

Multiomic profiling of the immune system at single cell resolution

Single Cell Immune Profiling v2 with Feature Barcode technology

The immune system is complex, composed of diverse cell types, many cell–cell interactions, and a range of antigen specificities. Traditional methods of immune surveillance allow researchers to interrogate one data type at a time, leaving them with an incomplete understanding of the immune response. Characterizing the immune response requires the ability to phenotype individual cells with multiple parameters and at massive scale.

The Chromium Single Cell Immune Profiling Solution provides a comprehensive approach to simultaneously examine cellular heterogeneity of the immune system, T and B cell repertoire diversity, and antigen specificity at single cell resolution. Discover new cell types and states using whole transcriptome analysis, or focus your search with targeted gene expression panels of interest. With our latest improvements, Single Cell Immune Profiling v2 vastly increases sensitivity, enabling detection of rare cell populations and phenotypes. Comprehensive immunophenotyping with this scale and resolution has never been more accessible.

Highlights

- Characterize adaptive and innate immune cell diversity
- Identify and characterize rare cell types and biomarkers
- Analyze tissue microenvironment, disease progression, and drug immune response
- Perform large scale antibody and TCR discovery for novel antigens
- Characterize immune response to infection by measuring clonal expansion and immune cell phenotypes
- Focus on relevant genes and pathways with pre-designed gene expression panels for immunology, oncology, and drug discovery, or design a fully custom panel

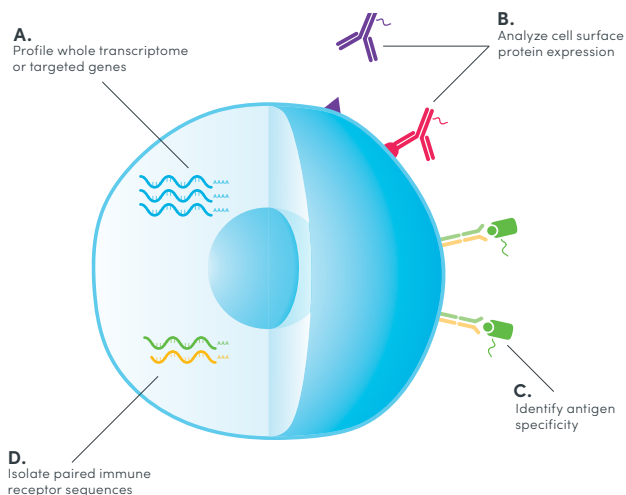


Figure 1. Multiomic interrogation of immune cell diversity. Detect whole transcriptome or targeted gene expression, along with cell surface proteins; paired, full-length receptor sequences of T or B cells; and antigen specificity. **A.** Chromium Single Cell Immune Profiling provides sensitive whole transcriptome analysis at the single cell level, for hundreds to tens-of-thousands of cells per reaction. Focus on your genes of interest with targeted gene expression using pre-designed or custom panels. **B.** Chromium Single Cell Immune Profiling with Feature Barcode technology yields multiomic analysis of gene expression and simultaneous cell surface protein detection with up to hundreds of antibodies at the single cell level. **C.** Feature Barcode technology also allows screening of the antigen specificity of T and B cells. Interrogation of the binding of barcoded antigens to immune cell receptors allows full characterization of the adaptive immune response. **D.** Obtain corresponding paired, full-length antigen receptor sequences to identify distinct clonotypes.

Gene Expression t-SNE

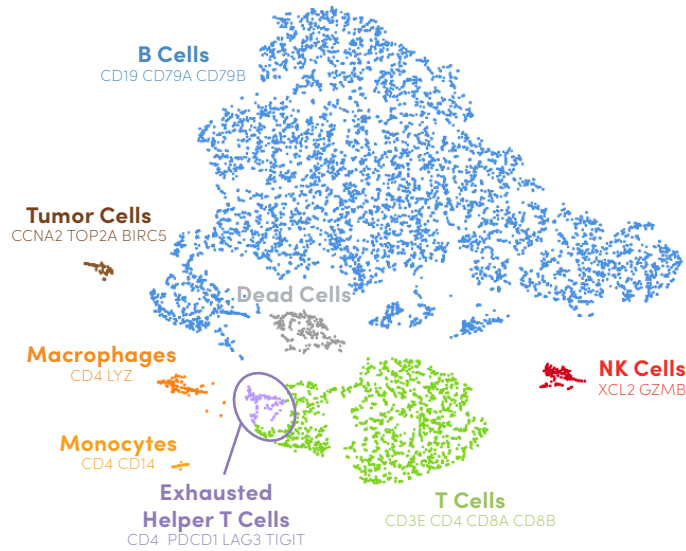


Figure 2. Diverse immune cell populations revealed in a complex cancer sample by single cell gene expression. To visualize the global differences in gene expression signatures across different cell types, the t-SNE dimensionality reduction technique was used, which represents each cell with a single dot. Here, a t-SNE projection shows 7,859 cells from a dissociated human melanoma sample that were run in a single channel. Cells were clustered by Cell Ranger based on whole transcriptome gene expression (as in Figure 1, A.) and manually annotated in Loupe Browser. Broad categories of immune cell populations were identified through gene expression signatures, including cell populations that were not identified by protein markers in Figure 3 such as macrophages (orange) and a small population of tumor cells (brown).

Antibody t-SNE

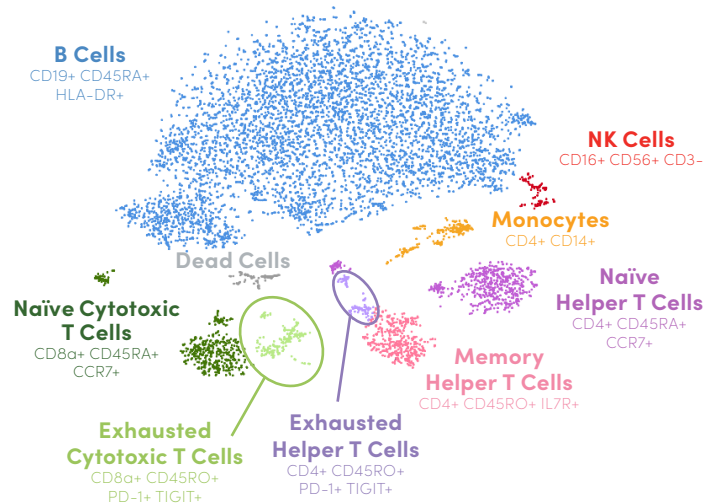
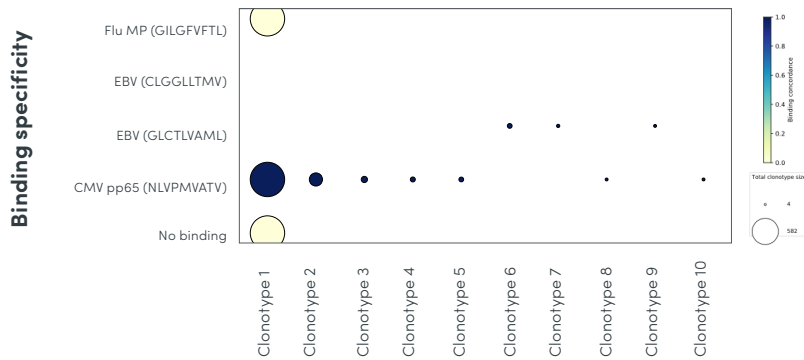
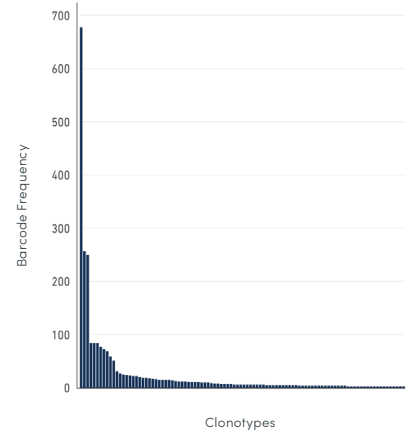


Figure 3. Multimic data with cell surface protein analysis enhances resolution of immune cell subtypes in a complex cancer sample. t-SNE projection of the same dissociated human melanoma sample as in Figure 2. Cells were clustered by Cell Ranger based on cell surface protein expression using a panel of 16 markers (as in Figure 1, B.) and manually annotated in Loupe Browser. Analysis of cell surface protein expression enabled further classification of T-cells into naïve, memory, and exhausted subpopulations.

A. Antigen Binding Specificities



B. TCR Clonotype Frequency



C. Full length sequences of Clonotype 1: TCR alpha and beta chains

TCR Alpha

TGGGGAGTCCACAGTTAGGCAGCGCACTTCTGAAAGGGTGTCAATTCTCGTTGTGTTCCACACTCATGGGGGATTTCTCAAAGTTTATGAAGTTGGCCTGAAGA
AAGCAGTTACTTCGACCCCTTTTGTGAACCACAGGAACTGAACATAGGGCAGAGCGTTTCTAGGACATTTATGGGAGCTGGTCTCTGATTGGCTAGGCAACCA
GATGCAGAATCCTGTTTTCTGAAGCAGAATATTTCCACAGATTTTGGCTGCAAAACGTTTTTCTGCTGTGGGTACGTGAGCAGGAAAC**ATGGAGAAGAATCCTTTG**
GCAGCCCATTAATACTCTCTGGTTTCATCTTGACTGCGTGAGCAGCATACTGAACGTGGAACAAAGTCTCAGTCACTGCATGTTCCAGGAGGAGACAGCACC
AATTTACCTGCAGCTCCCTCCAGCAATTTTATGCCTTACACTGGTACAGATGGGAACTGCAAAAAGCCCGAGGCCTGTTTGTAAATGACTTTAAATGGGG
ATGAAAAGAAGAAAGGACGAATAAGTGCCACTCTTAATACCAAGGAGGGTTACAGCTATTTGTACATCAAAGGATCCCAGCCTGAAGACTCAGCCACATACCTCTG
TGCCTTATCACCGGTAACAGTTCTATTTTGGGACAGGGACAAGTTTGACGGTCAATCCAAATATCCAGAACCCTGACCTGCGCTGTACCAGCTGAGAGAC

TCR Beta

GAGAGTCTGCTCCCTTTTCATCAATGCACAGATACAGAAGACCCCTCCGTCATGCAGCATCTGCC**ATGAGCATCGGCCTCCTGTGCTGTGCAGCCTTGTCTCTC**
CTGTGGGCAGGTCCAGTGAATGCTGGTGTCACTCAGACCCCAAAATTCAGGTCTGAAGACAGGACAGAGCATGACACTGCAGTGTGCCAGGATATGAACCA
TGAATACATGCTGGTATCGACAAGACCCAGGCATGGGGCTGAGGCTGATTCATTACTCAGTTGGTGTGGTATCACTGACCAAGGAGAAGTCCCAATGGCTA
CAATGTCTCCAGATCAACCACAGAGGATTTCCCGCTCAGGCTGCTGTGCGCTGCTCCCTCCAGACATCTGTGACTTCTGTGCCAGCAGCCTGTTACCGGGA
CAGGGGCATACGGTACACCTTCGGTTCGGGACCAGGTTAACCGTTGTAGAGGACCTGAACAAGTGTCCACCCGAGGTGCGTGTGTTGAGCCATCAGA

Figure 4. Characterization of anti-CMV T cells from a CMV+ donor by antigen binding specificity, clonotype frequency, and paired, full-length T-cell receptor sequences. **A.** The ten largest antigen-binding clonotypes are plotted along with their binding specificities and binding concordances, as defined by dCODE Dextramer® Reagent binding (as in Figure 1, C). “No binding” is defined as a clonotype showing no binding to peptide-MHC (pMHC) on the y-axis. The presence of a circle indicates that at least one member of the clonotype was specific for a particular pMHC. Circle size indicates the total number of cells for the given clonotype. Circle color indicates the proportion of cells within the clonotype that bind the specific pMHC, with darker colors having higher binding concordance. **B.** TCR clonotypes are ranked by frequency. The top CMV-specific clonotype has 678 cells. Data were output by Cell Ranger and visualized using Loupe V(D)J Browser. **C.** Shown are paired, full-length TCR α and β chain V(D)J sequences (see Figure 1, D.) for the top expanded clonotype in Figure 4, B. V(D)J nucleotides are color coded as follows: 5’UTR (gray), V (red), D (purple), J (green), C (blue), and CDR3 (bold). Full-length sequences are output by Cell Ranger.

Solution Features

- Profile thousands of genes by barcoding mRNA at the 5' end, at the single cell level
- Simultaneously profile immune repertoire (BCR/TCR) and gene expression from the same cell
- Obtain paired, full-length receptor sequences from T cells and/or B cells with complete isotype resolution, for human or mouse samples
- Combine gene expression analysis with detection of hundreds of cell surface proteins at high resolution
- Compatible with Targeted Gene Expression Panels
- Visualize clonal expansion and sample heterogeneity using easy-to-use, fully integrated software: Cell Ranger analysis and Loupe Browser or Loupe V(D)J Browser
- Compatible product partners for Feature Barcode technology (oligo-conjugated antibodies, oligo-conjugated MHCs)
- Dual index libraries for superior sequencing data quality

System Features

- Efficiently partition 500–10,000 cells per channel, for up to 80,000 cells per run
- Scalable; run up to 8 samples in parallel
- Cell size flexibility, no lower limits
- High cell capture rates of up to 65%
- Low doublet rates of under 0.9% in 1000 cells
- Based on Next GEM technology

Research areas

- Infectious Disease & Vaccines
- Autoimmunity, Inflammation & Allergies
- Immuno-oncology & Immunotherapies
- Transplantation Immunology
- Cellular & Molecular Immunology

Applications

- Immune Cell Atlasing
- B-Cell & T-Cell Receptor Profiling
- Antibody Discovery
- Tissue-Infiltrating Lymphocyte Characterization
- Cellular Immunotherapy Discovery

Additional resources

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Gene Expression and/or Immune Repertoire Profiling	
Products	Product Code
Chromium Next GEM Single Cell 5' Kit v2, 16 rxns	1000263
Chromium Next GEM Single Cell 5' Kit v2, 4 rxns	1000265
Chromium Next GEM Chip G Single Cell Kit, 48 rxns	1000120
Chromium Next GEM Chip G Single Cell Kit, 16 rxns	1000127
Library Construction Kit, 16 rxns	1000190
Dual Index Kit TT Set A, 96-rxns	1000215

Feature Barcode Kits	
Reagent Kits	Product Code
5' Feature Barcode Kit, 16 rxns	1000256
Dual Index Kit TN Set A, 96-rxns	1000250

V(D)J Amplification Kits		
Species and Target	Reagent Kits	Product Code
Human T Cell	Chromium Single Cell Human TCR Amplification Kit, 16 rxns	1000252
Human B Cell	Chromium Single Cell Human BCR Amplification Kit, 16 rxns	1000253
Mouse T Cell	Chromium Single Cell Mouse TCR Amplification Kit, 16 rxns	1000254
Mouse B Cell	Chromium Single Cell Mouse BCR Amplification Kit, 16 rxns	1000255

Products	Product Code
Chromium Controller & Accessory Kit, 12 Mo. Warranty	120223
Chromium Controller & Accessory Kit, 24 Mo. Warranty	120246
Cell Ranger go.10xgenomics.com/vdj/cell-ranger	Download
Loupe Browser go.10xgenomics.com/vdj/loupe-cell	Download
Loupe V(D)J Browse go.10xgenomics.com/vdj/loupe-vdj	Download
Compatible Partner Product: Biolegend TotalSeq™-C go.10xgenomics.com/totalseq-C	Download
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