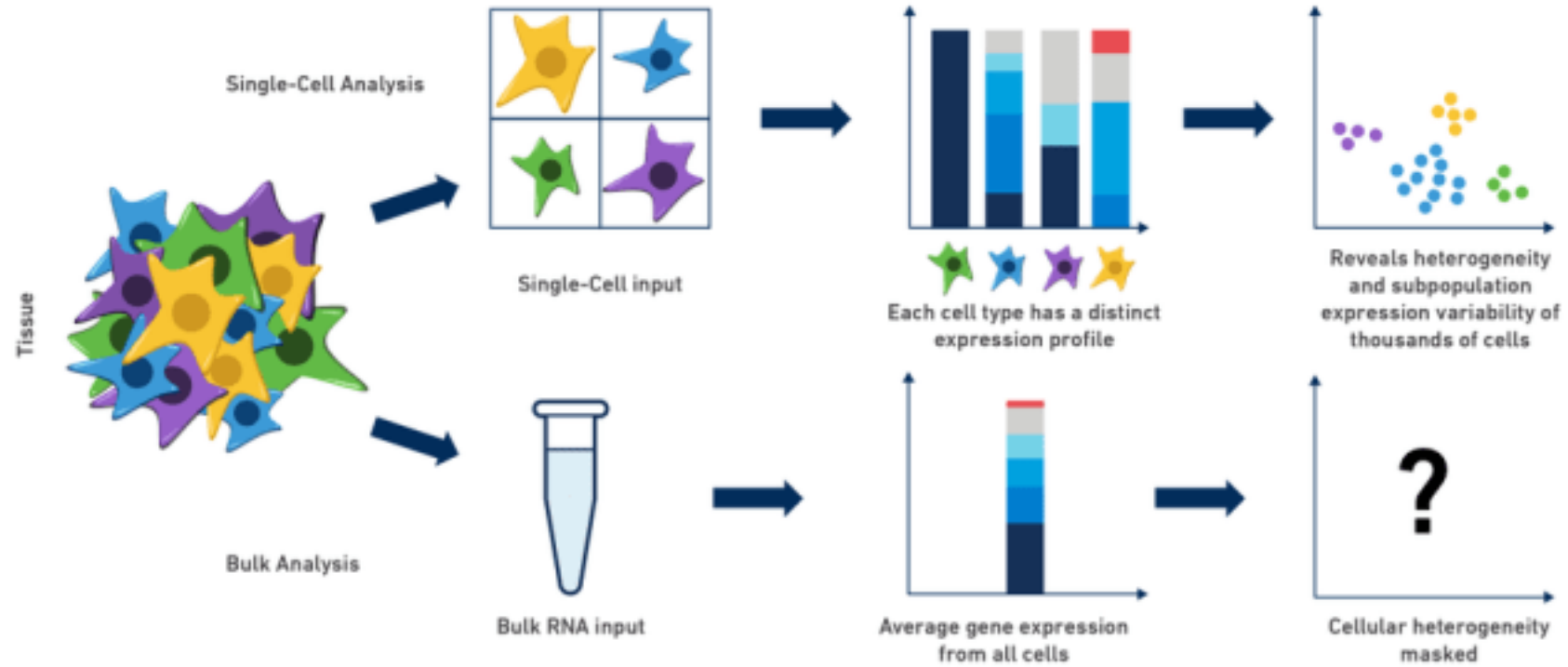


Single Cell RNA Sequencing

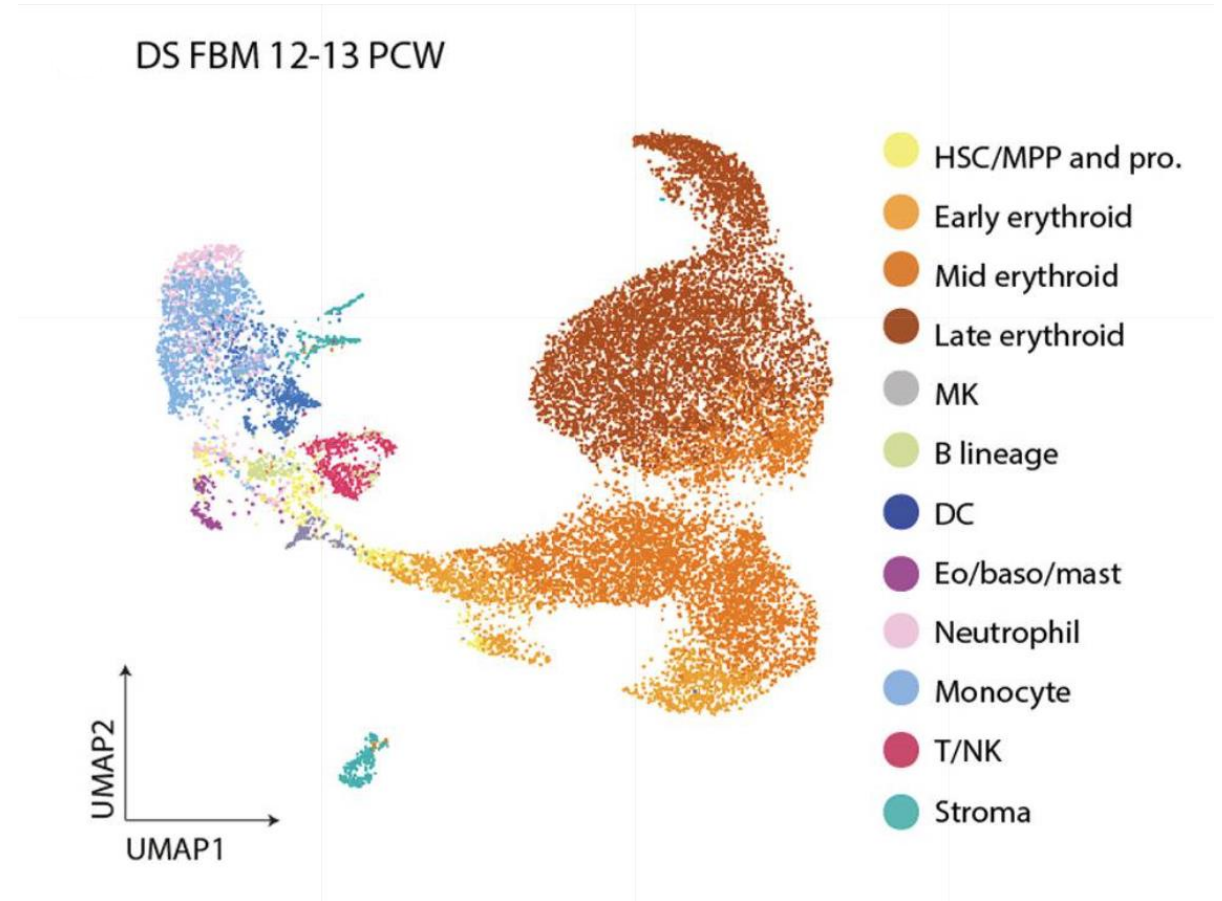
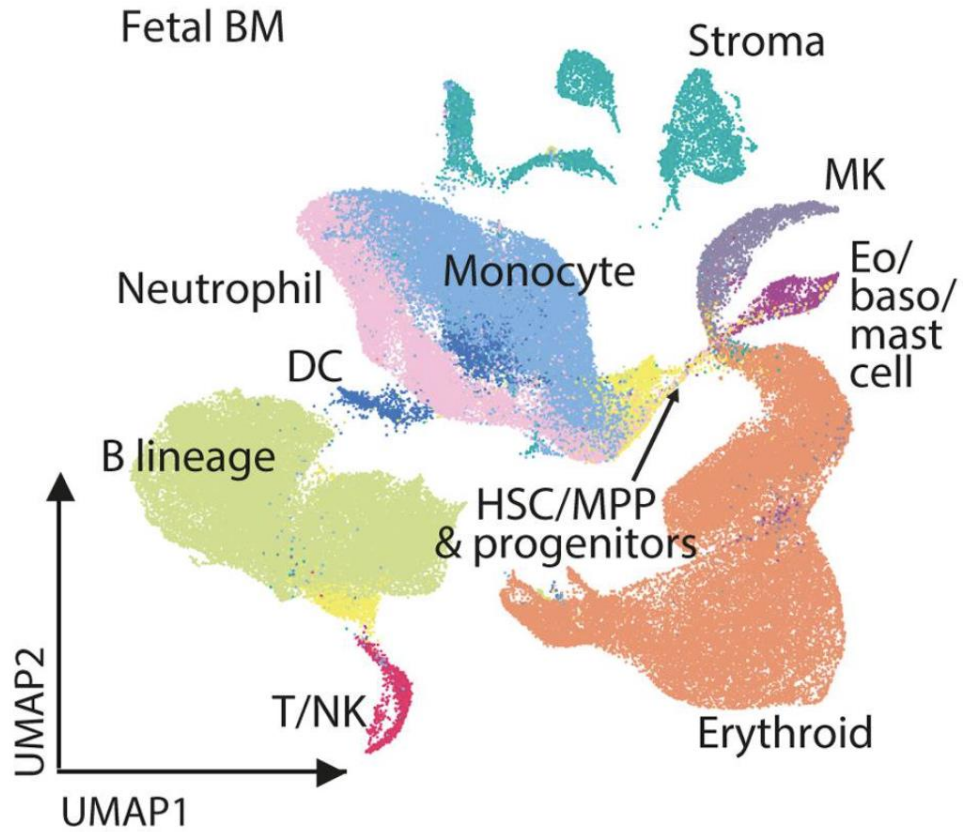
Chris Ozeroff

Short Read 2023

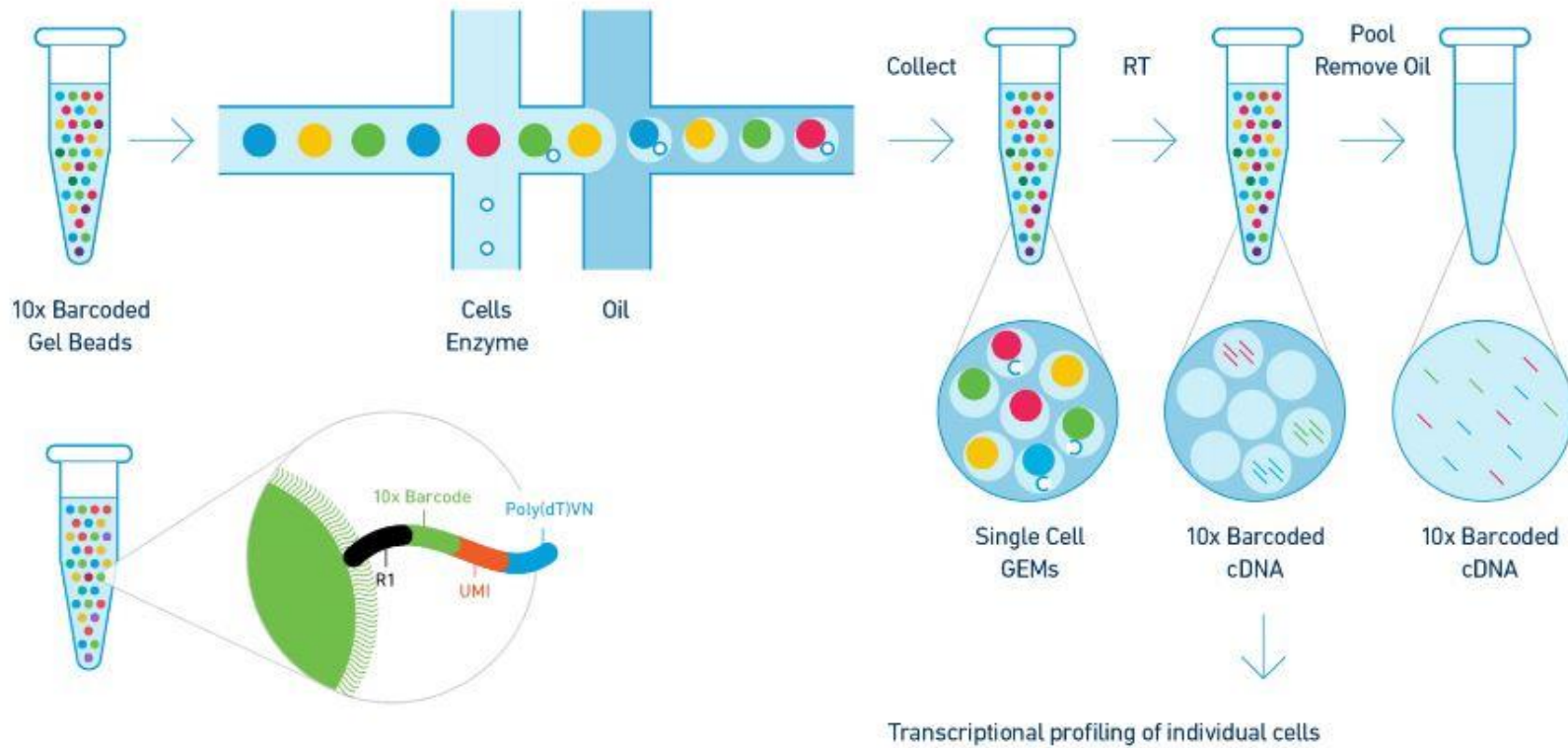
Single Cell RNA Sequencing



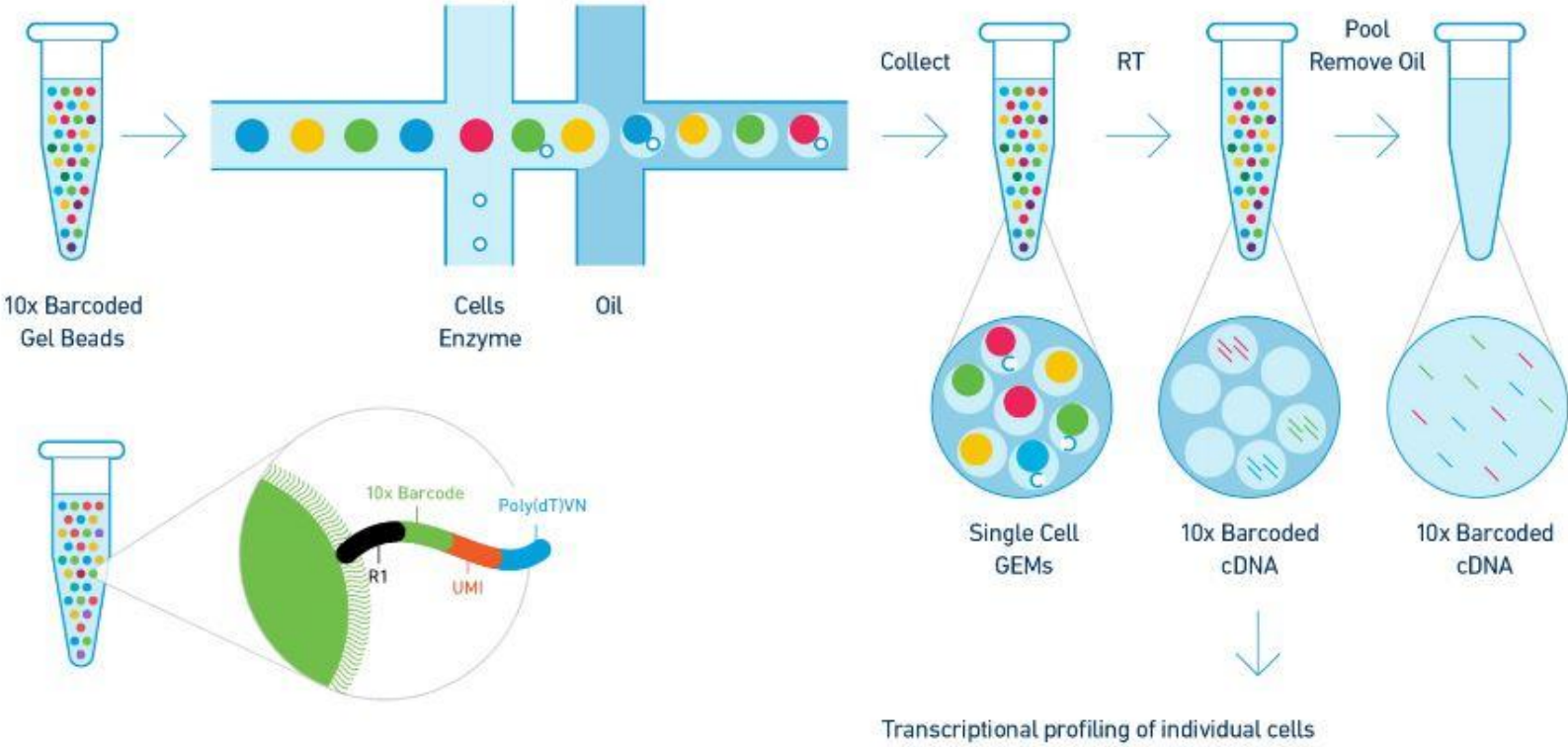
Down Syndrome vs. Typical Fetal Bone Marrow



10X Genomics Droplet Method and Cell Ranger

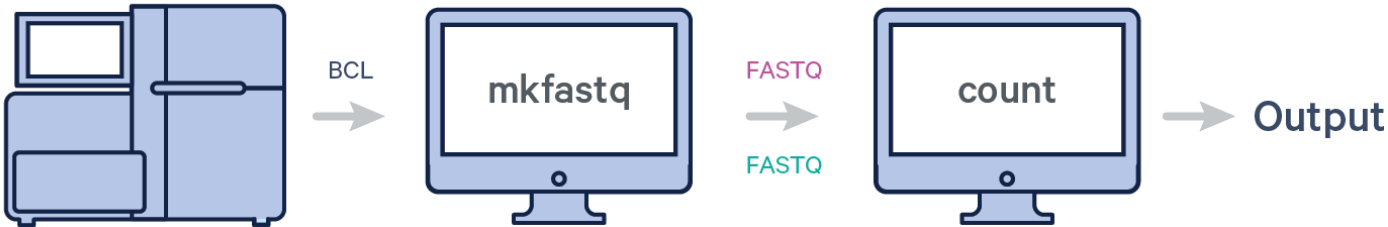


10X Genomics Droplet Method and Cell Ranger



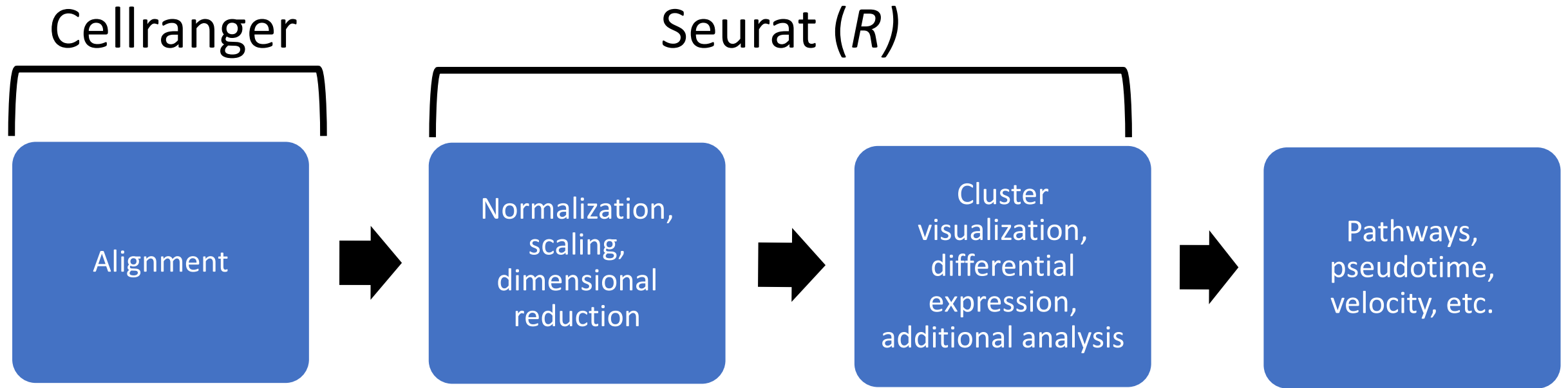
Sequencing

----- Cell Ranger Pipeline -----



Genomics Technology Core,
University of Missouri, 2021

scRNA-seq workflow



SEURAT

R toolkit for single cell genomics

Seurat

Remove debris/doublets



Normalization



Scaled data



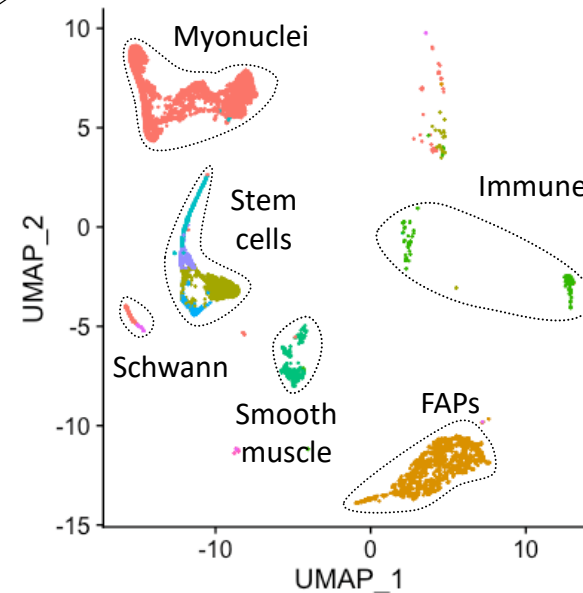
PCA



UMAP/clustering



A Sunday Afternoon on the Island of La Grande Jatte, Georges Seurat (1884)

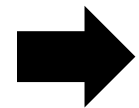


“...named for Georges Seurat to invoke the analogy between the intricate spatial patterning of single cells and a pointillist painting.”
(doi: [10.1038/nbt.3192](https://doi.org/10.1038/nbt.3192))

Seurat workflow

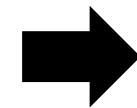
Processing individual Seurat objects

- Add metadata
- Merge Seurat objects
- Normalize data
- Find genes with variable gene expression
- Scale data
- Run PCA reduction



Integrating Seurat objects

Split Seurat objects and identify anchors (genes) to “integrate” objects



Plotting and clustering

UMAP reduction and visualization