

Flow Chart A: Detailed description for 10X Genomics Chromium single-cell microfluidic technology for GEM generation

The figure depicts a six-step visual for how the microfluidic device achieves single-cell resolution of nucleic acids. Each step consists of an icon separated by blue arrows. A text box in the lower left quadrant of the figure. The first entry in the text box states “Inputs:” followed on the next two lines by “10x Genomics gel beads, reagents and single cell suspension. The second entry in the text box states “Outputs:” followed on the next several lines by “Digital gene expression and Feature Barcode profiles from every partitioned cell (cell multiplexing, cell surface protein expression, or CRISPR perturbations)”. Below, the steps of the flow chart are described as a list in which each step is stated above each arrow.

Step 1: 10x Barcoded Gel Beads

Icon consists of a microcentrifuge tube with multiple differently color circles denoting the uniquely barcoded gel bead. Beads are in a light blue background.

Step 2: Encapsulation

Icon demonstrating encapsulation of cells and enzyme reagents with uniquely barcoded gel bead. A light blue ‘T’-shaped icon denotes the microfluidic channel. The multicolored gel beads enter the ‘T’ junction from the left and small clear circles, which denote cells and enzymes enter from below. To the right of the ‘T’ junction the gel bead and cells are touching and are surrounded by a light blue circle in a darker blue background labeled ‘oil’.

Step 3: Collect

Icon consists of a microcentrifuge tube with multiple differently color circles denoting the uniquely barcoded gel bead. Beads are in a light blue background, in a similar fashion as Step 1. A circle below the tube shows a close-up of the oil encapsulated gel bead and cells, each in individual light blue circles embedded in a darker blue background and is described as “Single Cell GEMs”.

Step 4: Reverse Transcription

Icon consists of a microcentrifuge tube with multiple differently color circles denoting the uniquely barcoded gel bead. Beads are in a light blue background, in a similar fashion as Step 1. A circle below the tube shows a close up of the light blue single cell GEMs. A subset of GEMs contains several colored lines that corresponds to the colored of the circle shown in Step 3. Text below is labeled “10x Barcoded cDNA and Feature Barcode DNA”.

Step 5: Pool Remove Oil

Icon consists of a microcentrifuge tube filled with a light blue. A light blue circle is below the tube and has a series of short colored lines corresponding to the colors of the lines described in Step 4. This is described as “10X Barcoded cDNA and Feature Barcode DNA”.

Step 6: Gene Expression and Feature Barcode Profiling of Individual Cells

Icon shows the grouping of cDNA fragments, denoted by a light grey line by cell number (Cell 1 and 5000 are shown) and gene or feature number. cDNA fragments from Cell 1 all have a blue circle on the left and all cDNA fragments from Cell 5000 have a green circle on the left. The number of cDNA fragments differs for each gene or feature, denoting differences in gene expression.

Flow Chart B: Detailed description for 10X Genomics single cell workflow.

The figure is a flowchart showing the six phases for the 10X Genomics single cell workflow starting from sample prep and ending with data visualization. Each phase has 1-2 icons separated by grey arrows. Above each phase's icons is a label denoting the phase. Below, the workflow is described as a list with each step having a brief description.

Phase 1: Sample prep

Microcentrifuge tube filled with dark blue dots in a light blue background. Circle below the microcentrifuge tube shows a cartoon of a zoomed portion of the microcentrifuge tube and contains the depiction of individual cells, indicating an isolated single cell suspension.

Phase 2: GEM Generation

Icon is a cartoon depiction of the 10X Genomics Chromium microfluidic controller.

Phase 3: Library Construction

Microcentrifuge tube filled with a light blue color. Circle below the microcentrifuge tube shows a cartoon of a zoomed portion of the microcentrifuge and contains numerous differently colored lines, indicating single cell barcoded cDNA ready for sequencing.

Phase 4: Sequencing

Icon shows a massively parallel sequencing machine.

Phase 5: Data Processing

A cartoon workflow consisting of four light blue boxes with numerous arrows connecting the boxes. No text is present.

Phase 6: Data Visualization

Icon shows a cartoon computer screen with dozens of small dots grouped by color and clumping together in different portions of the screen. Visualization depicts a t-SNE or UMAP plot.