# High-Throughput Single Cell Immune Profiling

**Product Catalog**

- Discover novel cell types and functions
- Recover a comprehensive immune repertoire
- Explore antigen specificity
- Discover new biomarkers
- and More!

## To Learn more

If you have any questions or would like to discuss how our services can help you with your research, please don't hesitate to contact us at P_contact@innomics.com. We look forward to hearing from you!

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# Contents

# What Is High Throughput Single Cell Immune Profiling

## 1 Introduction

Our Single Cell Immune Profiling service is based on 10x Genomics' high throughput single cell processing platform and provides an integrated service for sample preparation, cell sorting, droplet formation, library construction, sequencing and bioinformatics. By analyzing both paired B-cell or T-cell receptors, and 5' mRNA gene expression information from the same cell, 10x Genomics' high throughput single cell Immune Profiling provides a powerful and convenient multi-omics solution.

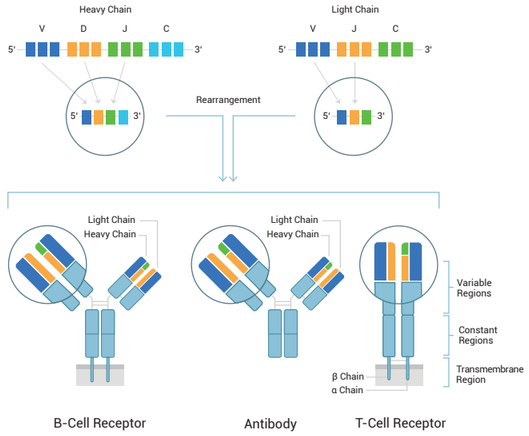


Fig 1. T Cell Receptor and B Cell Receptor Structures*

The 10x Genomics microfluidics chip technology works by pooling gel beads, coated with barcoded primers, cells and enzymes, and loading them on to the chip. Within the Chromium controller, barcoded gel beads are mixed with cells or nuclei, enzymes, and partitioning oil to form “GEMs" (Gel Bead-in-emulsion), which are single-cell emulsion droplets.

Within the GEM, a reaction takes place where gel beads are dissolved and genetic materials from a single cell are captured and barcoded. Barcoded fragments are pooled for down-stream reactions to create sequencing libraries and after sequencing reads are mapped back to the corresponding single cell.

*https://medicine.yale.edu/keck/ycga/sequencing/10x/singcellsequencing/



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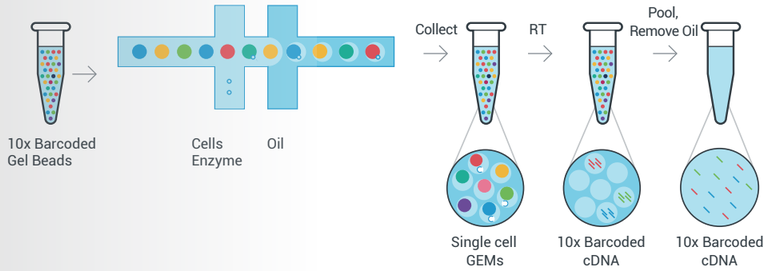



Fig 2. 10x Genomics Library Construction Mechanism*

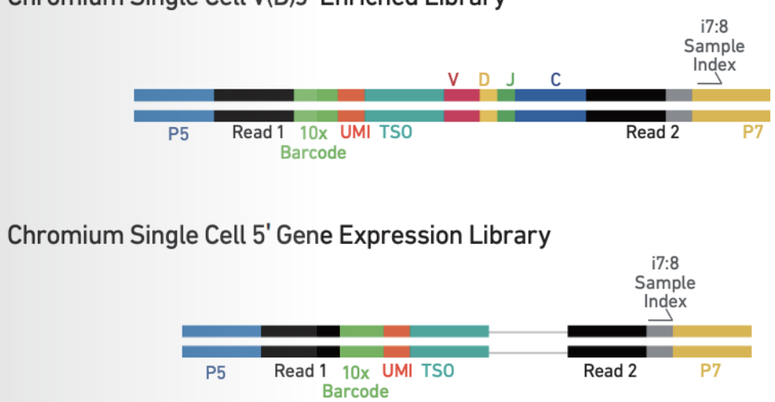
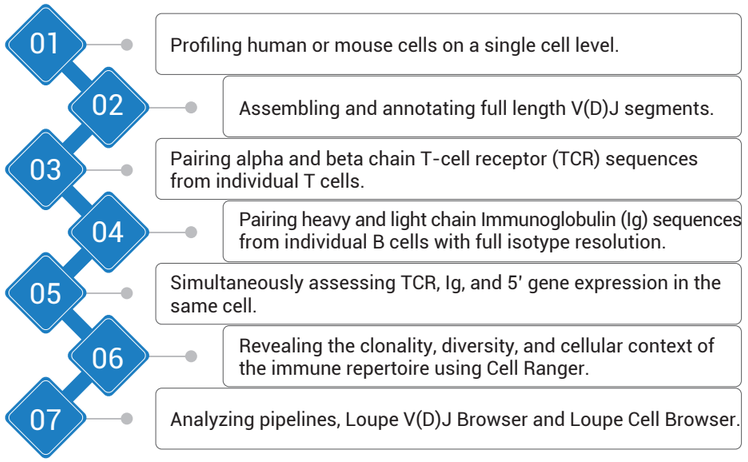



Fig 3. 10x Genomics 5' RNA-Seq & V(D)J Library Sequence Paradigm*

*10x Genomics

# What Can High Throughput Single Cell Immune Profiling Do




# Why Choose Our High Throughput Single Cell Immune Profiling

## 2 Advantages

· **Dr. Tom:** A proven convenient intuitive web-based data visualization system for RNA data analysis.
· **High throughput** processing ability, up to 80,000 cells processed at one time.
· **Highly automated** processing platform, able to complete thousands of cells partitioning within 10 minutes; Amplification and library construction are done in the reaction system; Cell Ranger pipelines provide direct analysis result.
· **Lower cost** for each individual cell.



## 3 Our Workflow

First, the frozen sample is shipped to our lab through one of our reliable shipping partners. Upon arrival we will then check the cell viability rate. If the rate is less than 80%, we will perform a cell debris removal to ensure sample quality. Then, the sample is loaded onto the 10x Genomics single cell system to construct the library. The library will then go through QC and sequencing.

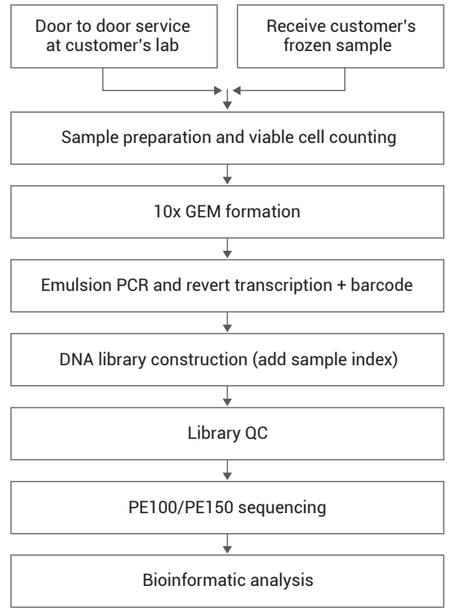



Fig 4. Service Flow Chart

## 4 Sample Requirements

| | | |
|---|---|---|
| Sample Requirements | Sample Type | Fresh/Frozen Tissue, Fresh PBMC, Cell Line |
| | Cell Condition | Cell diameter <40um; Cell viability rate>80%;<br>Cell number>5*10^5 cells/sample;<br>Clear background of suspension;<br>No large clumps and debris; No Ca2+ and Mg2+ |
| Sequencing Strategy | 5' mRNA-Seq library: PE100, 1 lane | |
| | TCR/BCR library: PE150, each 0.5 lane, total 1 lane | |
| Turnaround Time | 40~50 working days (case by case) | |

# 5

## Case Study 1

**CD4+ T cells contribute to neurodegeneration in Lewy body dementia. Gate D, Tapp E, Leventhal O, et al.. (2021). Science. 12;374(6569):868-874.**

To study the mechanism regulating T cell brain homing in Lewy body dementia and uncover potential mechanisms of brain entry in LBD, cerebrospinal fluid (CSF) of 11 healthy subjects and 11 PD-DLB were used in single cell RNA-Seq and single cell Immune profiling.

Fig 5

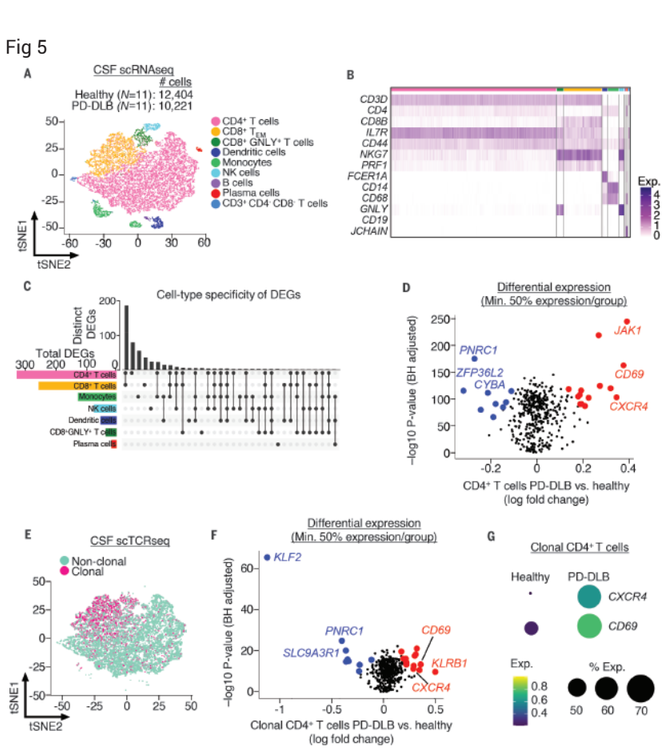



Fig 5

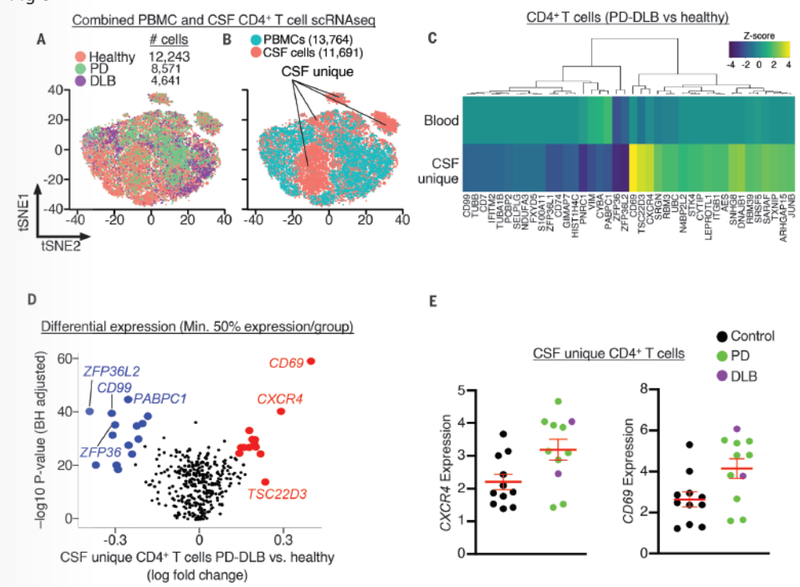
Single cell RNA-Seq revealed CD4+ T cells are the most transcriptionally altered immune cell subtype. JAK1, CD69 and CXCR4 are highly differentially expressed in PD-DLB cells. Enhanced CD4+ T cell cytokine signaling and activation can be observed in PD-DLB CSF. Single cell TCR sequencing revealed increased expression of CD69 and CXCR4 in PD-DLB, detected higher expression of KLRB1 and localized CD3+KLRB1+ T cells to phosphorylated alpha-synuclein deposits in the parenchyma of PDD brains. Thus, indicate LBD may involve enhanced activation of proinflammatory CD4+Th17 cells.



Fig 6

Single cell RNA sequencing of PBMCs compared CD4+ T cells of peripheral immune system and CSF of the same PD subjects. It uncovered CD4+ T cell populations that were specific to the CSF and CXCR4, CD69 and TSC22D3 are up regulated as the primary gene defining CSF specific T cells. Also, CD4+ T cell CXCR4 and CD69 have higher expression level in PD-DLB. Thus, CXCR4 may regulate homing of CD4+ T cells to LBD brain.

Fig 6




## Case Study 2

**Dissecting the heterogeneity of DENV vaccine-elicited cellular immunity using single-cell RNA sequencing and metabolic profiling. Waickman, A.T., Victor, K., Li, T. et al.. (2019). *Nat Communications.* 10, 3666.**

To study the molecular mechanisms of vaccine-elicited T cell immunity, samples from a subject who demonstrated strong NS1 and NS3 biased memory T cell responses following TAK-003 administration were used.

Fig 7

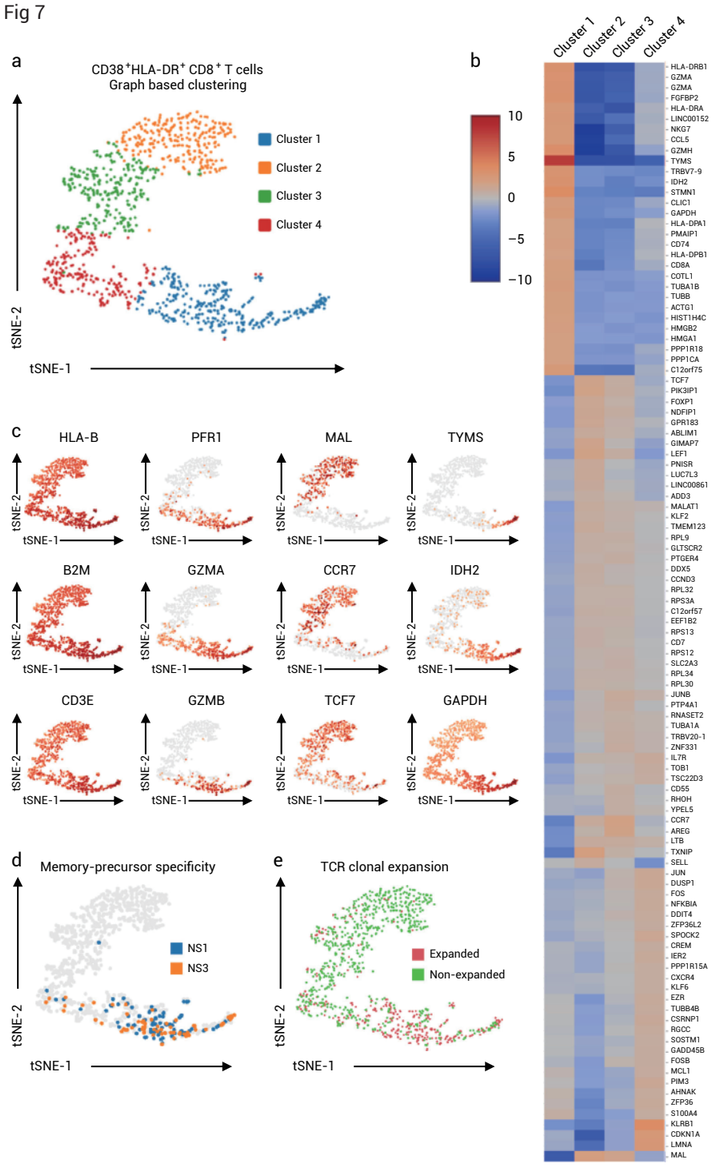



Fig 7

CD8+CD25+CD69+ T cells were isolated by flow cytometric cell sorting and subject to single cell RNA-Seq analysis. Distribution of CD38+HLA-DR+ CD8+ T cells at day 14 with TCR clonotypes overlapping with NS1- and NS3-reactive memory CD8+ T cells at day 120 (memory precursors).



Fig 8

TfR1+HLA-DR+ CD8+ T cells were sorted from a TAK-003 inoculated individual 14 days post immunization and subjected to single-cell RNAseq analysis. scRNA-Seq gene expression analysis of sorted CD38+HLA-DR+ CD8+ T cells and TfR1+HLA-DR+ CD8+ T cells 14 days post TAK-003 administration. Merged datasets showing cellular library origin, memory precursor specificity, and Cluster 1 origin cells.

Fig 8