SNARE-seq2 Analysis - version 2.0

Part 1;

Generate docker container. A /media/Scratch\_SSD\_Voyager/docker\_scratch folder have been specifically created for docker users.

$ docker run --name zhanglab\_omics -it -d -v /media/Scratch\_SSD\_Voyager/docker\_scratch:/scratch zhanglab/omics\_v2

Part 2:

Generate the fastq files.

RNA:

$ ./generate\_fastq.sh -d /media/SeqStore/MiSeqOutput/190803\_M00159\_0229\_000000000-CJRNF/ -m y70,i6n2,y102 -s ../config/RNA\_SampleSheet.csv -c 12 -b 0 -l 1 -o Miseq\_20190803\_CTRI\_LAPMAP\_20190731/raw\_fastq &> Miseq\_20190803\_CTRI\_LAPMAP\_20190731/bcl2fastq.log

Chromatin:

$ ./generate\_fastq.sh -d /media/SeqStore/MiSeqOutput/190527\_M00159\_0193\_000000000-CBYW2/ -m y75,y98,i8,y75 -s ../config/CHROMATIN\_SampleSheet.csv -c 12 -b 0 -l 1 -o Miseq\_20190527\_hBICCN\_20190523/raw\_fastq &> Miseq\_20190527\_hBICCN\_20190523/bcl2fastq.log

Part 3:

Generate the <sample>.list files

ie: "**mBICCN\_20190730A.list**"

$ cat mBICCN\_20190730A.list

A1 A2 A3 A4 A5 A6 A7 A8 A9 A10 A11 A12

E1 E2 E3 E4 E5 E6 E7 E8 E9 E10 E11 E12

Part 4:

Modify analyze shell scripts

RNA variables (**analyze\_rna.sh**):

**pools**=”...”

**samples**="mBICCN\_20190730A" # match the prefix names for list file.

**config**="/scratch/SNARE2\_MiSeqRuns/config"

**raw\_fastq**="..."

**star\_index**="/scratch/refdata-cellranger-GRCh38-3.0.0/star/"

**gtf\_file**="/scratch/refdata-cellranger-GRCh38-3.0.0/genes/genes.gtf"

CHROMATIN variables (**analyze\_chromatin.sh**):

**raw\_fastq\_dir**="..."

**config**="/scratch/SNARE2\_MiSeqRuns/config"

**pools**="..."

**samples**="mBICCN\_20190730A" # match the prefix names for list file.

**genome\_mmi**="/scratch/refdata-cellranger-atac-GRCh38-1.1.0/fasta/genome.fa.mmi"

**genome\_csize**="/scratch/refdata-cellranger-atac-GRCh38-1.1.0/fasta/genome.fa.fai"

**genome\_name**="hg38" # *hg38* or *mm10* for human and mouse.

Part 5:

Attach to docker container. If using a previous container, run “docker start <container\_name>” first.

$ docker attach zhanglab\_omics

Part 6:

* Make a <rna analysis> or <chromatin\_analysis> folder in the docker’s scratch space.
* Copy all <samples>.list files to this folder. RNA and Chromatin parts have the same <samples>.list files.
* Copy the modified analyze\_rna.sh or analyze\_chromatin.sh to this folder.
* Run the script for each analysis.
* “.rds” files from RNA analysis can be analyzed with the “Seurat” package in R.
* “.snap” files from CHROMATIN analysis can be analyzed with “SnapATAC” package in R.

RNA:

$ sh analyze\_rna.sh &> analyze\_rna.log &

CHROMATIN:

$ sh analyze\_chromatin.sh &> analyze\_chromatin.log &