



GETTING STARTED WITH 10X GENOMICS SINGLE CELL PRODUCTS

Before You Start

Contact the Genomics Core (genomics@vai.org) or Marie Adams (marie.adams@vai.org) to plan your single cell experiment before growing cells or procuring tissue to ensure availability for processing and the best success for your project. For new projects, we will schedule a **required** meeting for all stakeholders. This includes your lab, Genomics Core staff and Flow Cytometry staff (when appropriate.)

Project Optimization

Obtaining a single cell suspension, an accurate cell count and minimizing dead cells and debris are all vital to a successful project. Optimization may take several tries; we will help you with methods and QC as necessary.

Actual Project Numbers (scRNA 3' v3.1 or 5' v2)

- Starting sample concentration should be between 500–1200 live cells/ μ L as counted by hemocytometer or automated cell counter. Cell viability should be >85%.
- Plan to bring 100K cells total, in a max volume of 110 μ L. Protocols exist for lower cell numbers if needed.
- Library can be made from 500–10,000 cells. Superloading is possible; discuss with Core before proceeding.
- Hashtagging and CITE/REAP seq can be accommodated; Core **MUST** know which antibody set is being used.
- 10x recommends 25K sequencing reads/cell + 5K reads/cell for CITE, VDJ or Hash library; however, in our experience, 30-40K reads/cell produces a cleaner dataset.

Actual Project Numbers (scATAC)

- Starting concentration should be 3000 nuclei/ μ L.
- Plan to bring 200,000 nuclei total in 10X nuclei buffer. Protocols exist for lower nuclei numbers if needed.
- Each library can be made from 500–10,000 nuclei.
- 10x recommends 25K sequencing reads/cell; however, in our experience, 30–40K reads/cell produces a cleaner dataset.

Costs

scRNA library prep (both 3' v3.1 and 5' v2 available):

- **Setup fee:** \$368 for 1–8 libraries (additional setup fee applies for each group of 8 samples)
- **Per sample cost:** \$1682
- **Hashing, CITE library additional cost (3' or 5'):** \$100
- **VDJ library additional cost (5' only):** \$100

scATAC library prep

- **Setup fee:** \$368 for 1–8 libraries (additional setup fee applies for each group of 8 samples)
- **Per sample cost:** \$1594

Sequencing

Cost is based upon number of cells and sequencing depth for experiment; here are some examples:

scRNA 3' v3.1:

Note: 10x 3' libraries require a custom read length and cannot be pooled with other non 3' libraries for sequencing

- **NextSeq:** 4000 cells x 50K reads/cell = 1 mid-output, 150 cycle flowcell = \$1643 (\$0.41/cell)
- **NovaSeq:** 8000 cells x 50K reads/cell = 1 lane, SP 100 cycle flowcell = \$2487 (\$0.31/cell)
- **NovaSeq:** 30,000 cells x 50K reads/cell = 1 full S1 100 cycle flowcell = \$6355 (\$0.21/cell)

scRNA 5' v2:

- **NovaSeq:** 8000 cells x 50K reads/cell, pooled on S2 100 cycle flowcell = \$1390 (\$0.18/cell)

scATAC:

- **NextSeq:** 4000 cells x 50K reads/cell = 1 mid-output 150 cycle flowcell = \$1643 (\$0.41/cell)
- **NovaSeq:** 8000 cells x 50K reads/cell, pooled on S2 100 cycle flowcell = \$1390 (\$0.18/cell)