Revealing Structural Variation that Matters with Optical Genome Mapping

bionano™

10.24.2022

Modern Cytogenetics Laboratories are Facing Analytical and Operational Challenges

Market Drivers & Pressures

Strong demand for analysis of chromosomal aberrations

Multiple medical guidelines drive need for more comprehensive cytogenetic testing across applications

Increasing test volume

Global demand for cytogenetic services is expected to grow by over 8% per year

Growing test complexity

Discoveries in genomics and evolving guidelines have resulted in an increasing number of biomarkers and reflex pathways

Challenges Labs Face

Complex Workflows

Traditional cytogenetic technologies like karyotyping and FISH have complex prep and manual analysis steps, limiting overall throughput

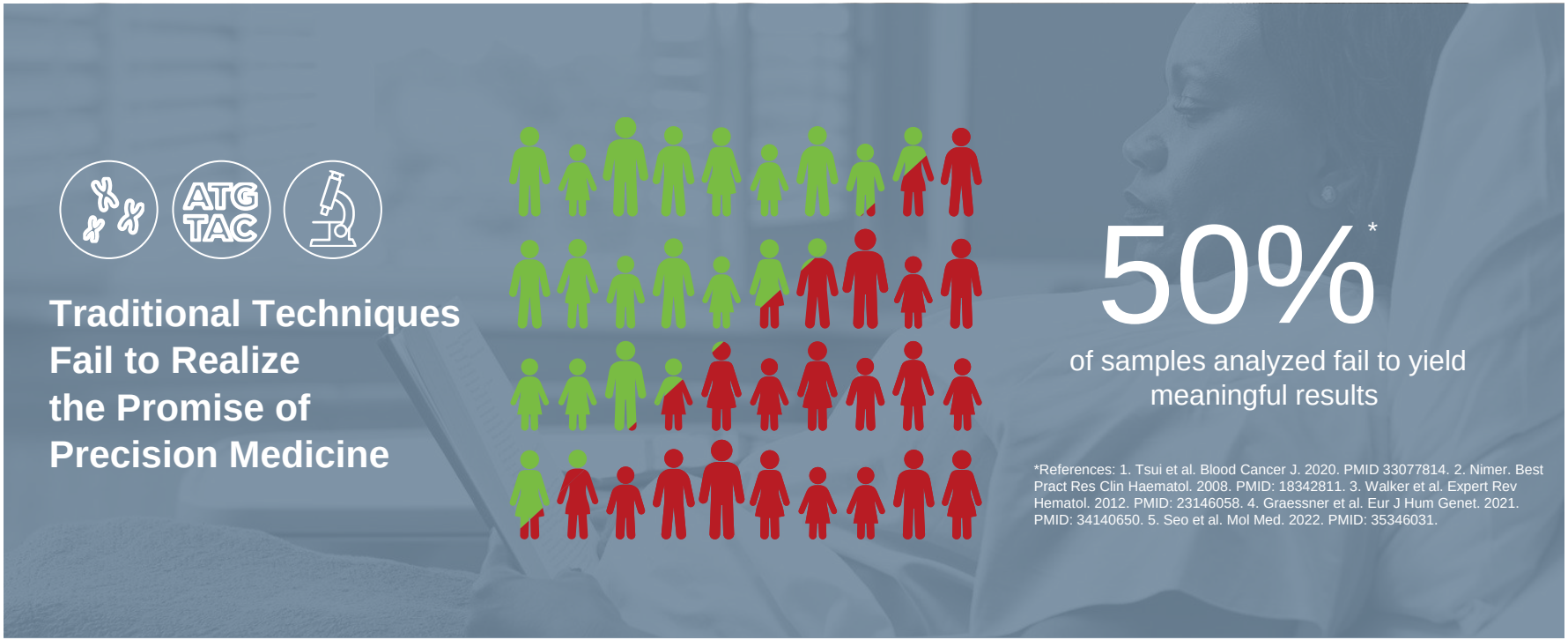
Resource constraints

The number of trained clinical cytogeneticists is expected to decline — creating challenges for meeting growing test demand

Technology limitations

Current cytogenetic technologies may be limited in their ability to meet the needs of growing test menus and complex biomarkers

Current Approaches to Assess Genomic Aberrations Fall Short



*References: 1. Tsui et al. Blood Cancer J. 2020. PMID 33077814. 2. Nimer. Best Pract Res Clin Haematol. 2008. PMID: 18342811. 3. Walker et al. Expert Rev Hematol. 2012. PMID: 23146058. 4. Graessner et al. Eur J Hum Genet. 2021. PMID: 34140650. 5. Seo et al. Mol Med. 2022. PMID: 35346031.

“ I do sign our chromosomal analysis by Karyotype on a regular basis, in diseases like AML or heme malignancies. In around maybe 40-50% of these cases, they come back with a normal Karyotype, or the FISH result is negative.”

Dr. Ravindra Kolhe, MD, PhD, FCAP
Professor, Dept of Pathology
Augusta University

OGM Solves for the Limitations of Current Methods for Chromosomal Aberration Detection

Method	Karyotyping	FISH	CMA	OGM
Resolution	>5-10 Mbp	>100-200 kbp	>50-100 kbp	**>500 bp**
Detection bias	Hypothesis-free	Single probe	Design bias	**Hypothesis-free**
Detection of balanced rearrangements	Yes	Yes	No	**Yes**
Cell culture needed	Yes	No	No	**No**
Average TAT	1-2 weeks	3-5 days	<1 week	**< 1 week**
Digital analysis	No	No	Yes	**Yes**
Sample-to-answer platform	No	No	Yes	**Yes**

Broeckel U et al. Multisite Study of Optical Genome Mapping of Retrospective and Prospective Constitutional Disorder Cohorts. medRxiv 2022.12.26.22283900; doi: https://doi.org/10.1101/2022.12.26.22283900; Stevenson RE et al. Multisite evaluation and validation of Optical Genome Mapping for prenatal genetic testing. medRxiv 2022.12.19.22283552; doi: https://doi.org/10.1101/2022.12.19.22283552; Pang A et al. Clinical Validation of Optical Genome Mapping for the Detection of Structural Variations in Hematological Malignancies. medRxiv 2022.12.27.22283973; doi: https://doi.org/10.1101/2022.12.27.22283973

OGM Provides Improvements as Compared to Classical Cytogenetics

Variant Type	Karyotype	FISH	CMA	OGM
Aneuploidy	✓	✓ Targeted	✓	✓
Deletion	✓ >5–10Mbp	✓ Targeted	✓	✓
Duplication	✓ >5–10Mbp	✓ Targeted	✓	✓
Translocation	✓ >5–10Mbp	✓ Targeted	✗	✓
Inversion	✓ >5–10Mbp	✓ Targeted	✗	✓
AOH	✗	✗	✓	✓ Germline
Repeat Expansion	✗	✗	✗	✓ Limited to large repeats
Repeat Contraction	✗	✗	✗	✓
SNV	✗	✗	✗	✗

Broeckel U et al., medRxiv 2022.12.26.22283900; doi: https://doi.org/10.1101/2022.12.26.22283900; Stevenson RE et al. medRxiv 2022.12.19.22283552; doi: https://doi.org/10.1101/2022.12.19.22283552; , Pang A et al.. medRxiv 2022.12.27.22283973; doi: https://doi.org/10.1101/2022.12.27.22283973

“Optical genome mapping has uncovered previously hidden structural variation that has been inaccessible largely because of the limitations and low resolution of the standard technologies that we relied upon in the past.“

Brynn Levy
Professor, Department of Pathology & Cell Biology
at College of Physicians and Surgeons
Columbia University

bionano

“30% of previously unsolved cases for B-ALL, which previously underwent karyotype + FISH + microarray + NGS, were solved using OGM.”

Dr. Gordana Raca
Children’s Hospital Los Angeles
California, USA

“With OGM we are changing subjectivity to objectivity in going from a visual microscope-based karyotype to a high-resolution digital output.”

Dr. Adam C. Smith
UHN Toronto
Toronto, Canada

Multisite Validation Study with OGM in Hematological Malignancies

OGM results highly concordant with SOC + OGM led to MORE REPORTABLE VARIANTS

207	**Datasets evaluated** (including replicated), from: • 68 Hematological malignancies samples • 27 controls • 2 cancer cell lines
100%	100% Concordance with SOC 100% Sensitivity 100% Specificity 100% Accuracy 100% Precision 100% PPV 100% NPV
96%	Reproducibility
37%	Cases for which OGM was able to find additional significant variants
5% to 10%	**Limit of Detection:** ~ 5% for Structural Variants ~ 10% for CNVs At 1.5Tbp (≥300× post-analytical effective coverage)

Different Sites Involved:

bionano laboratories

University of Rochester Medical Center

Augusta University Medical College of Georgia

Children's Hospital Los Angeles

> The results of this validation study demonstrate OGM's superiority over traditional SOC methods for the detection of SVs for the accurate analysis of various hematological malignancies.
>
> OGM ability to detect SVs at high resolution and high sensitivity holds promise for OGM to be a first tier cytogenomic assay for heme cancers.

SOC: standard-of-care

Pang A et al. Clinical Validation of Optical Genome Mapping for the Detection of Structural Variations in Hematological Malignancies. medRxiv 2022.12.27.22283973; doi: https://doi.org/10.1101/2022.12.27.22283973

Multisite Validation Study with OGM in Hematological Malignancies

OGM results highly concordant with SOC + OGM led to MORE REPORTABLE VARIANTS

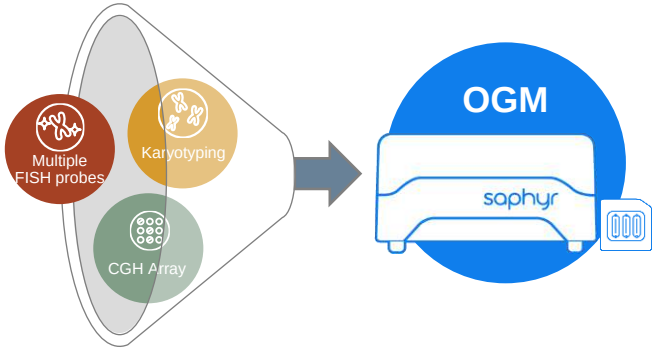
OGM Performance compared to SOC

High analytical concordance
100% concordance with SOC

Better characterization and resolution of SVs and CNVs
highest resolution and sensitivity attainable till date

Revealed novel, pathogenic SVs not reported by SOC
37% of the cases contained **novel** clinically significant SVs not reported by SOC

OGM workflow allows consolidation and simplification, leading to cost and time savings for the lab



Combination of karyotyping, multiple FISH probes, and arrays are typically used to obtain information. **Many aberrations are cryptic to these methods**. **OGM** helps **consolidate** and **simplify** workflow, while also **maximizing findings**.

Pang A et al. Clinical Validation of Optical Genome Mapping for the Detection of Structural Variations in Hematological Malignancies. medRxiv 2022.12.27.22283973; doi: https://doi.org/10.1101/2022.12.27.22283973

Multisite Prospective and Retrospective Study with OGM in Postnatal

OGM results highly concordant with SOC + OGM led to MORE REPORTABLE VARIANTS

>1,000	**Datapoints collected in this large study**, and compared to SOC (Karyotyping, FISH, microarray, Southern Blotting and PCR)
99.6%	Full or partial **concordance achieved between OGM and SOC results** (98.7% fully concordant)
10.1% to 14.8%	**Incremental variants reported**, when OGM was combined with SOC (10.1% gain in prospective cases; 14.8% in Autism Spectrum Disorder cases)

Many US sites involved in study:

University of Rochester Medical Center

University of Iowa Hospitals & Clinics
University of Iowa Health Care

Medical College of Wisconsin

Columbia
Columbia University Irving Medical Center

Praxis Genomics

Augusta University
Medical College of Georgia

Greenwood Genetic Center

Moffitt Cancer Center

> The increase in reportable yield by OGM was directly attributable to **genomic aberrations that were not discovered using SOC methods.**

SOC: standard-of-care

Broeckel U et al. Multisite Study of Optical Genome Mapping of Retrospective and Prospective Constitutional Disorder Cohorts. medRxiv 2022.12.26.22283900; doi: https://doi.org/10.1101/2022.12.26.22283900

Modern Cytogenomic Analysis. Lower Operational Burden.

Reveal more variation that matters

Create an objective, high-resolution digital karyotype

1000x resolution with automated data analysis and interpretation tools

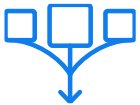
>99% concordance with traditional techniques

Multiple studies demonstrate OGM's ability to accurately and reliably identify chromosomal aberrations found by karyotyping, FISH and CMA

Reveal more genetic variation that matters

Analyze all guideline-recommended biomarkers and drive discovery with identification of novel findings undetected by traditional techniques

Simplify and scale your laboratory operations



Streamline your workflow

Single, alternative workflow to karyotyping, CMA and FISH that can simplify comprehensive chromosomal analysis

Lower your resource burden

Onboard new staff members faster, reduce the need for cell culture, and lower your hands-on time overall

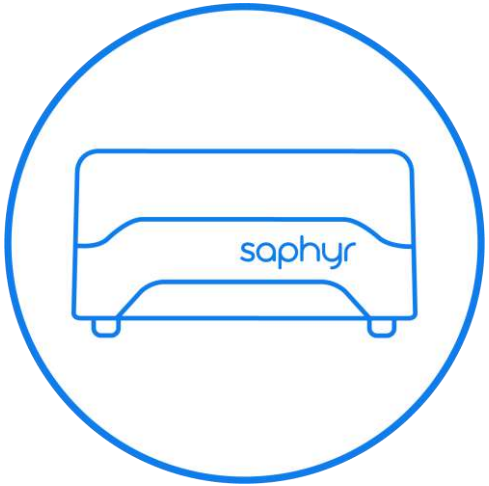
Sample to answer in <1 week

Eliminate complex reflex workflows and find the answers you want in less time

Next Steps

Consultative Discussion

Dive deeper on OGM with a Technical Specialist specific to the needs in your lab.

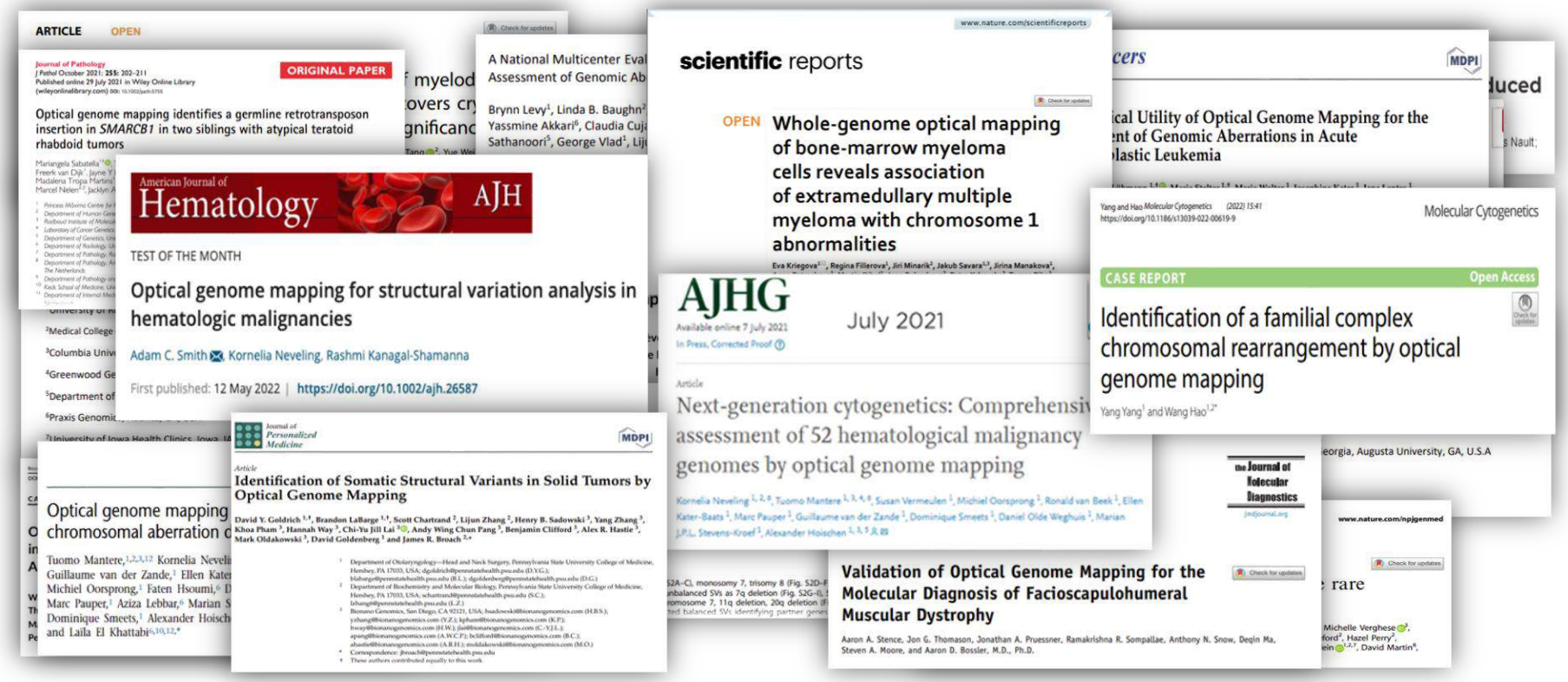


Data Services Project

See what OGM can do with your samples in our lab.

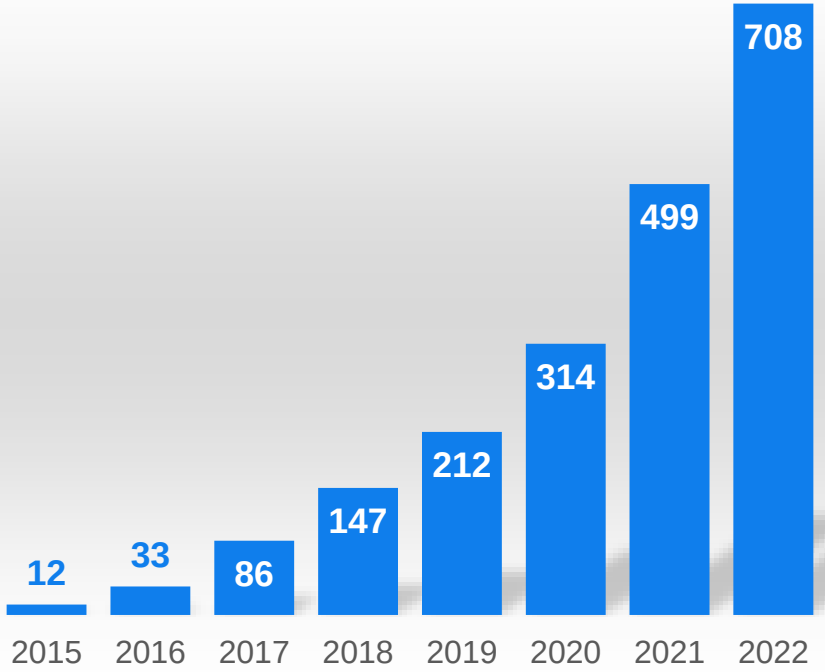
Multiple Institutions Have Benefited from OGM

Numerous Publications in Oncology and Constitutional Space



Sample of some key publications, not representative of entire reference library.

Growing number of Bionano OGM publications



Sample of some key publications, not representative of entire reference library.

How We Reveal Structural Variants: Optical Genome Mapping on the Bionano Saphyr® System

bionano™

A End-to-End Solution for Optical Genome Mapping

Sample-to-Answer in as few as 4 days with Sample Preparation, Data Generation, and Analysis

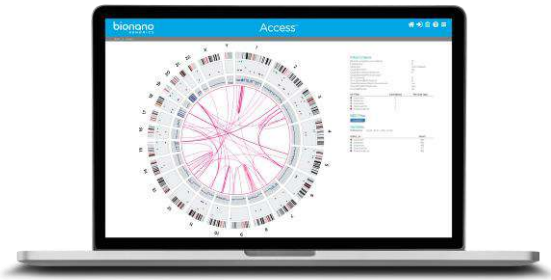
Sample Preparation
(SP and DLS reagent kits)

2 – 3 days

OGM Data Generation
(the Saphyr® System)

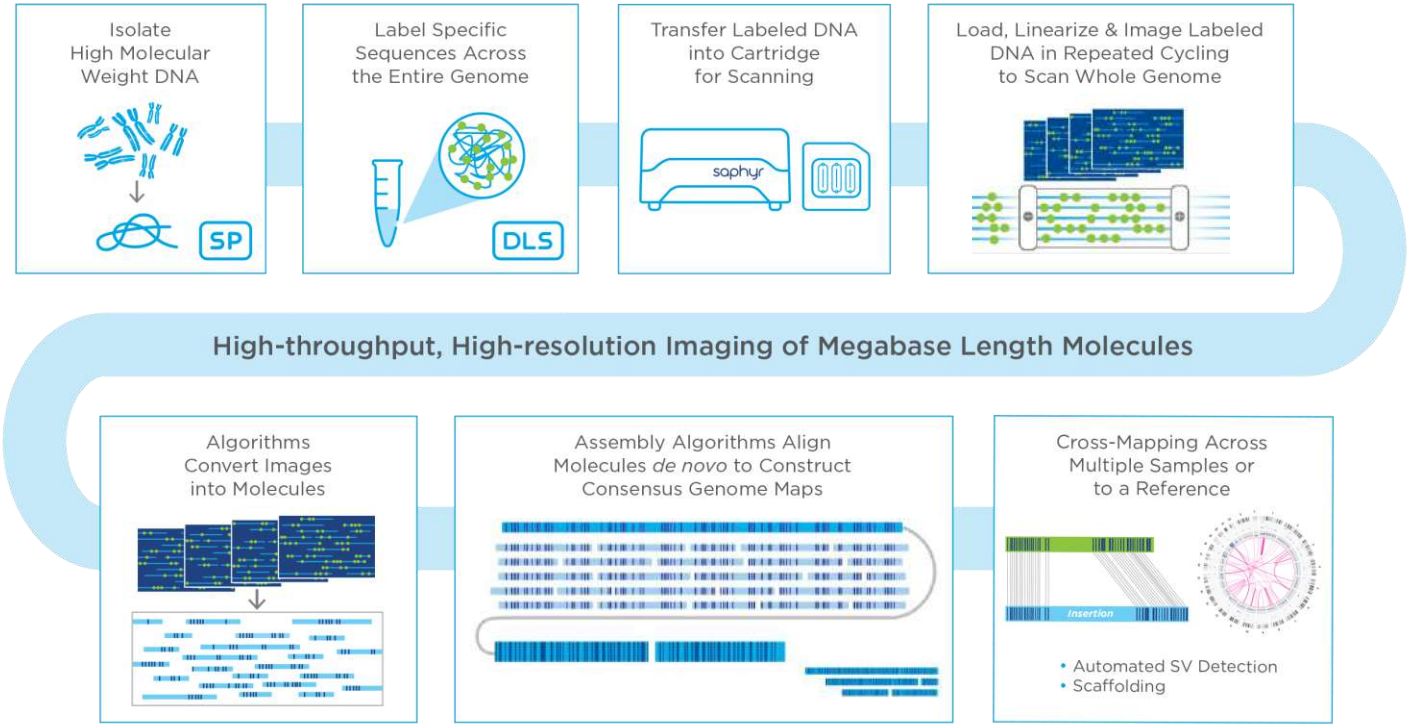
1 - 2 days

Data analysis and Interpretation
(Access and Solve software)
(VIA – coming soon!)



1 - 2 days

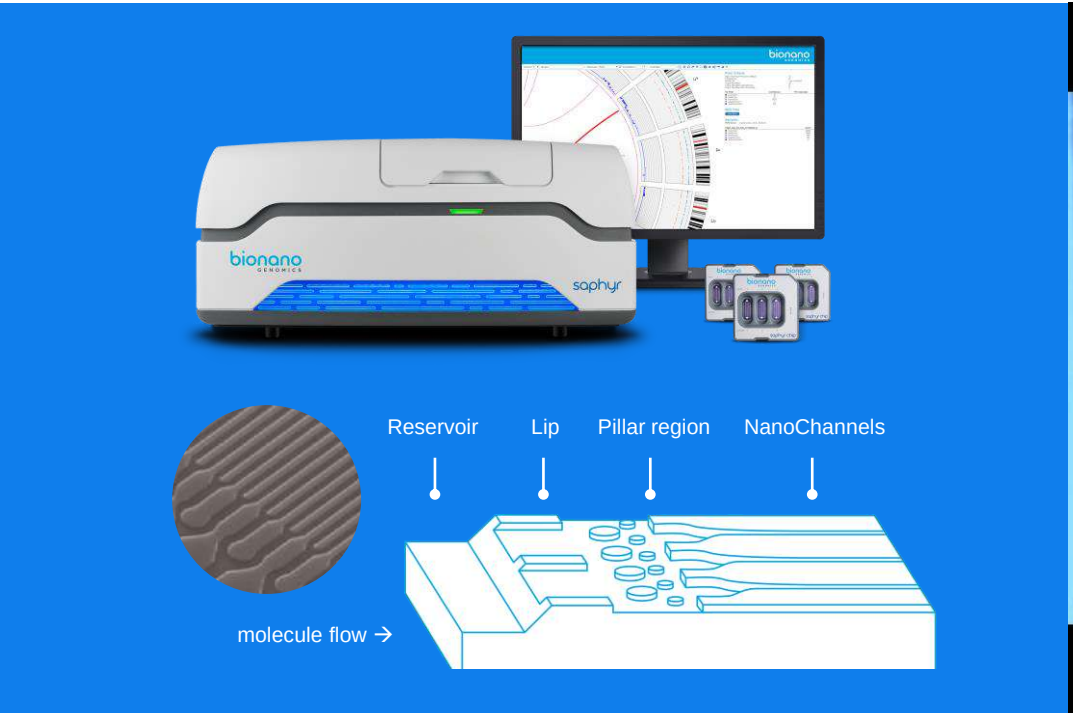
Overview of the Saphyr OGM Workflow

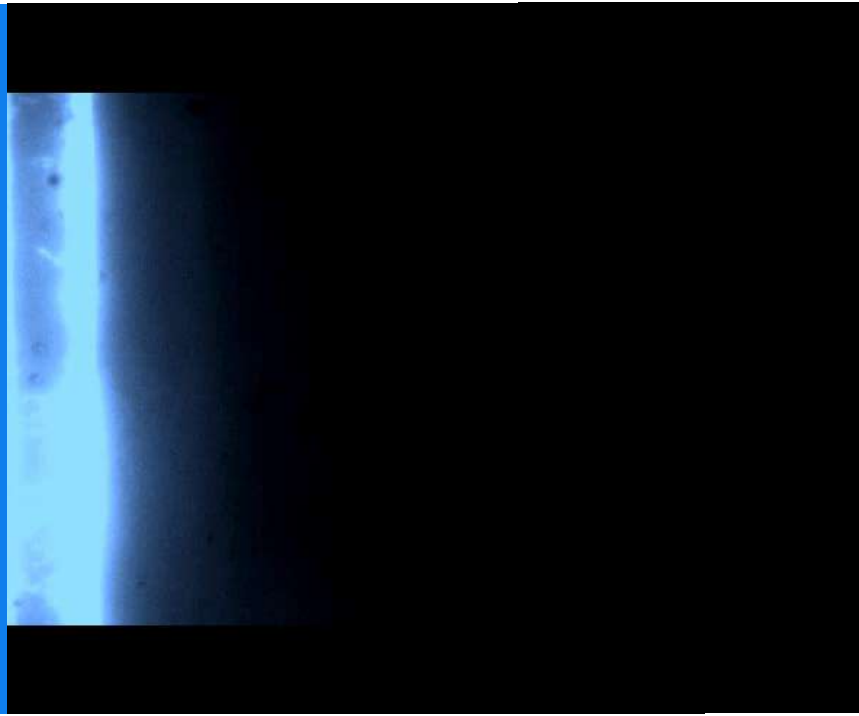


Sample Types (fresh or frozen)

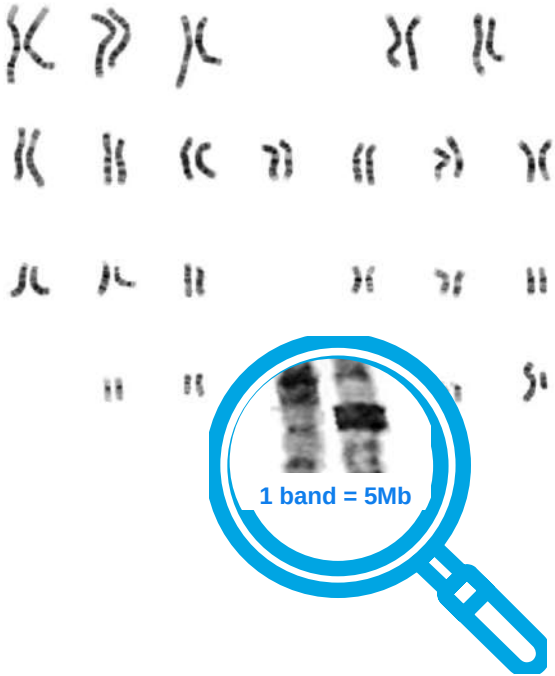
- Blood (650 µl)
- Bone marrow aspirate
- Cultured cells
- Tissue
- Tumor (5mg)
- Cultured amniocytes or CVS samples (1M)

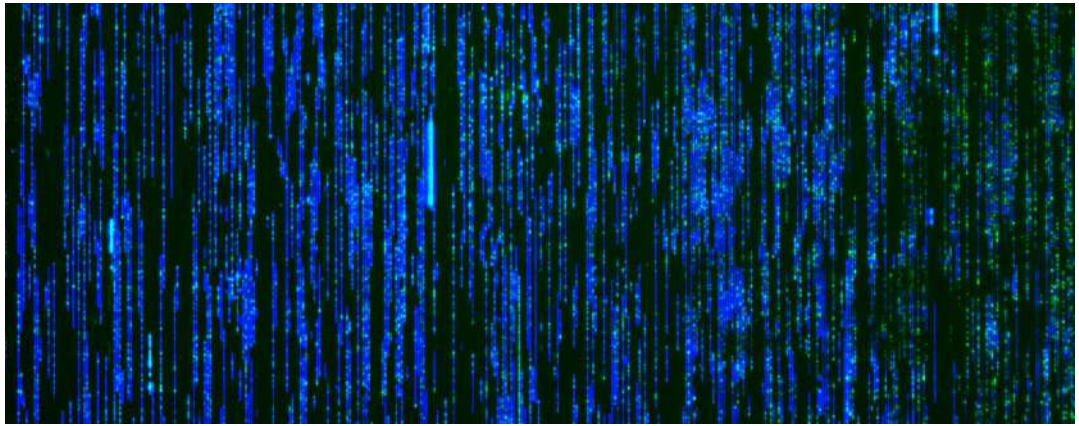
OGM with Saphyr Uses Nanochannel Arrays to Linearize Ultra-High Molecular Weight DNA





Optical Genome Mapping as High-Resolution Digital Karyotyping





OGM provides

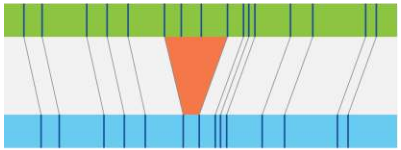
~1,000x times more "bands" in the form of labels and can detect chromosomal aberrations as small as 500 bp

10,000x resolution

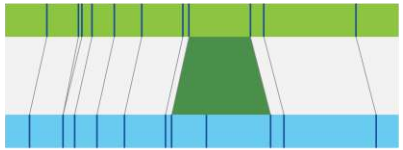
All Classes of Structural Variants Can Be Detected by OGM on the Saphyr System

Gain/Loss

Deletion

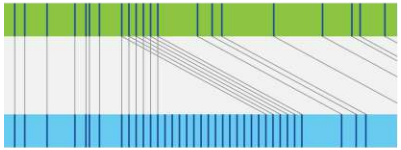


Insertion

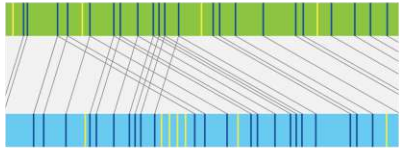


Copy Number Change

Repeat array expansion

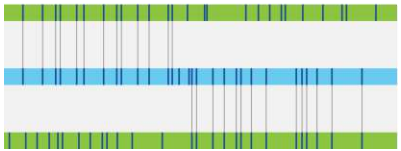


Tandem duplication

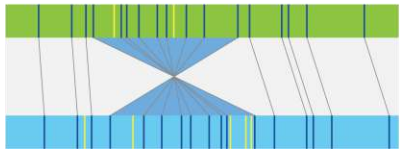


Balanced

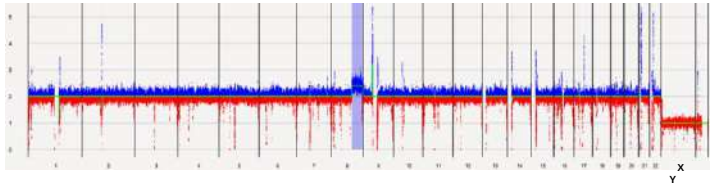
Translocation



Inversion

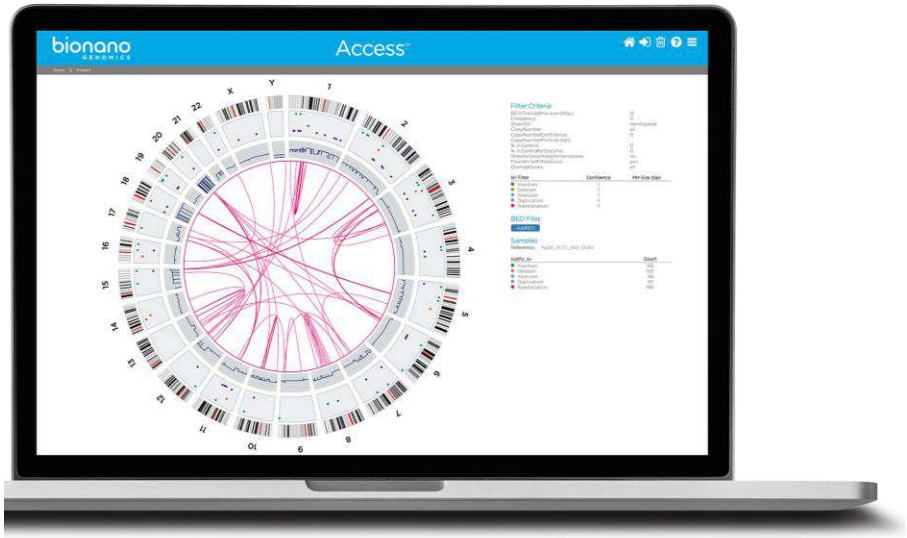


CNV



- Compared to karyotyping, OGM offers much higher resolution in a fully digitized analysis workflow.
- Compared to CMA, OGM closes the gap on balanced rearrangements, including translocations and inversions.

Circos Plot Allows Visualization of Chromosomal Aberrations



Bionano Access™ Software

Visualization Options

- Circos plot
- Genome Browser

Report

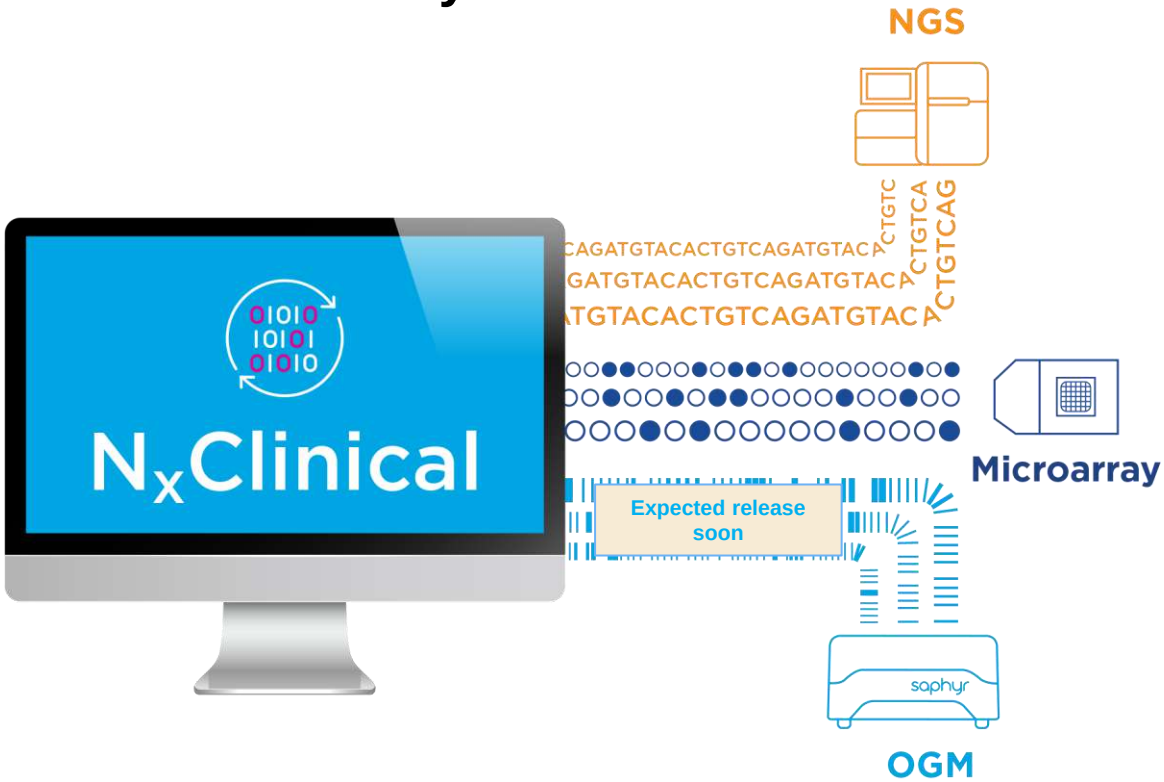
- Export SVs as VCF file and/or PDF report

Filtering Options

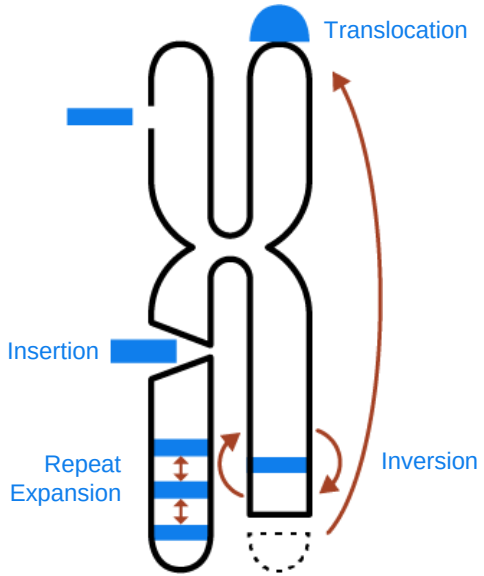
- Size, type of event, confidence score
- Control database
  (>300 healthy individuals)
- Gene panel or genomic coordinates

The only software decision support tool for detection of copy number variants from NGS and array data.

- Enables accurate, efficient variant interpretation from a single screen view
- Visualizes multiple tracks of genomic data for at-a-glance context of variant calls
- Releases continuous, community-driven updates that match the pace of innovation in genomics



OGM Performance



Application	Germline DNA Analysis	Cancer Analysis
Data collected	400 Gbp	1.5 Tbp
Coverage setting	100x	400x
Effective coverage	80x	300x
Variant allele frequency	≥50%	≥5%
Analysis pipeline	*De Novo* Assembly	Rare Variant Analysis
Resolution by variant type at >90% sensitivity		
Insertions	>500 bp	>5 kbp
Deletions	>700 bp	>7 kbp
Stable Repeat Expansion/Contractions	>500 bp	>5 kbp
Duplications	>30 kbp	>150 kbp
Translocations	>70 kbp	>70 kbp
Inversions	>30 kbp	>70 kbp

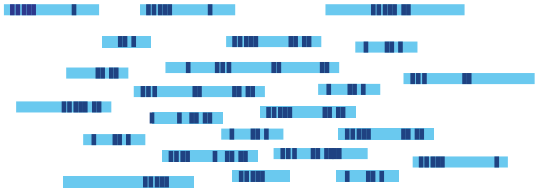
Based on Internal Findings. Bionano data on file.

Bioinformatics Pipeline

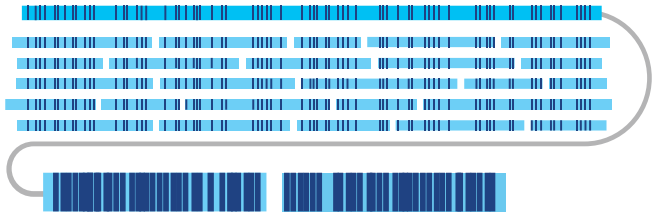
The *De Novo* Pipeline Calls Structural Variants by Comparing Maps to a Reference

- Whole Genome Assembly
- Detects variants 500bp and ≥50% variant allele frequency
- Recommended for constitutional samples

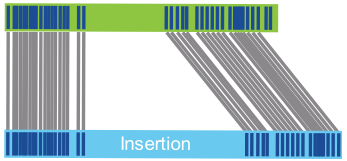
1 **Pairwise alignment of all molecules**



2 **Aligned molecules create a consensus map**



3 **Comparison to a reference map**

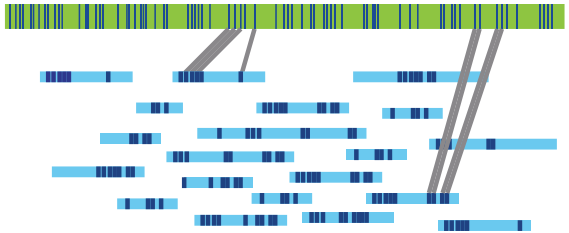


Bioinformatics Pipeline

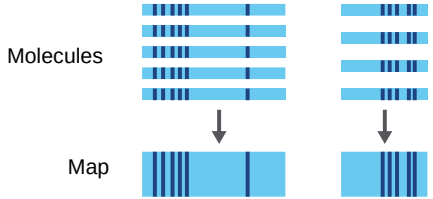
The Rare Variant Analysis Pipeline Calls Structural Variants by Comparing Maps to Each Other

- Single molecule tool
- Detects variants 5 kbp and at >5% variant allele fraction
- Recommended for cancer/mosaic samples

1 **Single molecule alignment to a reference**



2 **Molecules with SVs are clustered**



3 **Local alignment to confirm SV**

Additional Publications

bionano

Genetic Disease - Postnatal

bionano

Multisite Prospective and Retrospective Study with OGM in Postnatal

OGM results highly concordant with SOC + OGM led to MORE REPORTABLE VARIANTS

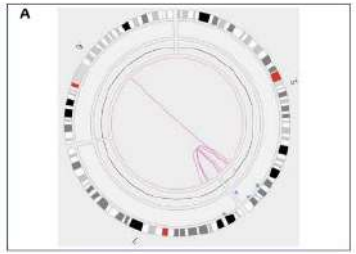
Examples of **NOVEL** findings by OGM:

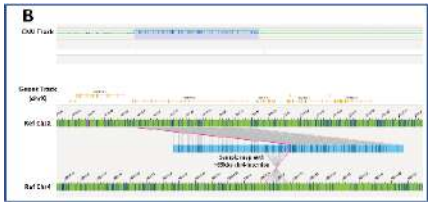
UNCLASSIFIED RETROSPECTIVE CASE (DEVELOPMENTAL DELAY, HYDROCEPHALUS, ANOPHTHALMIA)

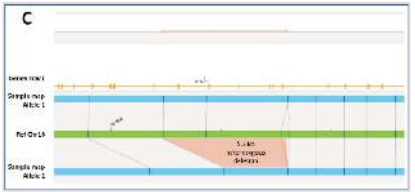
- Karyotyping results incomplete.
- OGM revealed a more complex rearrangement involving 3 chromosomes, copy number loss, several translocation breakpoints and intrachromosomal foldback junctions.

RETROSPECTIVE AUTISM SPECTRUM DISORDER CASE, WITH NORMAL SOC. OGM REVEALED:

- A heterozygous 5.5 kbp deletion in *AP1G1*, size and boundary which suggested impact to exons 11-12 or 12-13.
- Subsequent analysis with qPCR confirmed deletion of exons 11 and 12.
- *AP1G1* is related to autosomal dominant Usmani-Riazuddin syndrome





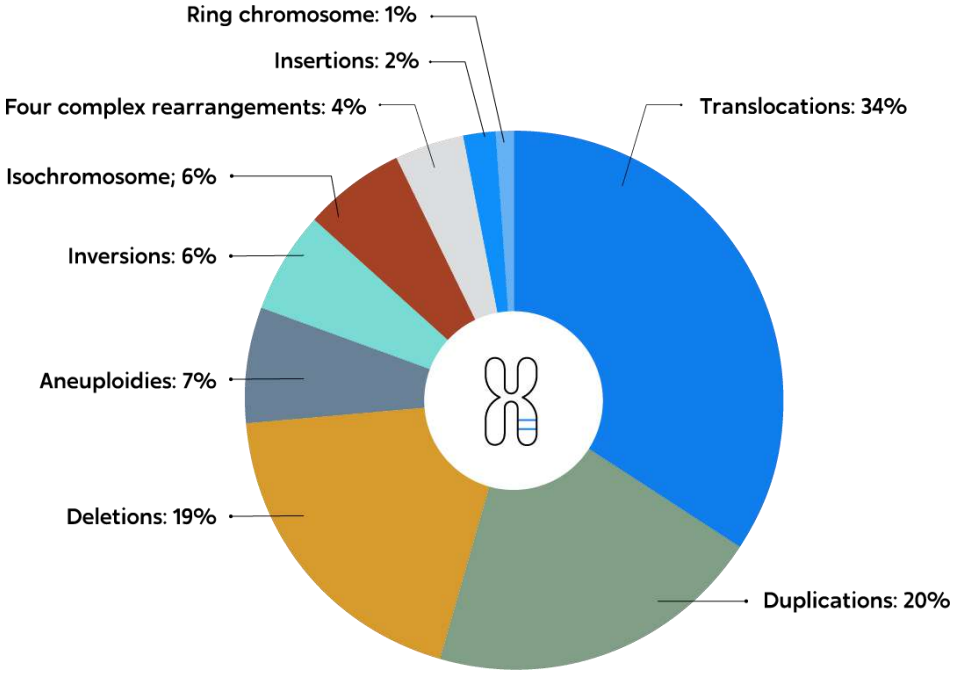


PROSPECTIVE DEVELOPMENTAL DISORDER CASE. OGM REVEALED:

- Xp22.12 duplications structure with a 69 kbp duplication from chr4 inserted in between the 390 kbp tandemly duplicated segment (chrX:19,547,445-19,937,089).
- The inserted segment from chr4 contains the entire *TLR3* gene and exon 1 of *FAM149A*, and its orientation and position supports a potential fusion with *BCLAF3* on chrX.

Broeckel U et al. Multisite Study of Optical Genome Mapping of Retrospective and Prospective Constitutional Disorder Cohorts. medRxiv 2022.12.26.22283900; doi: https://doi.org/10.1101/2022.12.26.22283900

OGM Shows 100% Concordance with SOC in European Consortium Constitutional Cohort Study



n=85 Standard of care method: array-based technology and karyotyping

Tuomo Mantere, et al. "Optical genome mapping enables constitutional chromosomal aberration detection." The American Journal of Human Genetics,Volume 108, Issue 8, 2021, pp 1409-1422, ISSN 0002-9297, https://doi.org/10.1016/j.ajhg.2021.05.012.

Case Study: OGM Detects a Pathogenic 410bp Duplication & Insertion

Not Observed with Previous Analysis by Multiple Methods

No findings from first line assessments and advanced Methods…

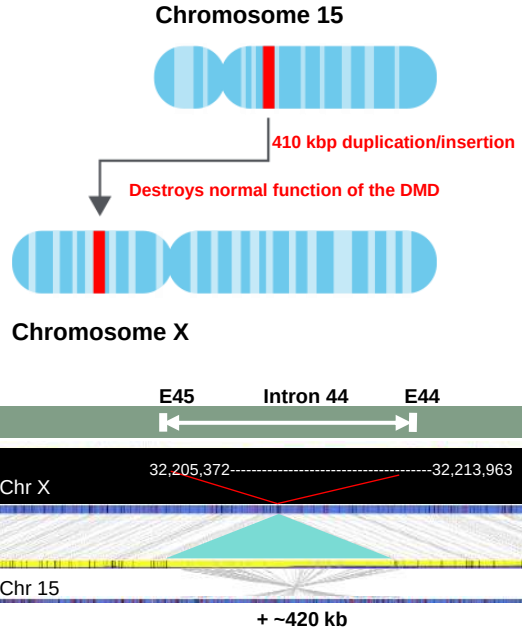
- Chromosomal Microarray
- PCR and Sanger Sequencing
- Multiplexed Ligation Polymorphism Assay (MLPA)
- Whole Exon & Whole Genome Sequencing

Children's National

Results from Dr. Eric Vilain – publication pending.

…OGM Explained the Molecular Basis of the Disease

Detected a 410kbp duplication & insertion in the DMD gene, resulting in out-of-frame transcription, consistent with Duchenne Muscular Dystrophy*



Genetic Disease - Prenatal

bionano

Multisite Evaluation and Validation of OGM in Prenatal Cases

In this study OGM provided, in a single assay, results 100% concordant with 2-3 SOC methods combined

200	**Datapoints across 123 prenatal samples** evaluated, and compared to Standard of Care methods (Karyotyping, FISH, microarray)
100%	OGM results: • **100% concordance** with SOC (for samples and variants) • **100% reproducibility** across 9 different sites, different operators and different instruments
75%	75% of the cases originally required 2 or 3 assays to generate the results, while **OGM led to same results with a single OGM assay!**

Different Sites Involved:

Columbia University Irving Medical Center · University of Rochester Medical Center · Cincinnati Children's · Greenwood Genetic Center · UCSF University of California San Francisco · Augusta University Medical College of Georgia · Brigham Health Brigham and Women's Hospital · Quest Diagnostics

> This study demonstrates that while three sequential cytogenetic assays were used to determine genomic structure, OGM accurately predicted the structure **in a single sample-to-answer assay with 100% concordance and 100% reproducibility.**

Stevenson RE et al. Multisite evaluation and validation of Optical Genome Mapping for prenatal genetic testing. medRxiv 2022.12.19.22283552; doi: https://doi.org/10.1101/2022.12.19.22283552

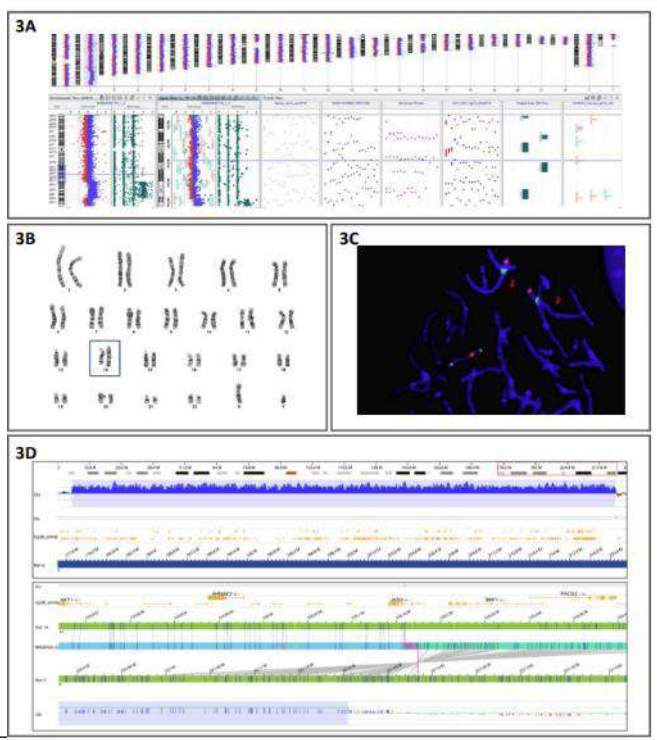
Multisite Evaluation and Validation of OGM in Prenatal Cases

In this study, OGM provided, in a single assay, results 100% concordant with 2-3 SOC methods combined

The ability to **quickly determine** the complexity of genomic aberrations is **critical for prenatal** analysis.

As shown in this example, in multiple cases, OGM resolved **multiple sequential tests in one assay!**

> OGM has the potential to become a first line assay as it would have likely **reduced the time** to attain comprehensive answers at a **reduced cost.**



Stevenson RE et al. Multisite evaluation and validation of Optical Genome Mapping for prenatal genetic testing. medRxiv 2022.12.19.22283552; doi: https://doi.org/10.1101/2022.12.19.22283552

Large-Scale Study Shows 100% Concordance Between OGM and Cytogenetics in Prenatal Cohort with Additional Findings

AUGUSTA UNIVERSITY
MEDICAL COLLEGE OF GEORGIA

94 amniocentesis samples previously characterized by:

- Karyotyping
- FISH
- CMA

Kolhe, et al. *medRxiv* (2022 May 16.)
https://doi.org/10.1101/2022.05.11.22274975

100%

OGM concordance with classical cytogenetics results

Chromosomal aberrations detected include aneuploidies, triploidy, deletions, duplications, translocations, isochromosomes, and LOH

101

Number of genetic aberrations identified

Resolution of the origin of 3 marker chromosomes and additional unknown material on another chromosome

46%

Samples with additional actionable findings detected

OGM detected 64 additional clinically reportable SVs in 43 samples

> We report the feasibility and relative ease of implementing OGM for prenatal diagnostic testing compared to SOC methods with the platform demonstrating high robust technical and analytical performance and recommend OGM as a first-tier test in prenatal settings.

OGM Enables Robust Detection of Cryptic Balanced Chromosomal Rearrangements in Recurrent Pregnancy Loss (RPL)

11 couples investigated for recurrent pregnancy loss (RPL)

100%

Cryptic events successfully identified by OGM

Includes 10 balanced reciprocal translocations and 1 unbalanced translocation

100%

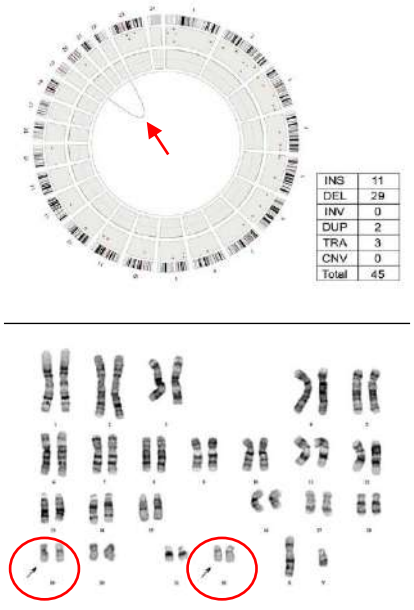
Confirmation with both FISH and sequencing analyses

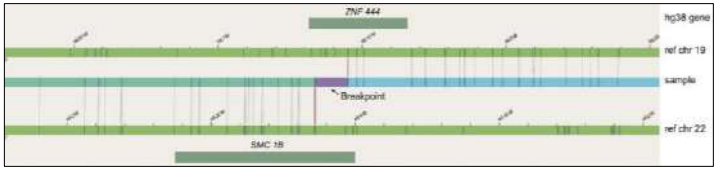
With further refinement of cryptic karyotypes

3

Rearrangements identified with highly repetitive sequences

Otherwise difficult to detect with NGS





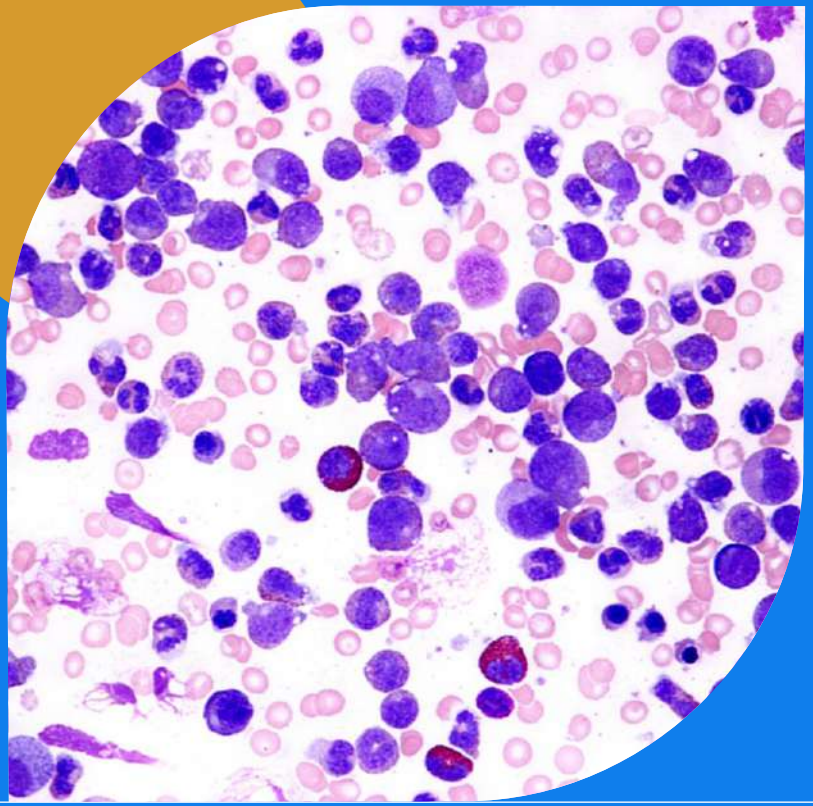
Example of balanced translocation detected by OGM. **Top left:** Circos plot view. **Top right:** Karyotype for which this event was cryptic. **Bottom:** map view.

> With the excellent performance, our research suggests that OGM can be used as a first-line method for detecting cryptic balanced chromosome rearrangements…

GENETICS & IVF INSTITUTE 集爱 SHANGHAI JI AI

Zhang S, Pei Z, Lei C, *et al* Detection of cryptic balanced chromosomal rearrangements using high-resolution optical genome mapping. *Journal of Medical Genetics* Published Online First: 16 June 2022. doi: 10.1136/jmedgenet-2022-108553

Oncology – Hematological Malignancies



bionano™

Large Body of Evidence Shows OGM has high-concordance with traditional cytogenetic methods, while unveiling more pathogenic SVs

Reference	Cohort Size	Clinical Referral	Number of Abnormalities Included (Soc)	Concordance with Cytogenetics Results	OGM Additional Findings
Radboud University Neveling et al., 2020	48	AML, MDS, CML, CLL, ALL, MM, MPN, T-PLL, LYBM	112	100%	18 potential gene fusions absent from COSMIC database. 26 insertions/deletions overlapping with well-established cancer genes
Cancer Genomics Consortium Levy et al., 2020	100	AML	NA	100%	3 translocations, 1 inversion, 2 deletions and 1 derivative chromosome
CHU Amiens Lestringant et al., 2021	10	B and T ALL	78	97%	4 fusions, 6 deletions, 2 gains, 1 duplication, 3 complex chromosomal rearrangements
Johns Hopkins University Stinnett et al. 2021	5	Leukemia/Lymphoma and Solid Tumors	30	100%	71 additional calls (7.7% involving cancer genes)
University Hospital Olomouc Kriegova et al. 2021	11	Multiple myeloma	NA	98%	
Augusta, Emory Sahajpal et al. 2022	69	CLL, AML, MDS, MM, lymphoma, PCM, CML, ET and others	164	99%	OGM detected chromosomal aberrations missed by karyotyping and FISH in 35 cases
Hannover Luehmann et al. 2022	12	ALL	NA	~98%	Many new and unknown SVs including gene fusion of JAK2 and NPAT
M.D. Anderson Yang et al., 2022	101	MDS	194	99%	OGM identified 224 cryptic, clinically significant SVs in 34% of pts.
TOTAL	**356**	**VARIOUS**	**>578**	**>99%**	

2022 Comprehensive Study by Augusta University

OGM outperformed classical methods

59 myeloid and lymphoid neoplasms + 10 controls (AML, CLL, MDS, MM, Lymphoma, MPD/MPN, CML)

Compared to Karyotyping, FISH, CMA

Performance Metric Evaluation of OGM for Hematological Neoplasms	
Performance Criteria	**Overall SVs (n=164)**
Sensitivity	**98.7%**
Specificity	**100%**
PPV/Precision	**100%**
NPV	**98%**
Accuracy	**99.2%**

- Sensitivity/positive percentage agreement = true positive/(true positive + false negative).
- Specificity/negative percentage agreement = true negative/(true negative + false positive).
- PPV = true positive/(true positive + false positive).
- NPV = true negative/(true negative + false negative).
- Accuracy = true positive + true negative/all results.
- NPV, negative predictive value; PPV, positive predictive value; SV, structural variation.

AUGUSTA UNIVERSITY
MEDICAL COLLEGE OF GEORGIA

- **LOD: 5%** allele fraction for aneuploidy, translocation, interstitial deletion, and duplication
- OGM identified **several additional structural variations,** refined breakpoints, and corrected interpretations

> Overall, OGM has outperformed the classical methods in this study and demonstrated its potential as a first-tier cytogenomic assay for hematologic malignancies.

Sahajpal NS, et al. Clinical Validation and Diagnostic Utility of Optical Genome Mapping for Enhanced Cytogenomic Analysis of Hematological Neoplasms. J Mol Diagn. 2022 Oct 17:S1525-1578(22)00290-2. doi: 10.1016/j.jmoldx.2022.09.009.

Multi-Center Study from Cancer Genomics Consortium

CANCER GENOMICS CONSORTIUM

OGM 100% Concordant with classical methods + found additional actionable pathogenic aberrations

100	100 AML cases previously evaluated with one or more classical cytogenetic methods (karyotyping, FISH, CMA)
100%	OGM detected 100% of all key pathogenic SVs and CNVs previously identified by traditional cytogenetic methods, with LOD of 5% allele fraction
13%	Cases had additional pathogenic findings identified by OGM
12%	Cases for which OGM findings would have altered ELN risk-level or identified eligibility for clinical trials

COLUMBIA
COLUMBIA UNIVERSITY IRVING MEDICAL CENTER

Quest Diagnostics

NATIONWIDE CHILDREN'S

PennState College of Medicine

CASE WESTERN RESERVE UNIVERSITY
think beyond the possible

AUGUSTA UNIVERSITY
MEDICAL COLLEGE OF GEORGIA

labcorp

Seattle Cancer Care Alliance
Fred Hutch · Seattle Children's · UW Medicine

PathGroup
Physician Centered. Patient Focused.

Fred Hutch Cancer Center

MAYO CLINIC

THE UNIVERSITY OF TEXAS
MD Anderson Cancer Center
Making Cancer History®

Levy et al.; Optical Genome Mapping in Acute Myeloid Leukemia: A Multicenter Evaluation. *Blood Adv* 2022; bloodadvances.2022007583. doi: https://doi.org/10.1182/bloodadvances.2022007583

Multi-Center Study from Cancer Genomics Consortium

CANCER GENOMICS CONSORTIUM

OGM 100% Concordant with classical methods + found additional actionable pathogenic aberrations

OGM findings that could have altered risk-level or led to eligibility to clinical trials

Sample	Significant Cytogenetic Findings From Karyotyping	Significant Cytogenetic Findings From OGM	Change in ELN	Clinical Trial Opportunity
1	t(8;21)	Atypical RUNX1::RUNX1T1 , del(5)		
2	Simple karyotype	CBFB:: MYH11		
3	Normal karyotype	ETV6::MECOM		
4	Simple karyotype	7q deletion, RUNX1 deletion		
5	Normal karyotype	NUP98::NSD1		
6	Normal karyotype	NUP98::NSD1		
7	Simple karyotype	KMT2A::MLLT3		
8	del(17p)	Complex		
9	Mono 17, monosomal karyotype	Complex		
10	del(5q), del(17p)	del(5q)		
11	del(5q), del(17p)	del(5q), del(17p), KMT2A r		
12	del(5q), mono 7, del(17p)	del(5q), mono 7		

Levy et al.; Optical Genome Mapping in Acute Myeloid Leukemia: A Multicenter Evaluation. *Blood Adv* 2022; bloodadvances.2022007583. doi: https://doi.org/10.1182/bloodadvances.2022007583

“

The results from this multi-institutional study indicate that OGM effectively recovers relevant SVs and CNVs found by standard of care methods and reveals additional SVs not reported.

Furthermore, OGM minimizes the need for labor-intensive multiple cytogenetic assays while concomitantly maximizing detection through a standardized workflow.

Oncology – Solid Tumors

bionano

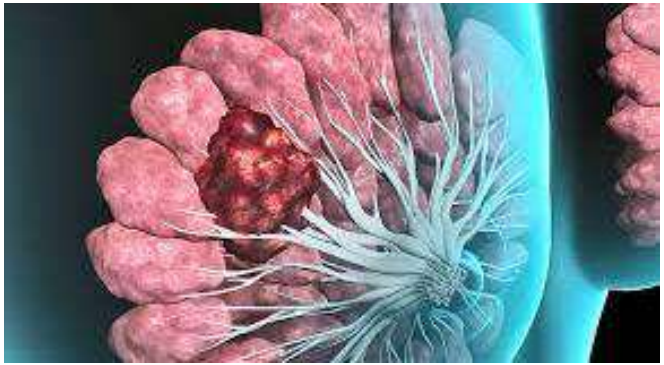
OGM as a tool for Homologous Recombination Deficiency (HRD)

OGM provides high sensitivity and specificity for SV detection and HRD stratification.
More cost effective, and simpler to analyze, as compared to WGS, even in samples with low tumor content

PART II OF THE STUDY:

Comparison between whole-genome sequencing and OGM in an HRD triple negative breast carcinoma (TNBC) sample

institut Curie



100%	Concordance between WGS and OGM, for all structural variant events ≥5kb in size
17	Additional Structural Variants found with OGM only

Our results demonstrate that the OGM technology is an **affordable way of getting an insight of the structural variants** present in **solid tumors, even with low tumoral cellularity.**

It **represents an alternative technology for HRD** analysis, which **should now be evaluated in independent series of tumors of different tissue origins.**

Vanhuele et al. Optical Genome Mapping for detecting Homologous Recombination Deficiency (HRD) in human breast cancers. bioRxiv 2022.12.23.521790; doi: https://doi.org/10.1101/2022.12.23.521790

OGM as a tool for Homologous Recombination Deficiency (HRD)

OGM provides high sensitivity and specificity for SV detection and HRD stratification.
More cost effective, and simpler to analyze, as compared to WGS, even in samples with low tumor content

PART II OF THE STUDY:

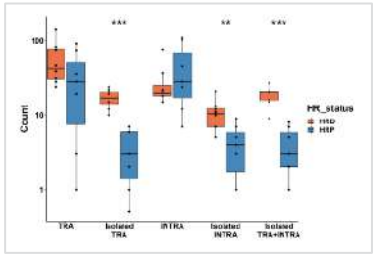
Expanded analysis of OGM performance, for HRD characterization, in a cohort of **15 triple negative breast carcinoma (TNBC) samples:**

- 8 samples were HRD (Homologous Recombination Deficient)
- 7 samples were HRP (Homologous Recombination Proficient)

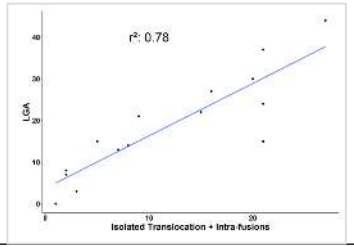
Samples from RadioPARP clinical trial.

RD status was previously detected by low coverage WGS approach

OGM allowed clear stratification of HRD and HRP samples

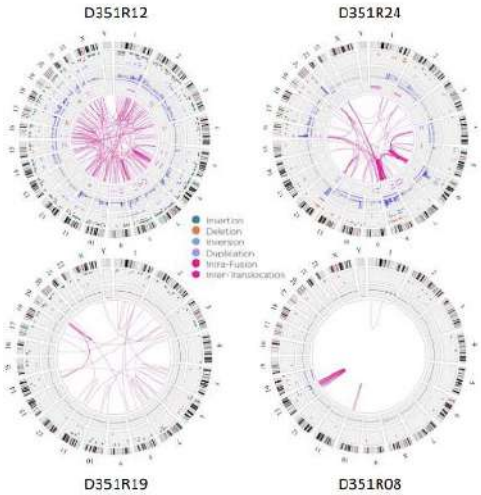


OGM Results:
high correlation to low coverage WGS.
(The only outlier sample was a sample with low tumor content, which may explain why shallow WGS underestimated the amount of large-scale genomic alterations as compared to OGM.)



OGM Circos Plots for 4 representative examples of triple-negative breast carcinomas (TNBC) samples

HRD CASES **HRP CASES**



Vanhuele et al. Optical Genome Mapping for detecting Homologous Recombination Deficiency (HRD) in human breast cancers. bioRxiv 2022.12.23.521790; doi: https://doi.org/10.1101/2022.12.23.521790

Optical Genome Mapping is Powerful for Detecting Pathogenic Structural Variants (SVs) in Solid Tumors

PennState College of Medicine

Several SVs map to known cancer genes, and many would not be readily identified by NGS gene panels

THYROID

17 translocations detected

BLADDER

3 translocations detected

LUNG

358 translocations detected

KIDNEY

36 translocations detected

OVARY

212 translocations detected

BREAST

555 translocations detected

PROSTATE

159 translocations detected

TONGUE

54 translocations detected

BRAIN

321 translocations detected

COLON

2 translocations detected

LIVER

78 translocations detected

Goldrich, D.Y.; LaBarge, B.; Chartrand, S.; Zhang, L.; Sadowski, H.B.; Zhang, Y.; Pham, K.; Way, H.; Lai, C.-Y.J.; Pang, A.W.C.; Clifford, B.; Hastie, A.R.; Oldakowski, M.; Goldenberg, D.; Broach, J.R. Identification of Somatic Structural Variants in Solid Tumors by Optical Genome Mapping. J. Pers. Med. 2021, 11, 142. https://doi.org/10.3390/jpm11020142
For Research Use Only. Not for use in diagnostic procedures.

Combining OGM + WGS led to resolution of complex rearrangements in Hepatocellular Carcinoma

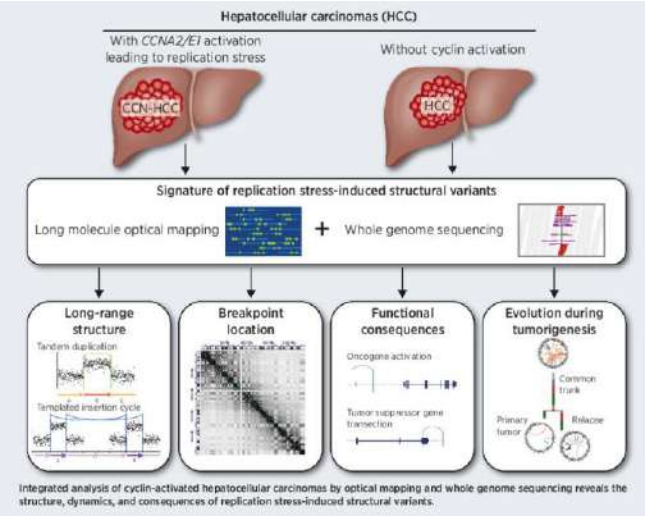
> By combining WGS and optical mapping, **we could reconstruct the structure of complex SVs both at large scale and single-base resolution** in a subgroup of HCC with cyclin-induced replication stress.

SORBONNE UNIVERSITÉ

- OGM and WGS were combined to explore SVs induced by replication stress in cyclin-activated hepatocellular carcinomas
- WGS alone could not resolve complex events involving several distant regions interconnected by abnormal intra/inter-chromosomal junctions

1.4x MORE

SVs were detected by OGM, as compared to WGS alone, ranging from classical tandem duplications to complex tumor initiating cells (TICs) with multiple template-switching events



Integrated analysis of cyclin-activated hepatocellular carcinomas by optical mapping and whole genome sequencing reveals the structure, dynamics, and consequences of replication stress-induced structural variants.

Bayard Q, et al. Impact of Replication Stress–Induced Structural Variants in Hepatocellular Carcinoma. *Cancer Res* 15 April 2022; 82 (8): 1470–1481.
https://doi.org/10.1158/0008-5472.CAN-21-3665

OGM identifies causative variant, solving cancer predisposition mystery in pediatric ATRT cancer

Sanger, MLPA, WES & WGS

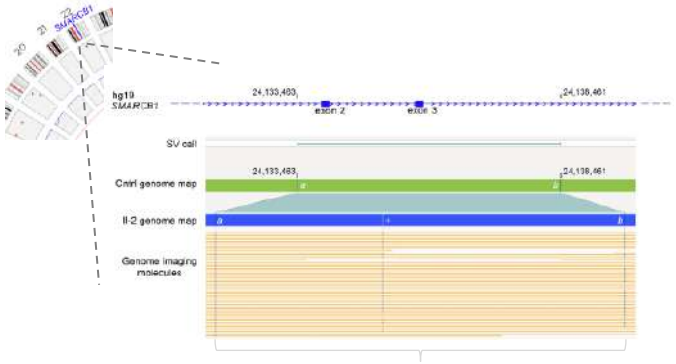
Sanger seq, MLPA, WES and WGS previously used with no driver alteration identified

2.7kb insertion

When OGM was applied, it identified a 2.7kb insertion previously missed, falling into *SMARCB1* gene, a known driver of ATRT

ATRT: Atypical teratoid rhabdoid tumor

> The high GC and repeat content explained why the variant was difficult to detect by NGS and incriminated a retrotransposon that disrupted the *SMARCB1* gene.



Insertion 2.790 bp

Radboudumc university medical center

Princess máxima center pediatric oncology

Sabatella M, et al. Optical genome mapping identifies a germline retrotransposon insertion in SMARCB1 in two siblings with atypical teratoid rhabdoid tumors. J Pathol. 2021 Oct;255(2):202-211. doi: 10.1002/path.5755.

Testimonials

bionano™

OGM reveals more CNVs and SVs, allowing Dr. Dubuc and his team to further their understanding of B-ALL.

Dr. Adrian Dubuc
Harvard Medical School Brigham and Women's Hospital
Massachusetts, USA

“OGM allows next generation cytogenetics and enables the identification of hidden structural variants as a cause of rare diseases.”

Alexander Hoischen, PhD
Radboud UMC
The Netherlands

OGM enables Dr. Lühmann to find novel aberrations in ALL.

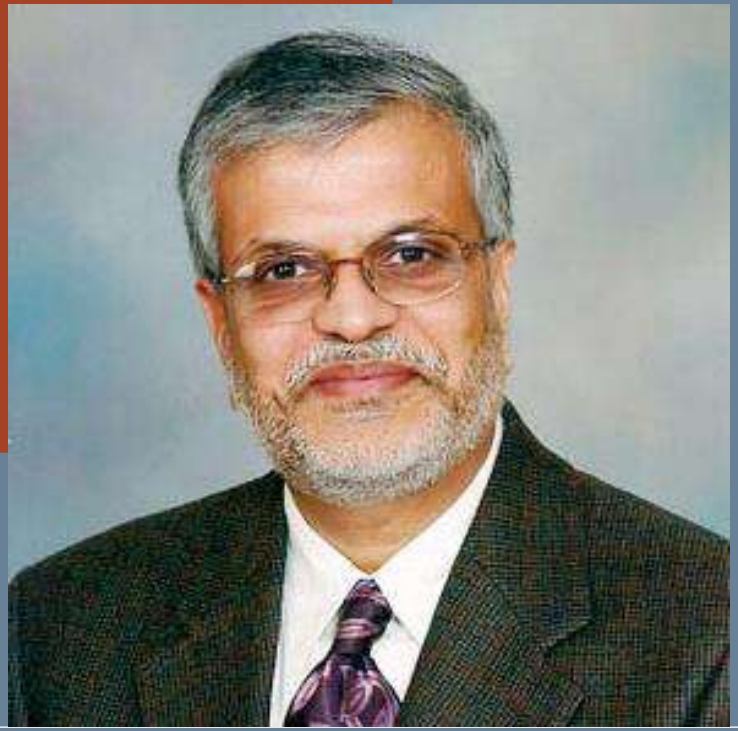
Jonathan Luhmann, PhD Student
Department of Human Genetics
Hannover Medical School

“OGM revealed important translocations, in Ewing sarcoma samples, that could be useful as a prognostic marker for patients with poor clinical outcome.”

Dr. Juan Diaz Martin
Instituto de Biomedicina de Sevilla (IBiS)
Sevilla, Spain

“Balanced translocations, primarily detected by karyotyping for the last 50 years, are solved by OGM.”

Dr. Gopalrao Velagaleti, PhD
UT Health San Antonio
Texas, USA



“The results of the study demonstrate that we are grossly under-evaluating the degree of genomic aberrations.”

Dr. Rashmi Kanagal-Shamanna
MD Anderson Cancer Center
Texas, USA

“OGM reveals more of what matters: more clinically relevant SVs leading to higher success rates and resolution of unsolved cases.”

Dr. Laïla El-Khattabi
Hôpitaux de Paris
(AP-HP)-Université de Paris