

TECHNICAL NOTE

Chromium Next GEM Single Cell V(D)J v1.1: Reagent & Workflow Updates

Introduction

The Chromium Single Cell Immune Profiling Solution provides a comprehensive, scalable solution to simultaneously examine the cellular context of the immune system and the immune repertoires of hundreds to tens of thousands of cells on a cell-by-cell basis. This document highlights the key changes in Chromium Next GEM Single Cell V(D)J v1.1 reagents, along with workflow updates for generating and analyzing Illumina-ready sequencing libraries.

Refer to the Chromium Next GEM Single Cell V(D)J Reagent Kits v1.1 User Guides for the complete protocol.

Chromium Next GEM Single Cell V(D)J v1.1 Workflow

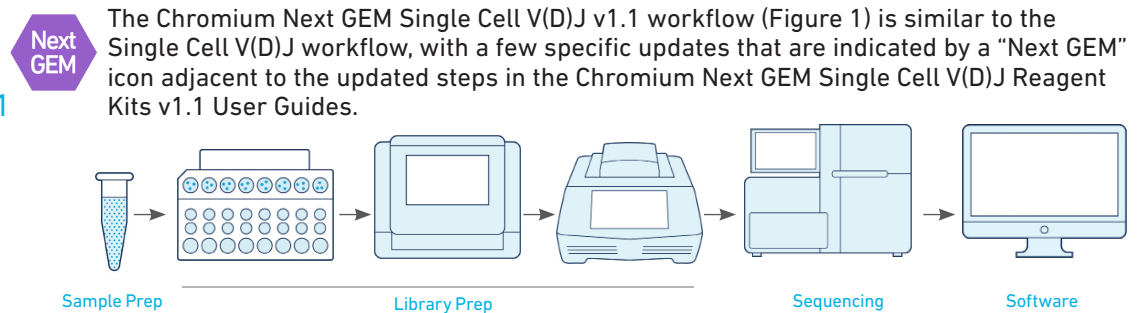


Figure 1. Chromium Next GEM Single Cell V(D)J v1.1 workflow.

Chromium Next GEM Chip G

The Chromium Next GEM Chip G is assembled in the Chromium Next GEM Secondary Holder. The location of the rows labeled 1-3 and the NO FILL (not used) wells are unique to the Chromium Next GEM Chip G (Figure 2A). GEMs are generated by combining a Master Mix containing cells and enzymes, barcoded Single Cell VDJ 5' v1.1 Gel Beads, and Partitioning Oil onto the Chromium Next GEM Chip G (Figure 2B).

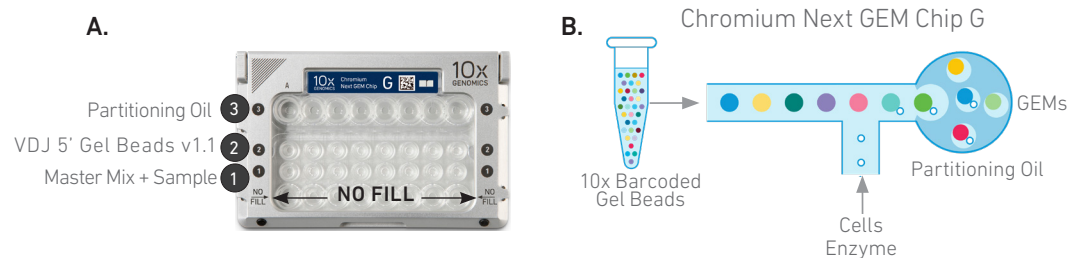


Figure 2. Chromium Next GEM Chip G assembled in a Chromium Next GEM Secondary Holder (A). GEM generation in Chromium Next GEM Chip G (B).

Sample Prep



Recommendations for preparing single cell suspensions are unchanged for Chromium Next GEM Single Cell V(D)J Reagent Kits v1.1 protocols.

Library Prep

Protocol Steps	Single Cell V(D)J	Next GEM Single Cell V(D)J v1.1
Reagent Kits		
10x Genomics Reagents	Chromium Single Cell V(D)J Reagent Kits Chromium Single Cell 5' Feature Barcode Library Kit Chromium i7 Multiplex Kits	Chromium Next GEM Single Cell V(D)J Reagent Kits v1.1 Chromium Single Cell 5' Feature Barcode Library Kit Single Index Kit T Set A
GEM Generation & Barcoding		
GEM-RT Reagents in Master Mix	RT Reagent Mix	RT Reagent B
Master Mix volume	68.3 µl	37.2 µl (updated reagent volumes in Master Mix)
Cell Suspension Volume Table	-	Updated volumes
Gel Bead	Single Cell 5' Gel Beads	Single Cell VDJ 5' Gel Beads v1.1 (updated handling instructions; vortex 30 sec, centrifuge 5 sec before use)

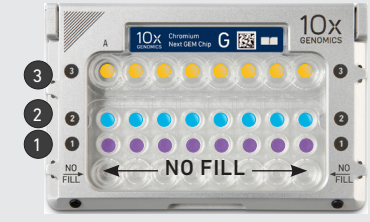


Chip Loading (Chips cannot be used interchangeably)



Master Mix + Sample Gel Beads Partitioning Oil

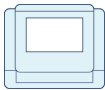
90 µl - row labeled 1
40 µl - row labeled 2
270 µl - row labeled 3



● **70 µl - row labeled 1**
● **50 µl - row labeled 2***
● **45 µl - row labeled 3****

* After loading the Gel Beads, wait 30 sec before loading Partitioning Oil.

After loading Partitioning Oil, attach the gasket and run the chip in the Chromium Controller **immediately.



Chromium Controller

Firmware Version 2.0 or higher

4.0 or higher

Run time ~6.5 min

~18 min

GEM Recovery

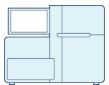
From top row labeled "NO FILL"



From top row labeled "3"



Sequencing



Recommendations for sequencing libraries generated using the Chromium Next GEM Single Cell V(D)J Reagent Kits v1.1 protocols are same as sequencing recommendations for libraries generated using the Chromium Single Cell V(D)J Reagent Kits protocols.

Software



Libraries generated using the Chromium Next GEM Single Cell V(D)J Reagent Kits v1.1 can be analyzed and visualized using the current versions of Cell Ranger and Loupe Cell Browser.

Chromium Next GEM Single Cell V(D)J v1.1 – Product List & Documents

Product list for generating Chromium Single Cell Libraries:

REAGENT KITS	REACTIONS	PART NUMBER (PN)
Chromium Next GEM Single Cell 5' Library & Gel Bead Kit v1.1	16 rxns 4 rxns	1000165 1000167
Chromium Single Cell 5' Library Construction Kit	16 rxn	1000020
Chromium Single Cell V(D)J Enrichment Kit, Human B Cell	96 rxns	1000016
Chromium Single Cell V(D)J Enrichment Kit, Human T Cell	96 rxns	1000005
Chromium Single Cell V(D)J Enrichment Kit, Mouse B Cell	96 rxns	1000072
Chromium Single Cell V(D)J Enrichment Kit, Mouse T Cell	96 rxns	1000071
Chromium Next GEM Chip G Single Cell Kit	48 rxns 16 rxns	1000120 1000127
Single Index Kit T Set A	96 rxns	1000213
INSTRUMENT		
Chromium Controller & Next GEM Accessory Kit	-	120223 (12 month warranty) 120246 (24 month warranty)
SOFTWARE		
Cell Ranger Analysis Pipeline (DOWNLOAD)		
Loupe Cell Browser (DOWNLOAD)		
DOCUMENTS		
User Guide : Chromium Next GEM Single Cell V(D)J Reagent Kits v1.1 (CG000207)		

If using Single Cell V(D)J Reagent Kits v1.1 protocols with Feature Barcode technology, the following products are required in addition to all the products listed above. Refer to the indicated documents for specific guidance.

Chromium Single Cell 5' Feature Barcode Library Kit	16 rxn	1000080
Single Index Kit N Set A	96 rxns	1000212
DOCUMENTS		
User Guide : Chromium Next GEM Single Cell V(D)J Reagent Kits v1.1 with Feature Barcode technology for Cell Surface Protein (CG000208)		
Demonstrated Protocol : Cell Surface Protein Labeling for Single Cell RNA Sequencing Protocols (CG000149)		
Demonstrated Protocol : Cell Labeling with Dextramers for Single Cell RNA Sequencing Protocols (CG000203)		

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Contact:
support@10xgenomics.com
 10x Genomics
 6230 Stoneridge Mall Road
 Pleasanton, CA 94588 USA

