The Power of Massively Parallel Partitioning

Inside the Chromium Controller

The Power of Partitioning

The Chromium System, powered by GemCode Technology, provides a precisely engineered reagent delivery method that enables thousands of micro-reactions in parallel.

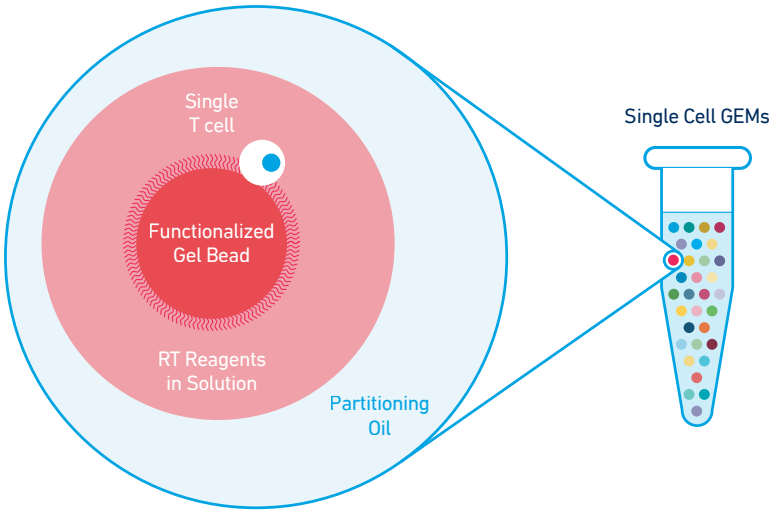

Figure 1. A GEM is a "Gel Bead-in-emulsion" droplet that encapsulates each micro-reaction within the Chromium System. Here we show a Single Cell GEM with a single T cell, reagents, and barcoded gel bead all partitioned within a single droplet.

Massive Partitioning and Barcoding

Encapsulate your sample into hundreds to tens of thousands of uniquely addressable partitions in minutes, each containing an identifying barcode for downstream analysis.

Each Gel Bead, infused with millions of unique oligonucleotide sequences, is mixed with a sample, which can be high molecular weight (HMW) DNA, individual cells, cells labeled using Feature Barcoding technology, nuclei, nuclei treated with transposase, or Cell Beads (see page 6). Gel Beads and samples are then added to an oil-surfactant solution to create Gel Beads-in-emulsion (GEMs), which act as individual reaction vesicles in which the Gel Beads are dissolved and the sample is barcoded (Figure 1). Barcoded products are pooled for downstream reactions to create short-read sequencer compatible libraries. After sequencing, the resulting barcoded short read sequences are fed into turnkey analysis pipelines that use the identifying barcodes to map reads back to their original HMW DNA, single cell, or single nucleus of origin.

Single Cell Partitioning for Transcriptome Analysis

The Chromium System also enables single cell transcriptional profiling of up to tens of thousands of single cells. Single cell suspensions loaded onto the system are partitioned into GEMs, where transcripts are tagged with cell-specific barcodes. The barcoded cDNA is then pooled for downstream processing and library preparation (Figure 2).

Our precise and efficient microfluidics allow 100–80,000+ cells to be recovered in droplets in each efficient run with a low doublet rate, facilitating the profiling of precious and rare cell populations. After sequencing, downstream bioinformatics tools use the cellular barcodes to group transcripts that originated from the same cell, revealing the transcriptome. Feature Barcoding technology uses specific oligonucleotide sequences to identify cell surface proteins or CRISPR perturbations associated with each individual cell for more robust cellular phenotyping and characterization.

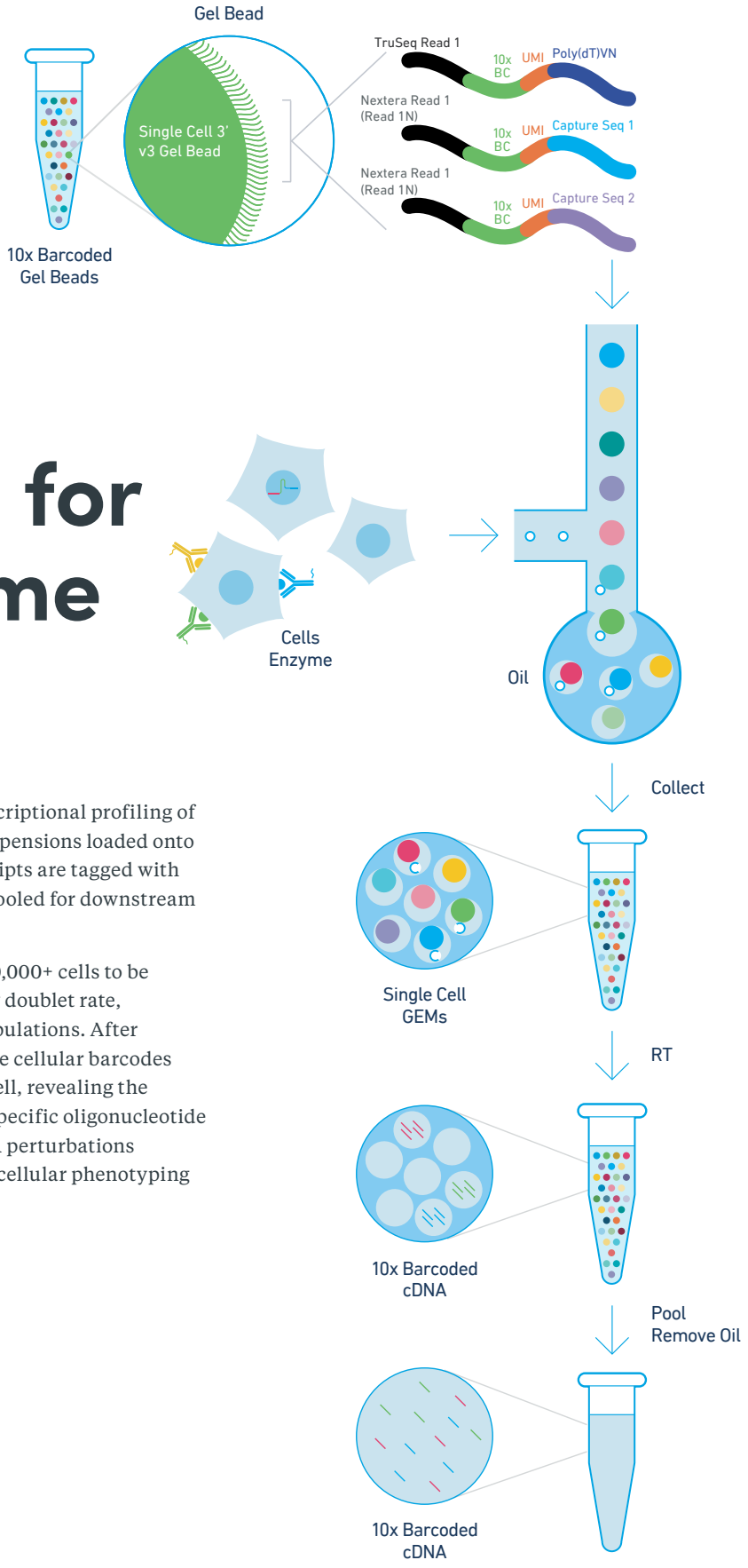

Figure 2. The Chromium partitioning workflow for single cell transcriptome analysis.

Single Cell Partitioning for Immunology Applications

Immune profiling of up to tens of thousands of T and B cells on a cell-by-cell basis can also be performed with the Chromium System. Single cell suspensions, including flow sorted cells, or cells labeled with antibodies or peptide-MHC multimers using Feature Barcoding technology, are loaded into the system and partitioned into GEMs. Transcripts are generated and tagged with cell-specific barcodes and the cDNA is then pooled for downstream processing and library preparation (Figure 3). For immune repertoire profiling, the cDNA undergoes targeted enrichment for T or B cell receptor transcripts prior to library preparation.

Our precise and efficient microfluidics allow 100–80,000+ cells to be recovered in droplets rapidly, with a low doublet rate, facilitating the profiling of precious and rare cell populations. After sequencing, downstream bioinformatics tools use the cellular barcodes to group transcripts that originated from the same cell, revealing the transcriptome and the full-length paired T or B cell receptor sequences of each individual cell. The Feature Barcode sequence is also used to identify the specific antigen or cell surface protein associated with each individual cell, allowing for a more accurate characterization of innate and adaptive immunity.

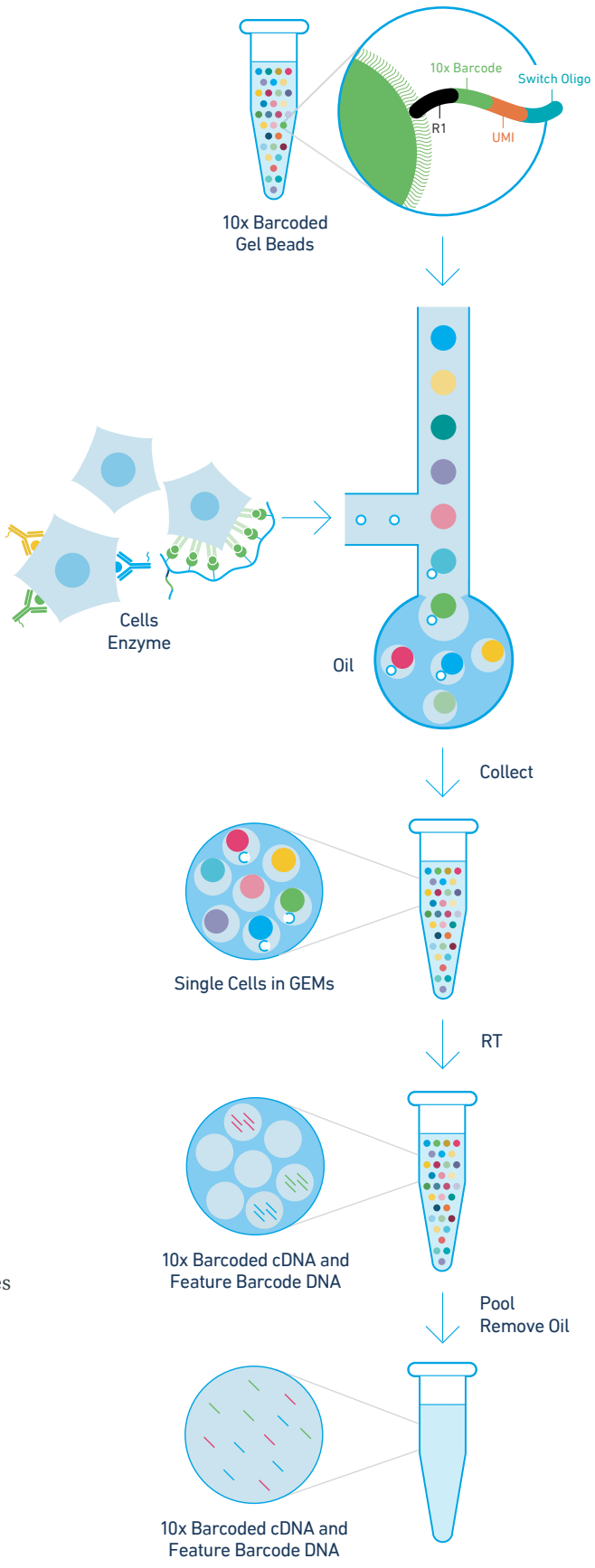

Figure 3. The Chromium partitioning workflow for single cell immunology applications.

Single Nuclei Partitioning for Epigenome Analysis

The Chromium System enables the analysis of chromatin accessibility at single cell resolution. In the Chromium Single Cell ATAC (Assay for Transposase Accessible Chromatin) workflow, single nuclei are first treated in bulk with transposase enzyme to preferentially insert sequencing adaptors into accessible DNA regions. Transposed nuclei are then partitioned into GEMs in the Chromium Controller. All accessible DNA fragments from the same nucleus share a common 10x barcode. The barcoded, accessible DNA fragments are subsequently pooled for downstream processing and library preparation (Figure 4).

The precise and efficient microfluidics allow 500–80,000+ nuclei to be recovered in droplets in each run with a low doublet rate, facilitating the open chromatin profiling of heterogeneous sample types and detecting rare cell populations. The barcoded library fragments can then be easily traced back to the open chromatin landscape for each nucleus from which they originated using intuitive bioinformatic tools.

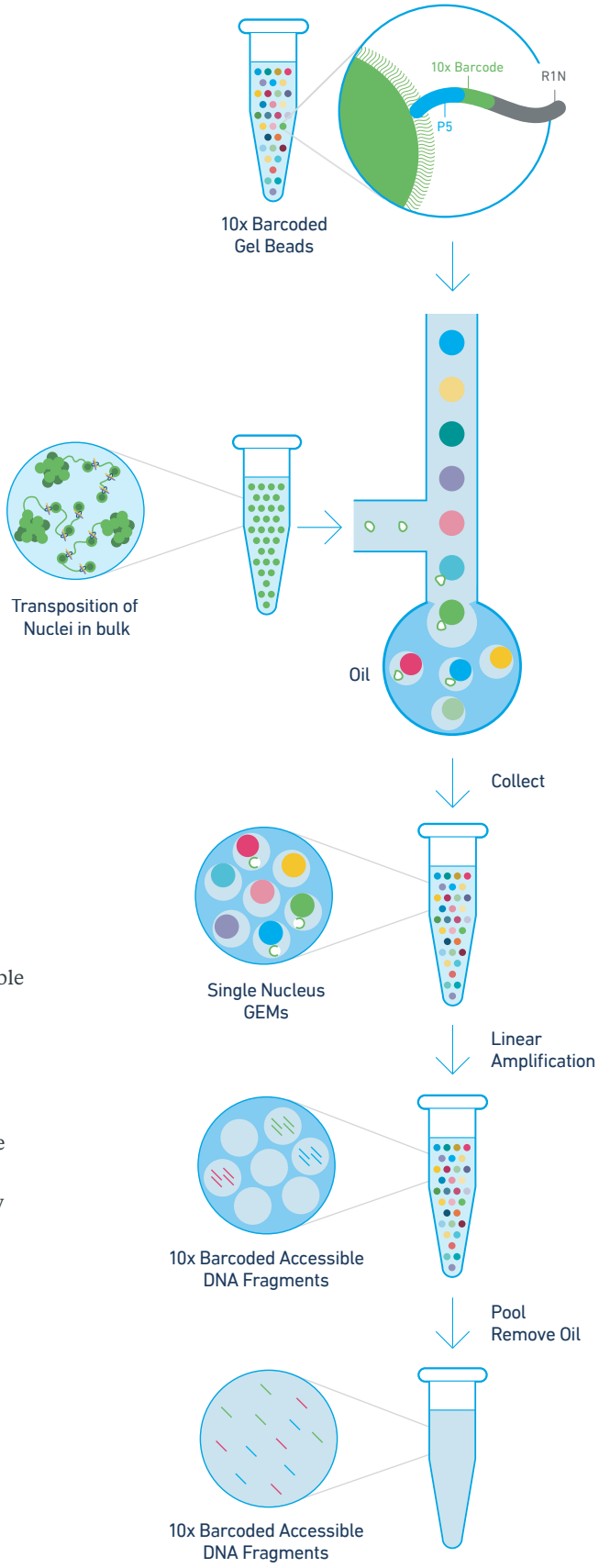

Figure 4. The Chromium partitioning workflow for single cell epigenomic profiling.

Single Cell Partitioning for Genome Analysis

Using the 10x technology, DNA from single cells is prepared in a two-step partitioning process. In the Chromium Single Cell DNA1 workflow, single cells are first encapsulated in hydrogel Cell Beads (CBs), allowing the cells to be subjected to protein digestion and denaturation, while still retaining intact DNA in the hydrogel matrix (Figure 5). After the cells have been lysed, CBs containing DNA are partitioned with 10x barcoded Gel Beads (GBs), to generate Cell Bead Gel Bead (CBGB) GEMs (Figure 2). DNA from single cells is barcoded within the CBGB partitions and the barcoded fragments are then pooled for library production. The barcoded library fragments can be easily traced back to the cells from which they originated using downstream bioinformatics tools.

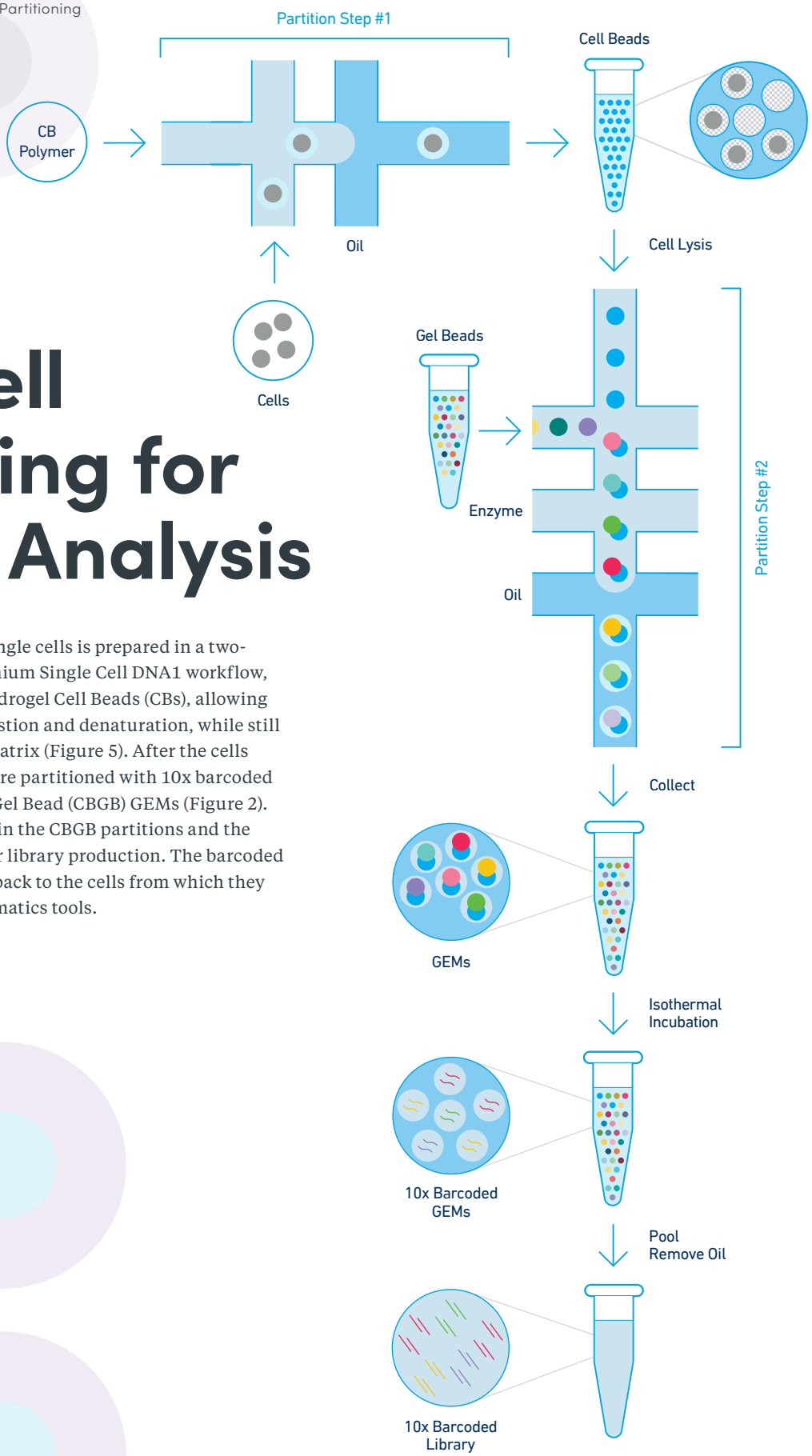

Figure 5. The Chromium partitioning workflow for single cell genome analysis.

1 Available for use only with the indicated Chromium Controller (PN-120223;120246) or Chromium Single Cell Controller (PN-120263;120212).

High Molecular Weight DNA Partitioning

For whole genome[2] or exome[2] analyses, the Chromium Controller allows researchers to create sequencing-ready libraries with >1,000,000 unique barcodes from ~1 ng of HMW genomic DNA (Figure 6). Massive partitioning of the genome provides haplotype level dilution and enables the barcoding of long input DNA molecules, which are then sequenced in bulk to produce a unique data-type known as Linked-Reads. The long range information encoded in barcoded Linked-Reads is leveraged by innovative bioinformatics pipelines to assemble sequences over long genomic distances, including across repetitive regions. The precise assembly of Linked-Reads leverages heterozygous loci to resolve individual haplotypes, enabling diploid *de novo assembly* and phased calling of the full spectrum of human genetic variations, including SNPs, small indels, and complex structural variants.

2 Available for use only with the indicated Chromium Controller (PN 120223 or 120246).

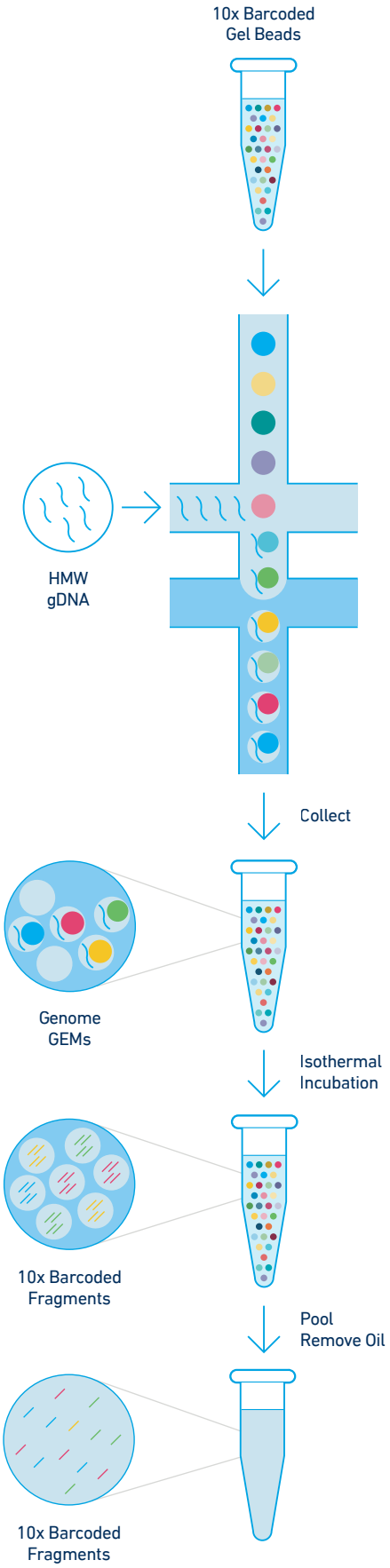

Figure 6. The Chromium DNA partitioning workflow for genome sequencing.

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