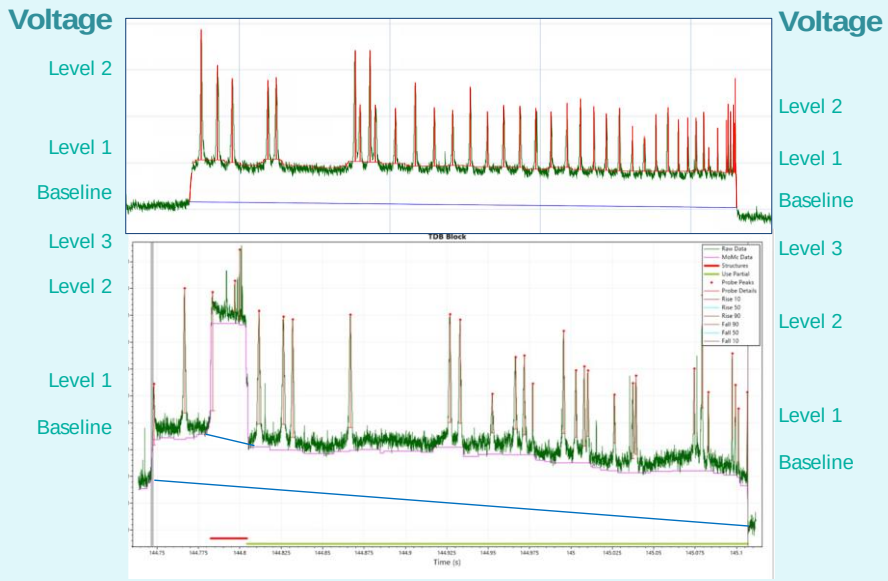
In addition to higher resolution/lower costs:

Reduced false positives:

- No Amplification
- No Ligation
- Double-stranded DNA analyzed

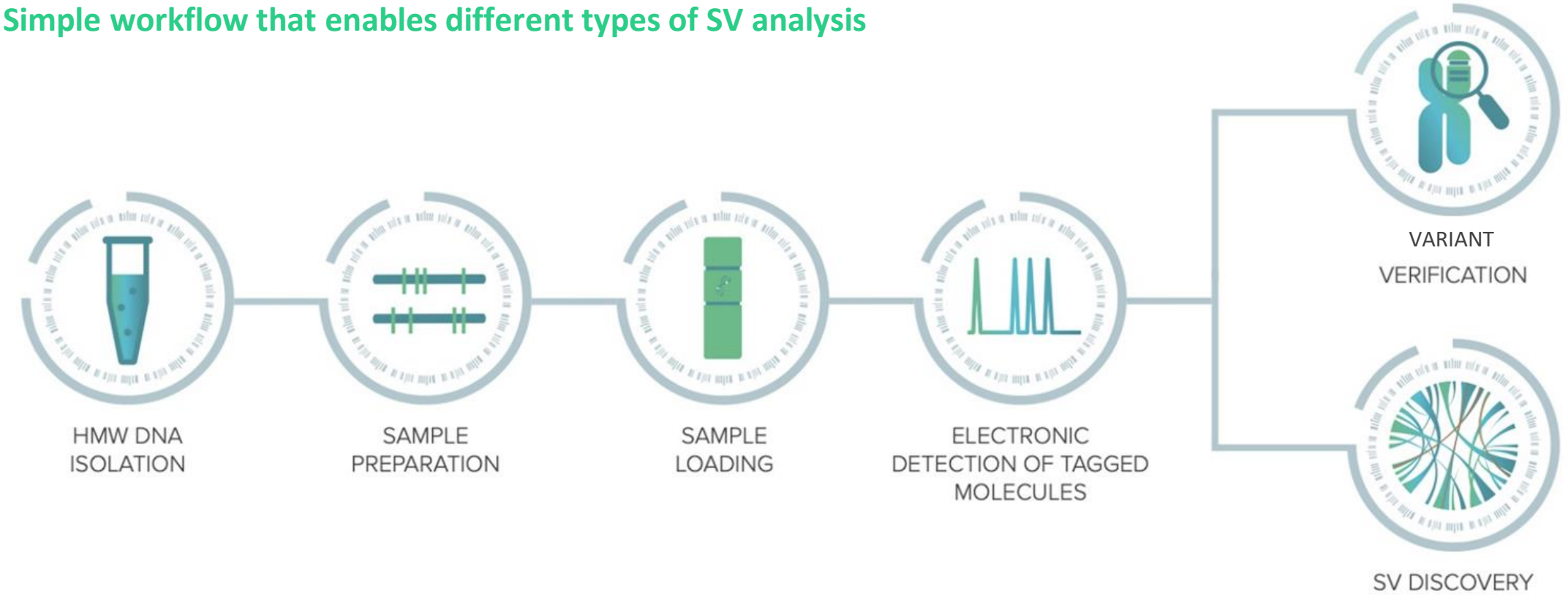
Adjacent molecules easily distinguished

- Large voltage changes when molecules co-translocate
- Differences in tag shape at beginning/end of molecules
- Eliminates false chimeric molecules found in some methods

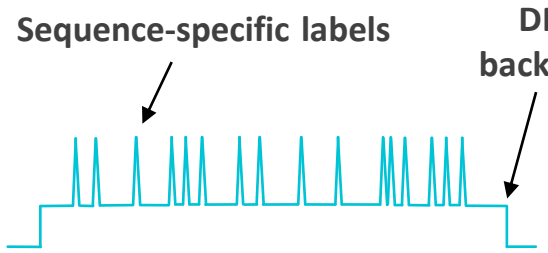


SV detection strategies – verification & discovery

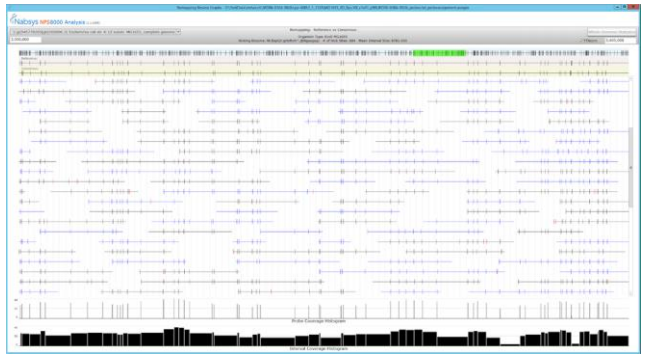
Simple workflow that enables different types of SV analysis



Analytical software for high resolution SV analysis

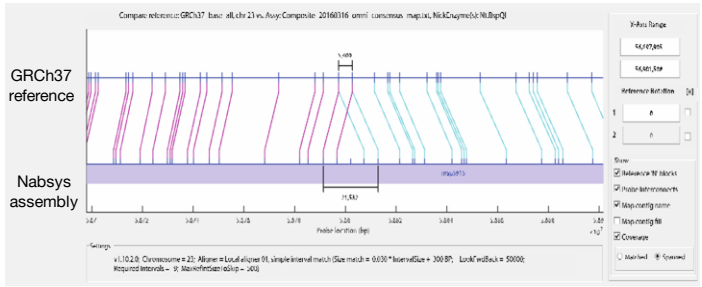


Superior signal detection enables high resolution SV analysis

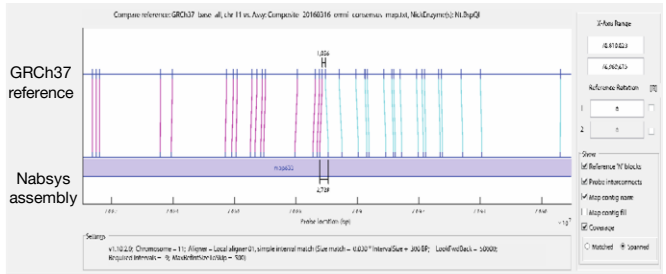


Analytical software calls SVs down to 300 bp in size

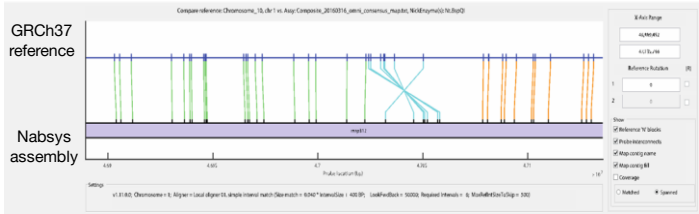
Repeat region



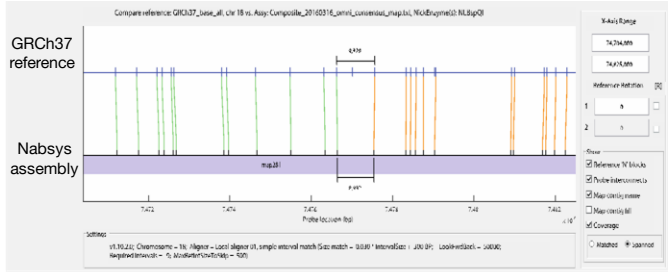
Insertion



Inversion



Deletion



SV analysis in the cloud

- Human Chromosome Explorer (HCE) developed by Hitachi and hosted in Google Cloud
- *De novo* assembly of long, single molecule maps
- SV analysis and reporting through a web browser

Google Cloud

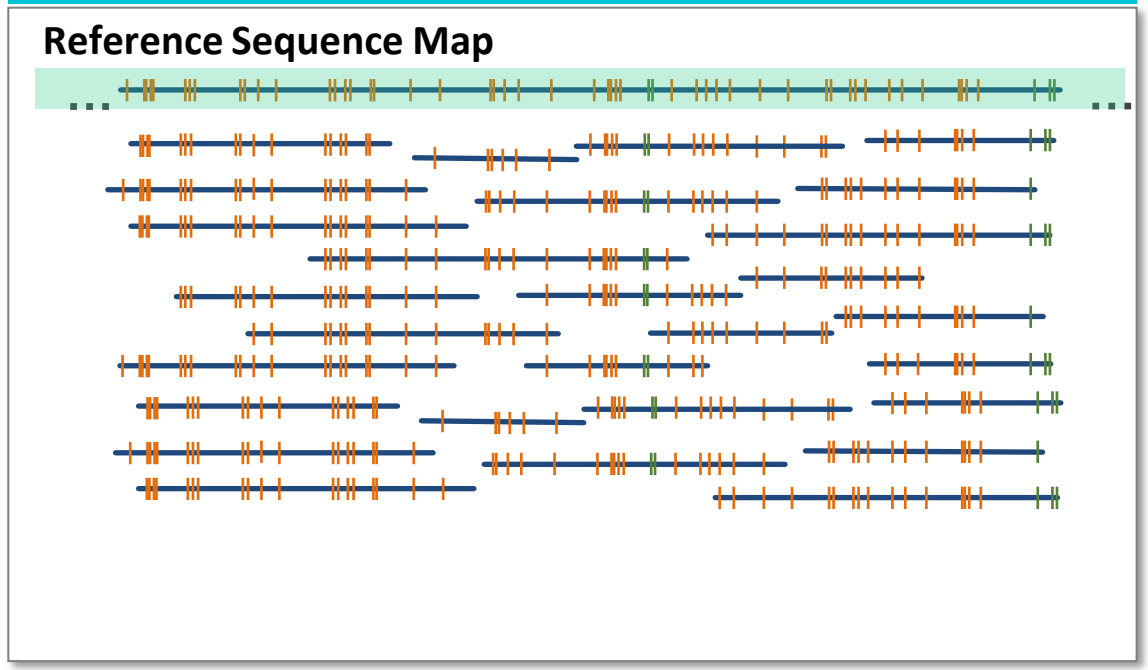
HITACHI
Inspire the Next

Nabsys

Two different analytical modes

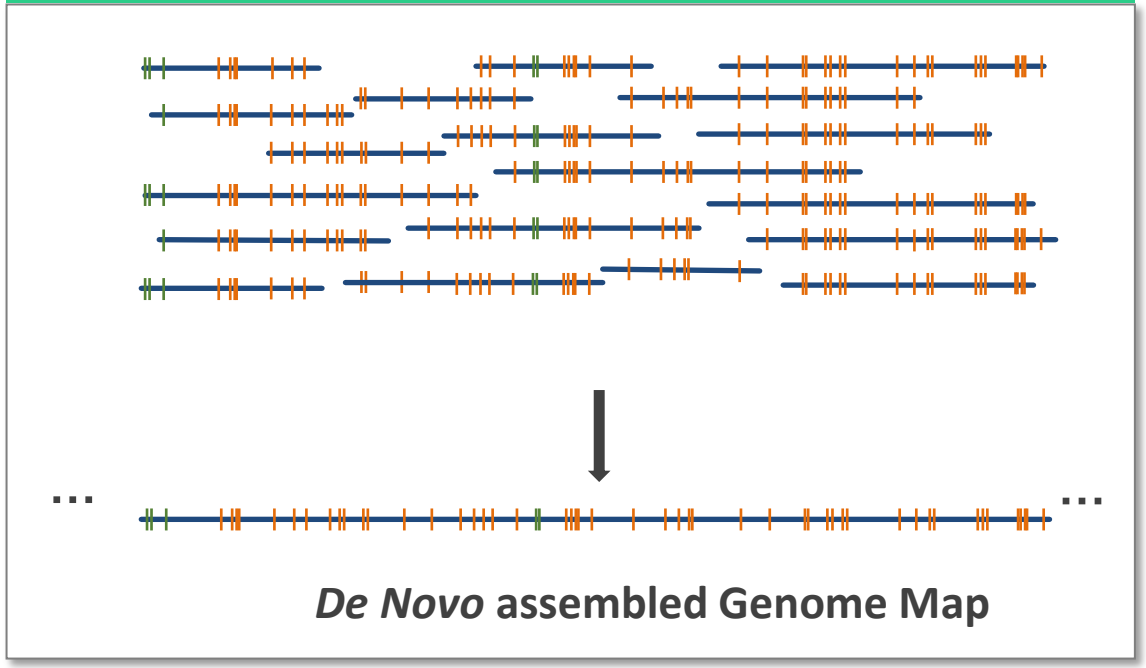
Map sequence specific molecules against a reference sequence in order to confirm the presence or absence of an SV

Remapping: **Variant Verification Mode**



De novo **assemble all sequence specific molecules into an entire genome based on labeling patterns and analyze the entire genome**

Map Assembly: **SV Discovery Mode**



Variant Verification Mode

Confirm and interrogate the presence or absence of SVs

Remapping: **Variant Verification Mode**

Confirm CNV calls made though whole genome sequencing (i.e. Illumina DRAGEN calls)

Interrogate certain regions of the genome (i.e. target genes)

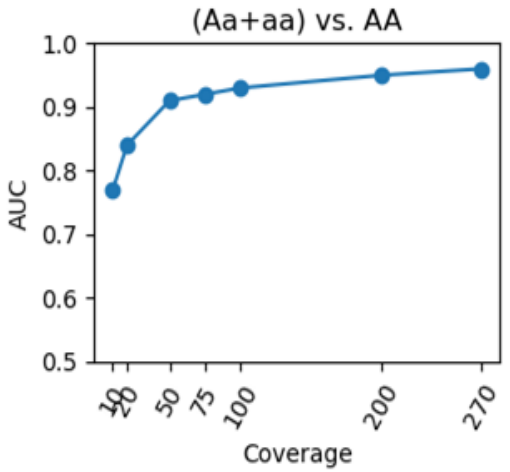
Look for different types of SVs related to disorders (i.e. repeat expansions)

Confirm unclear microarray data

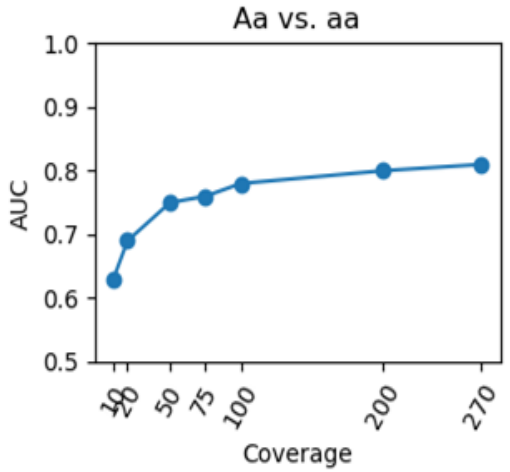
Low coverage requirements, low cost, faster run times and amenable to the development of LDTs and targeted assays

HG002 Performance vs. coverage: No significant decrease in performance at 50X coverage

Presence/Absence of SV



Heterozygous vs Homozygous SV



SV Discovery Mode

***De novo* assembly of an entire genome based on mapped reads for whole genome SV analysis**

Map Assembly: **SV Discovery Mode**

Perform whole genome SV analysis beyond cytogenetics and OGM

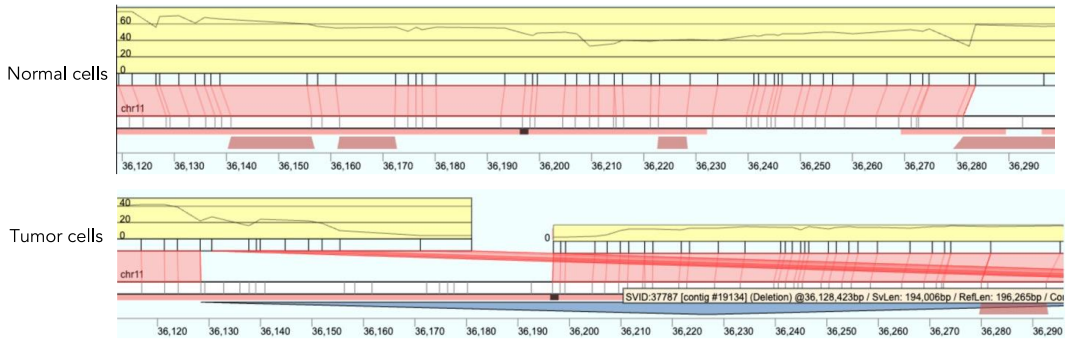
Identify genome wide breakpoints and instabilities

Whole genome scaffolding to resolve complexity for sequencing technologies

Cell line fingerprinting

Higher coverage requirements that maximize use of the detector. No reliance on a reference genome for assembly and good for whole genome discovery applications

HCE can be used to analyze whole genomes and requires greater than 150X coverage

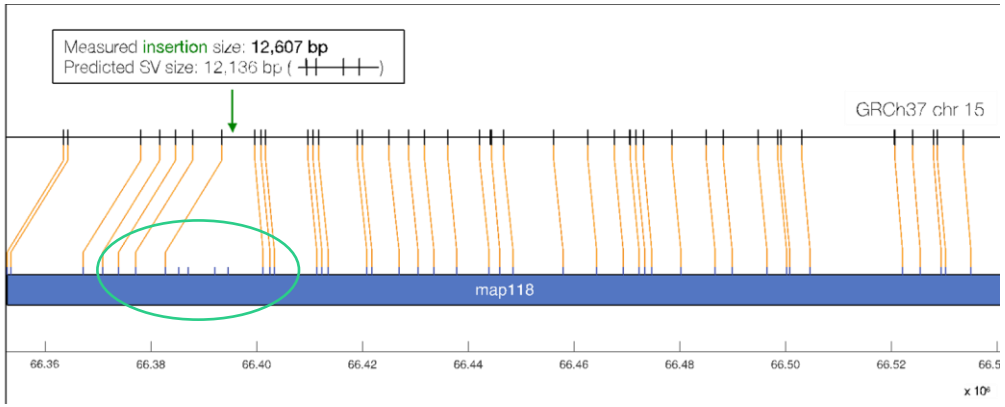


Higher coverage requirements that maximize use of the detector. No reliance on a reference genome for assembly and good for whole genome discovery applications

Nabsys

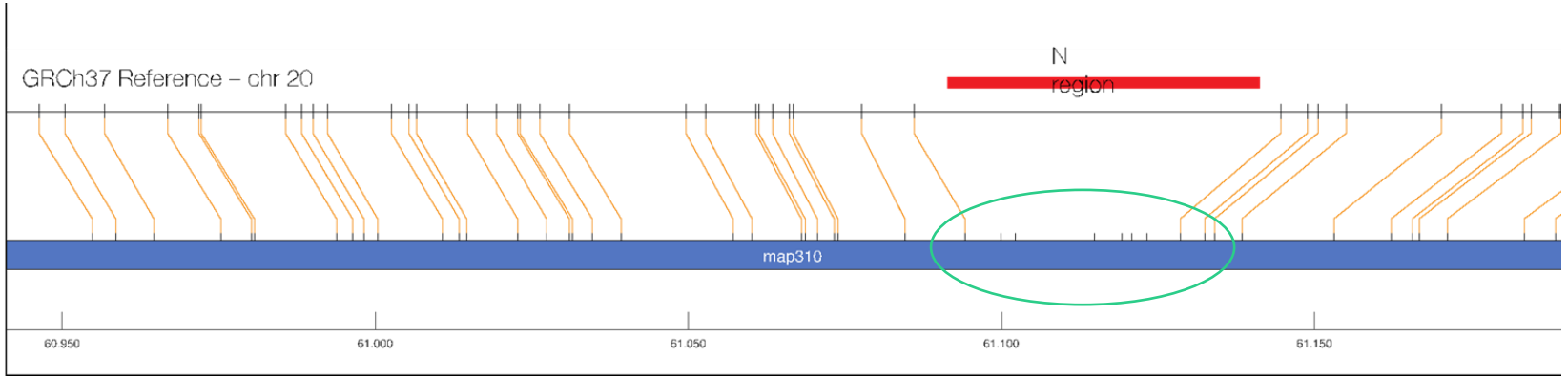
Two examples of high confidence Nabsys calls

Variant verification mode



- Low coverage enabled detection of 4 labels that did not map to reference sequence indicates an insertion
- Confirmation of a *de novo* ~12kb insertion in chromosome 15 of GRCh37

SV Discovery Mode



- De novo assembly of single molecule reads from human genome NA24385 spans N region on chromosome 20
- Incorrect gap size identified

Applications

Applications and case studies

Applications in human genomics

Functional Genomics

Rare Disease

Oncology

CLINICAL GENOMICS

PRECISION MEDICINE

Cell and Gene Therapy

Pharmacogenomics

Emerging Technology

Nabsys

OhmX consolidates cytogenomics workflows

Unlike legacy cytogenomic tools, OhmX is able to see all types of structural variants at high resolution, in a hypothesis-free way.

Variant Type	OhmX	Karyotyping	FISH	Microarrays
Aneuploidy	✓	✓	✓ targeted	✓
Deletion	✓	✓ >5-10Mbp	✓ targeted	✓
Duplication	✓	✓ >5-10Mbp	✓ targeted	✓
Translocation	✓	✓ >5-10Mbp	✓ targeted	X
Inversion	✓	✓ >5-10Mbp	✓ targeted	X
Repeats	✓	X	X	X

Nabsys

Case study in Functional Genomics

The Genome in a Bottle Consortium is a public-private-academic consortium hosted by NIST to develop technical infrastructure to enable translation of whole human genome sequencing to clinical practice

Contributing technologies (~17 different callers) to tackle both small and large variants

- Illumina
- PacBio
- 10X Genomics
- Complete Genomics
- Bionano Genomics
- Nabsys

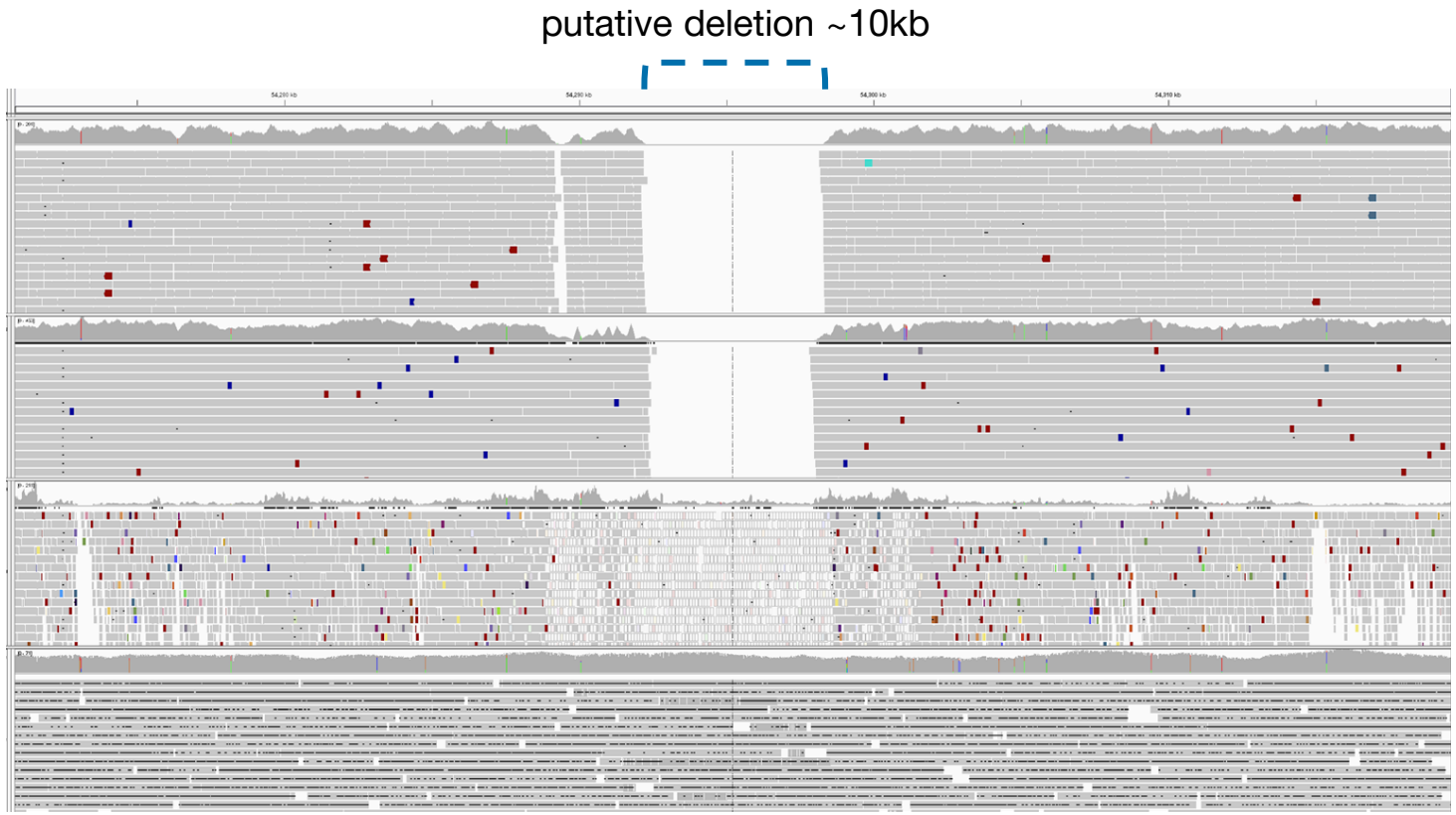
NIST

JMB

Nabsys

There is a need for orthogonal technologies

Discrepancy over a 10Kb deletion in the human reference across different technologies



CNVnator calls deletion
Illumina data collected by different groups is able to call short variants highlighted in colors but does not see the deletion

10x calls with low confidence and says its heterozygous

PacBio identifies deletion and calls it homozygous

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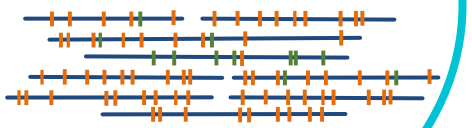
Use of multiple technologies

Leveraging strengths of different data sets to generate variant calls for use in benchmarking and validating new technologies and pipelines

Short-Read Sequencing

Long-Read Sequencing

Whole Genome Mapping



Nabsys

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Summary

OhmX platform product summary

OhmX address limitations of other SV platforms

Nabsys platform is designed for high resolution whole genome structural variant analysis at cost that should support wider adoption of SV analysis in research and cytogenetics

Integrated ecosystem
Detection down to 300 bp optimal for use alongside NGS data

High Resolution
Better diagnostic yield for cytogenetics

Low Cost
Low instrument and consumable compared to long-read & OGM

OhmX

Nabsys

Nabsys

Thank you

Sal Mazza, Director of Sales