# Supplemental Materials

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1.	Cellranger bash commands and parameter files	. 1
2.	ScRNA Benchmarking bash command and parameter file	. 2
	ClubAC bash command and parameter file	
.).	ClubAC pash command and parameter tile	ົ

## 1. Cellranger bash commands and parameter files

#### Bash commands:

cellranger count --id=10X\_20\_010\_GE\_HTO -- transcriptome=/home/david.mentrup/cellRanger/refdata-cellranger-hg19-3.0.0 --libraries GE\_010\_HTO\_Hash\_library.csv -- feature-ref GE\_010\_HTO\_Hash\_feature\_ref.csv --expect-cells=10000 --localcores=24 --localmem=64

cellranger count --id=10X\_20\_011\_GE\_HTO -- transcriptome=/home/david.mentrup/cellRanger/refdata-cellranger-hg19-3.0.0 --libraries GE\_011\_HTO\_Hash\_library.csv -- feature-ref GE\_011\_HTO\_Hash\_feature\_ref.csv --expect-cells=5100 --localcores=24 --localmem=64

cellranger count --id=10X\_20\_010\_GE\_Epi --transcriptome=/home/david.mentrup/cellRanger/refdata-cellranger-hg19-3.0.0 --libraries GE\_010\_Epi\_CITE\_\_library.csv --feature-ref GE\_010\_EPI\_CITE\_feature\_ref.csv --expect-cells=10000 --localcores=24 --localmem=64

### GE\_010\_HTO\_Hash\_library.csv

fastqs,sample,library\_type

/home/david.mentrup/ProjectCITE/data/Run01/10X\_20\_OK\_05\_R59/fastq\_files/P16358\_1001/02-FASTQ/200403\_A00621\_0207\_AHKYCGDRXX,P16358\_1001,Gene Expression /home/david.mentrup/ProjectCITE/data/Run01/10X\_20\_OK\_05\_R59/fastq\_files/P16358\_1003/02-FASTQ/200403\_A00621\_0207\_AHKYCGDRXX,P16358\_1003,Antibody Capture

#### GE\_011\_HTO\_Hash\_library.csv

fastqs,sample,library\_type

/home/david.mentrup/ProjectCITE/data/Run01/10X\_20\_OK\_05\_R59/fastq\_files/P16358\_1002/02-FASTQ/200403\_A00621\_0207\_AHKYCGDRXX/,P16358\_1002,Gene Expression /home/david.mentrup/ProjectCITE/data/Run01/10X\_20\_OK\_05\_R59/fastq\_files/P16358\_1004/02-FASTQ/200403\_A00621\_0207\_AHKYCGDRXX/,P16358\_1004,Antibody Capture

### GE 010 Epi CITE library.csv

fastqs,sample,library\_type

/home/david.mentrup/ProjectCITE/data/Run01/10X\_20\_OK\_05\_R59/fastq\_files/P16358\_1001/02-FASTQ/200403\_A00621\_0207\_AHKYCGDRXX/,P16358\_1001,Gene Expression /home/david.mentrup/ProjectCITE/data/Run01/10X\_20\_OK\_05\_R59/fastq\_files/P16358\_1005/02-FASTQ/200403\_A00621\_0207\_AHKYCGDRXX/,P16358\_1005,Antibody Capture

#### GE 010 HTO Hash feature ref.csv

id,name,read,pattern,sequence,feature\_type

H1, Hash1 TotalSeqB, R2, 5PNNNNNNNNN(BC)NNNNNNNNN, GTCAACTCTTTAGCG, Antibody Capture

H2,Hash2\_TotalSeqB,R2,5PNNNNNNNNNNN(BC)NNNNNNNNN,TGATGGCCTATTGGG,Antibody Capture

H3, Hash3\_TotalSeqB, R2, 5PNNNNNNNNNN(BC)NNNNNNNNN, TTCCGCCTCTCTTTG, Antibody Capture

#### GE\_011\_HTO\_Hash\_feature\_ref.csv

id,name,read,pattern,sequence,feature type H3, Hash3 Total Seq B, R2, 5PNNNNNNNNNNNN(BC)NNNNNNNNNNTTCCGCCTCTCTTTG, Antibody Capture

#### GE 010 Epi CITE library.csv

id,name,read,pattern,sequence,feature\_type CD19,CD19 TotalA,R2,^(BC),CTGGGCAATTACTCG,Antibody Capture CD3,CD3\_TotalA,R2,^(BC),CTCATTGTAACTCCT,Antibody Capture CD16,CD16\_TotalA,R2,^(BC),AAGTTCACTCTTTGC,Antibody Capture CD4,CD4\_TotalA,R2,^(BC),TGTTCCCGCTCAACT,Antibody Capture CD11c,CD11c TotalA,R2,^(BC),TACGCCTATAACTTG,Antibody Capture CD56\_NCAM,CD56\_TotalA,R2,^(BC),TCCTTTCCTGATAGG,Antibody Capture CD14,CD14 TotalA,R2,^(BC),TCTCAGACCTCCGTA,Antibody Capture CD8,CD8\_TotalA,R2,^(BC),GCGCAACTTGATGAT,Antibody Capture CD45,CD45\_TotalA,R2,^(BC),TCCCTTGCGATTTAC,Antibody Capture CD34,CD34 TotalA,R2,^(BC),GCAGAAATCTCCCTT,Antibody Capture CD15,CD15 TotalA,R2,^(BC),TCACCAGTACCTAGT,Antibody Capture

#### 2. ScRNA Benchmarking bash command and parameter file

#### Bash command:

snakemake --configfile BMMC\_small\_config.yml --cores 16 --use-singularity

#### BMMC small config.yml

output dir: BMMC small/Output

datafile: BMMC\_small/BMMC\_Ref\_count.csv labfile: BMMC\_small/BMMC\_Ref\_Label.csv

column: 1

number of features: 2000 tools\_to\_run:

- singleCellNet
- SingleR
- kNN9
- kNN50
- scVI
- scmapcell
- Seurat\_CCA
- Seurat\_PCA
- SVM
- SVM\_rejection
- LDA
- LDA\_rejection
- RF
- CHETAH
- scmapcluster
- scID

- SingleR
- NMC

## 3. ClubAC bash command and parameter file

#### Bash command:

snakemake --configfile BMMC\_AML\_5000genes\_config.yml --cores 16 --use-singularity

#### BMMC\_AML\_5000genes\_config.yml

output\_dir: BMMC\_AML/output\_5000genes

refdatafile: BMMC\_AML/bmmc\_Ref

reflabelfile: BMMC\_AML/BMMC\_Ref\_Label.csv testdatafile: BMMC\_AML/AML\_S01\_CITE testclusterfile: BMMC\_AML/AML\_Cluster.csv

column: 1

number\_of\_features: 5000

tools\_to\_run:

- scmapcell
- Seurat\_CCA
- Seurat\_PCA
- SVM
- SVM\_rejection
- LDA
- LDA\_rejection
- RF
- CHETAH
- scmapcluster
- scID
- SingleR