bionano GENOMICS

CANCER RESEARCH

HEME MALIGNANCIES AND SOLID TUMOR RESEARCH

Starting at
$450
per genome

Cancer samples are just too complex for low coverage whole genome sequencing. Complex rearrangements as well as unsequenceable regions of the genome present an additional challenge for short and long read sequencing technologies.

Bionano Genome Imaging detects unbiased structural variations at sensitives much higher than sequencing based technologies, and down to 1% variant allele fraction.

Extreme genomic rearrangements are hallmarks of cancer manifestation. Deciphering such complex genomic structures requires a combination of short read sequencing as well as long range information with high coverage to unlock the heterogeneity while providing a complete picture of the genome. To date, NGS applications in the clinic are limited to either low coverage across the whole genome, or high coverage exome sequencing that disregards 98% of the genome. The repetitive nature of the genome makes two thirds largely inaccessible by short-read sequencing, and the short reads make elucidating the complex rearrangements seen in cancer impossible. The throughput of current long-read sequencing technologies is too limited to be realistically applied to cancer diagnostics or discovery, and these technologies remain expensive which is an impediment to getting a true complete view of the rearranged genome.

1% Variant allele fraction detection

1600X Coverage per sample

Bionano Genome Imaging uncovers large structural variations beyond what short and long read sequencing can see.

Bionano Genome Imaging directly visualizes patterns of labels on megabase-size intact DNA molecules, at up to 1600X coverage, to detect structural variations. Every type of structural variant is detected with sensitivities as high as 99%, and with positive predictive value of more than 97%. It can detect balanced translocations, repeat expansions, events flanked by repeats, and even rearrangements of large segmental duplications. Unlike sequencing based methods, that are typically unable to detect insertions or identify where the extra sequence is inserted, Bionano detects both deletions and insertions starting at 500 bp with high sensitivity. And because it uses a single molecule imaging technology, mosaic variants down to as little as 1% variant allele fraction can be detected as well.

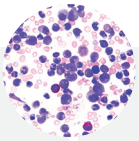
The workflow is simple and starts with mega-base size DNA isolation from blood, cells, tissue or tumor biopsies. A single enzymatic reaction places 500,000 fluorescent labels all throughout the genome at a specific sequence motif occurring approximately 15 times per 100 kbp in the human genome. The long, labeled DNA molecules are linearized in nanochannel arrays on a Saphyr chip® and imaged in an extremely high throughput, automated manner by the Saphyr® Instrument. Changes in patterning or spacing of the labels are detected automatically, genome wide, to call all structural variants.

bionanogenomics.com/cancer

BIONANO CONSOLIDATES KARYOTYPING, FISH AND aCGH IN A SINGLE AUTOMATED ASSAY THAT CAN BE ANALYZED WITH JUST A FEW CLICKS

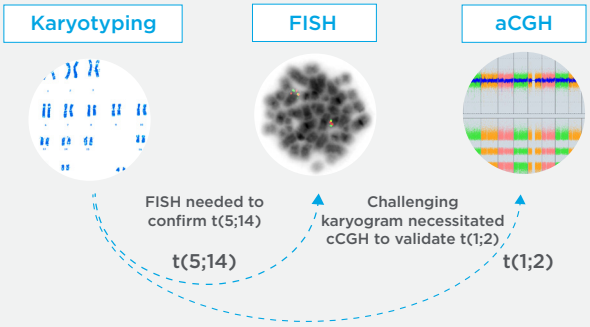
Identification of complex rearrangements in cancer often requires a combination of approaches and are typically performed sequentially. This sequential approach manages costs but increases the turn-around time and complicates understanding the genetic makeup of the sample. A leukemia sample presented a challenging karyogram, necessitating aCGH as a complement to validate t(1;2). Suspicion of a previously missed t(5;14) called for a FISH analysis. Results from karyotyping and FISH were inconclusive since the acceptor chromosome couldn't be clearly identified. The two events were captured by Bionano in a single experiment and benign structural variants were automatically filtered out without complex pipelines.[1]

Challenging Leukemia Sample



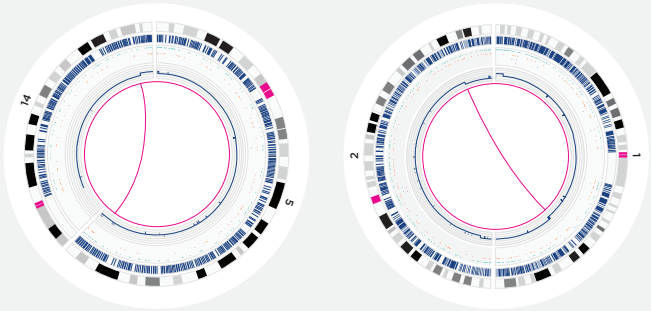
Current Methods

3 different platforms required to identify all events



Bionano Genome Imaging

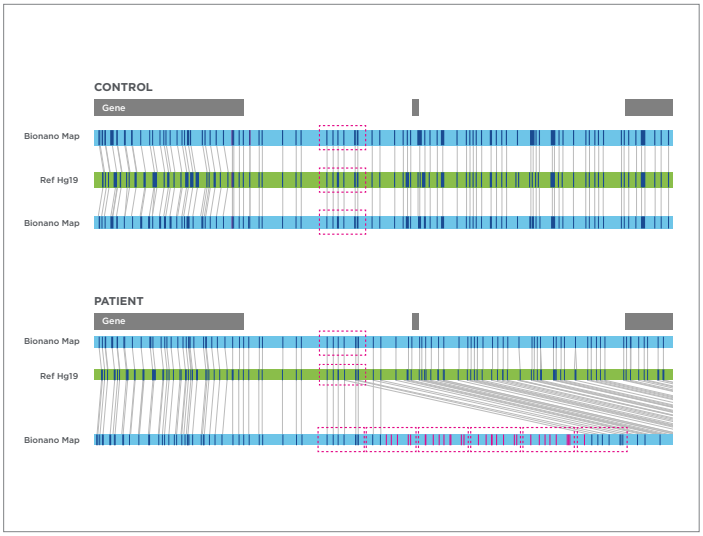
Consolidates 3 Into 1 convenient assay with all events identified



BIONANO IDENTIFIES VARIANTS IN, AND AROUND GENES IMPLICATED IN CANCER, THAT ARE MISSED BY SHORT AND LONG READ SEQUENCING

Because they are too long:

A case of familial cancer known to be linked to a particular gene was studied for a decade without any molecular evidence of events perturbing that gene. Bionano identified a 38kb cassette that was amplified in 6 tandem copies just upstream of this gene. Saphyr imaged single molecules spanning all 230 kbp of this tandem repeat which allowed it to be observed directly, as opposed to inferred algorithmically as is the case with other molecular methods.[2]

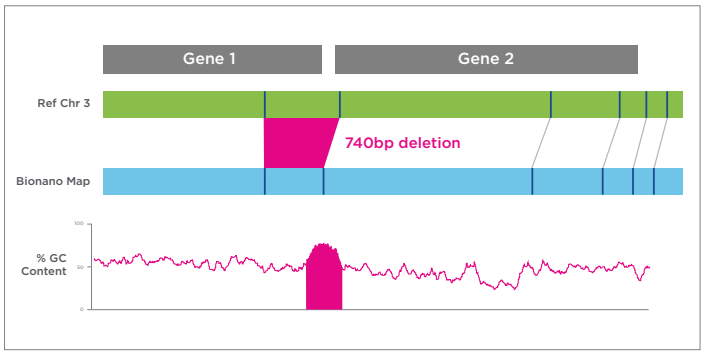


Because they are too rare:

The study of complex rearrangements in heterogenous cancer samples with long read sequencing is done at the expense of coverage, throughput or cost. Reaching low allelic fraction necessary to identify SV in biopsies requires deeper coverage than long read sequencing can offer for a reasonable price. By providing up to 1600X coverage for a human sample, Bionano can automatically detect variants present in as little as 1% allele fraction.

Because they are inaccessible by sequencing:

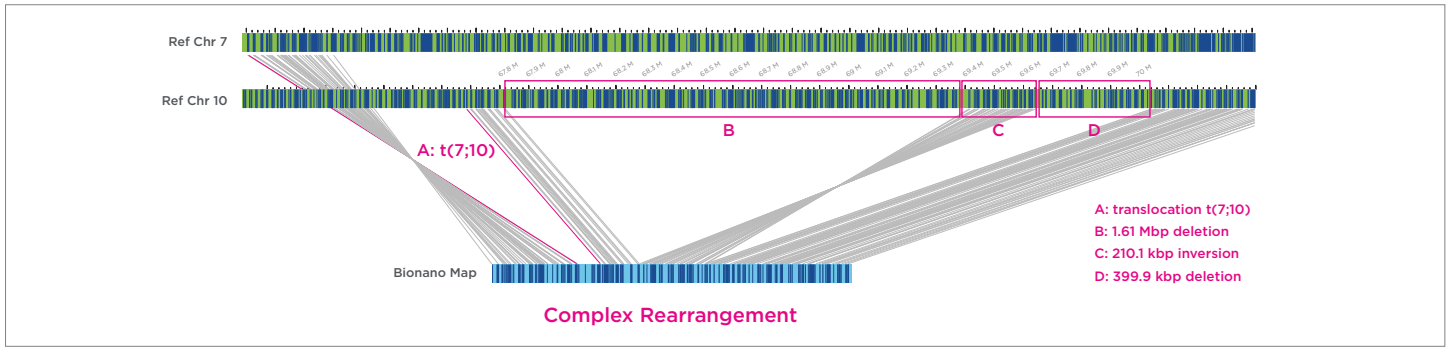
In a patient derived model of uveal melanoma, no mutation or epigenetic event was found explaining the loss of expression of gene 1 using NGS, which is typically associated with this cancer. Bionano identified a 740 bp deletion in the promoter of gene 1. This region was missed by sequencing due to the high GC content of that region.[3]



BIONANO IDENTIFIES MUTATION SIGNATURES AND BRINGS ORDER, WITHOUT COMPLEX PIPELINES

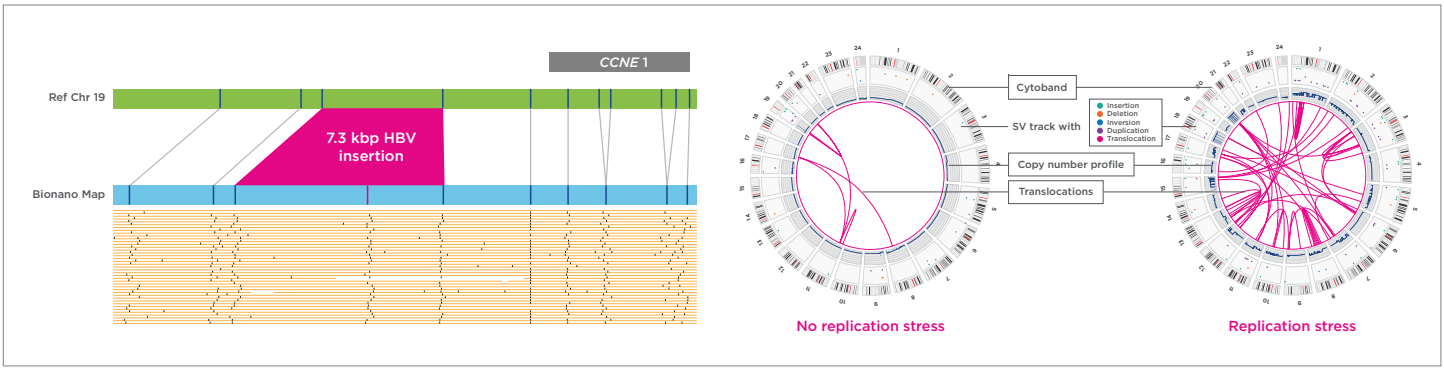
In highly rearranged cancer samples:

Cancer samples often display a high number of structural variants and the limitations of the short read length of NGS are particularly detrimental for making sense of complex successions of event, such as in chromothripsis. In this patient-derived breast cancer cell line, a consensus map resulting from the alignment of dozens of molecules spanning the region allowed Bionano to identify a succession of a translocation, deletions and an inversion missed by short-read and long read-sequencing.



In samples where stratification is needed:

The ability to stratify patient samples based on mutational profile usually requires extensive bioinformatic data curation downstream of sequencing. In a hepatocellular carcinoma study, our built-in pipeline automatically provides enough information to distinguish samples with or without a replication stress signature. The circos plot on the right in the image below shows replication stress resulting from a Hepatitis B virus insertion 9 kb upstream of the Cyclin E gene. Patients could be stratified based on the accumulation of DNA damage to determine if they are candidates for novel innovative therapies such as PARP inhibitors.[4]



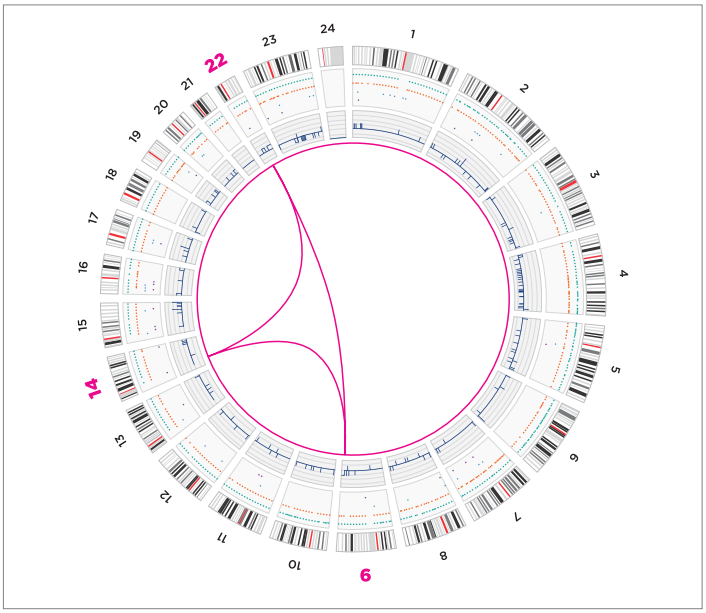
BIONANO IS A STREAMLINED TOOL FOR PERSONALIZED MEDICINE

Because it automatically identifies classical actionable fusions:

Rapid identification of BCR-ABL1 translocations in Acute Lymphoblastic Leukemia is urgent to leverage potential therapeutic options. Imatinib is a specific inhibitor of the fusion protein resulting from that translocation and identification of eligible patients is the cornerstone of personalized medicine in leukemia. With a unique workflow, Bionano Genome Imaging easily identifies complex and rare events such as this 3-way translocation of the Philadelphia chromosome t(9;22)(q34;q11).[5]

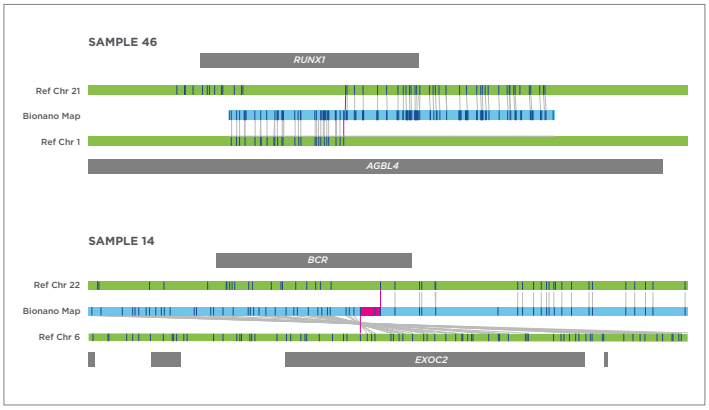
Because it identifies new fusions never discovered before:

In a study conducted on 48 leukemia samples, Bionano identified 2178 rare structural variants of which 95 were rare interchromosomal translocations and 23 were unique calls containing potential gene fusions, leading to new avenues of research for drug development.[5]



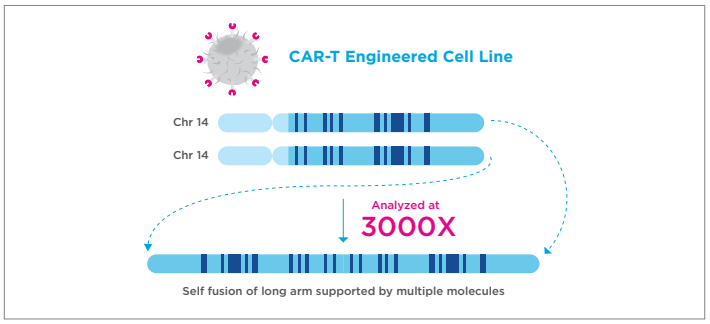
Because it easily identifies rare/new fusions of known genes:

In a large systematic comparative study between Bionano and classical cytogenetics, Bionano identified novel, non-recurrent fusions never reported before. In both cases shown here, one of the two fusion partners is well known in leukemia from other gene fusions. These events were missed by classical cytogenetics either because of their low allelic frequencies or because of the targeted FISH approach classically used in a diagnostics context.[5]



Because the ultrasensitive, automatic, genome wide detection of rearrangements brings safety in CAR-T cells manufacturing:

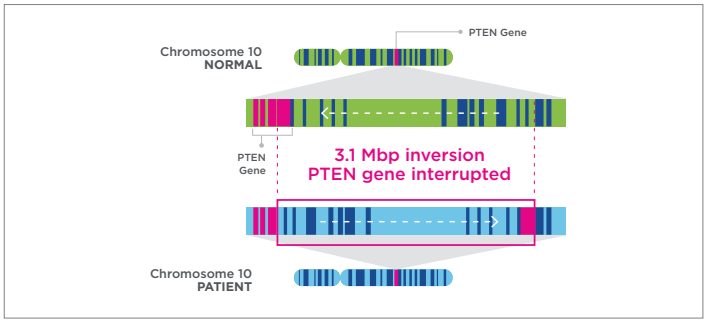
CAR-T cells are a powerful new therapeutic tool, but these engineered immune cells are often poorly characterized or remain unchecked for unwanted alterations. Bionano can rapidly collect several thousand folds of coverage of a human genome at a low cost, allowing for the detection of undesired rearrangements. Here, an unwanted inverted self-fusion of a large part of the long arm of chromosome 14 was detected in an engineered cell line that was mapped at 3000x coverage, and several imaged molecules validated this fusion.



COMBINED WITH NGS, BIONANO PROVIDES A COMPREHENSIVE UNDERSTANDING OF CANCER EVENTS, FROM SNVs TO MULTI MEGA BASE PAIR STRUCTURAL VARIATIONS

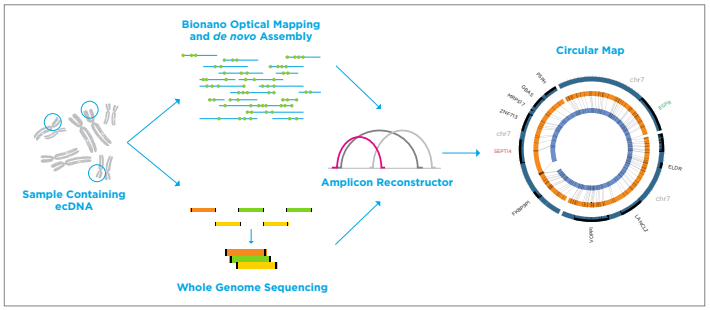
To perform complete tumor profiling for cancer discovery:

In 12 leukemia genomes analyzed with Bionano and whole genome sequencing (WGS), dozens of genes were affected by multiple structural and single nucleotide variants. Most of these genes were not previously associated with leukemias or with cancer in general. The potential for novel biomarker discoveries, even in such a small sample size, is unexpectedly large. Many of the SVs were affecting regulatory sequences, absent from whole exome sequencing approaches. Intergenic mutations are a known mechanism for tumor escape from chemotherapy. TCGA data showed that expression differences in several of these novel biomarker genes is associated with patient outcome and provides useful information for patient follow up regarding response to chemo. In the example shown here, a 3.1 Mbp inversion was detected by Bionano in the PTEN gene, while a SNV was detected by WGS in the other PTEN allele. This compound heterozygous mutation made this patient a candidate for trials with drugs attempting to rescue PTEN mutants.[6]



To elucidate complex genome structure:

Oncogenes are commonly amplified on particles of extrachromosomal circular DNA (ecDNA) in cancer, but our understanding of the ecDNA structure and its effect on gene expression is limited. Bionano was used to build a detailed map of ecDNA in a colorectal adenocarcinoma cell line, showing the circular structure, gene amplifications and fusion breakpoints.[7]



Combination with NGS data through Genoox:

Dozens of algorithms exist to call structural variants from NGS data, but all struggle with low sensitivities and high false positives. When Bionano's SV calls are used to inform the SV calling algorithms, the signatures of most SVs can be found in the short-read data. The Genoox integrated pipeline uses Bionano calls to identify short read pairs that confirm the variants, therefore validating the calls and refining the breakpoints. The Genoox AI-based classification engine produces a single report combining all variant calls from NGS and Bionano, classified by likely pathogenicity.

References: 1. Dr. Alex Hoischen, ESHG 2019 Workshop Series 2. Dr. Sven Bocklandt, AMP 2019 Series – Dr. Sven Bocklandt 3. David Gentien, Curie Institute in Paris (Festival of genomics 2020, manuscript in progress) 4. Dr. Eric Letouzé, Bionano Genome Imaging unravels complex structural rearrangements induced by replication stress in liver cancer – Letouze. 5. Neveling K et al. bioRxiv 2020.02.06.935742 6. Xu J et al. bioRxiv 563270 7. Luebeck J et al. bioRxiv 2020.01.22.916031

Contact your Bionano Regional Business Manager to get started.

info@bionanogenomics.com | 858.888.7600 | bionanogenomics.com

Part #30367 Rev 1