Requirements:

1. Unmapped SmartSeq2 FASTQ files
2. SmartSeq2 barcode Excel file
3. mus\_index folder
4. barcode\_prep.R
5. merge\_barcode.sh
6. qc\_align.sh
7. tpm\_fpkm.R
8. Conda environment for alignment. Required packages:
   1. Fastqc
   2. Multiqc
   3. Salmon

Workflow

1. Download or move all required files to project folder (ie. SN0189514)
2. Prepare barcode file with barcode\_prep.R
   1. Rscript barcode\_prep -f [SmartSeq2 barcode Excel filename]
3. Create merged files with well identifiers using merge\_barcode.sh and sequencing run (ie. HV2HTBGXC)
   1. sh merge\_barcode.sh well\_barcode.txt [sequencing run]
4. Activate conda alignment environment
   1. conda activate smartseq\_qc\_align
5. Conduct quality control and alignment with qc\_align.sh
   1. sh qc\_align.sh