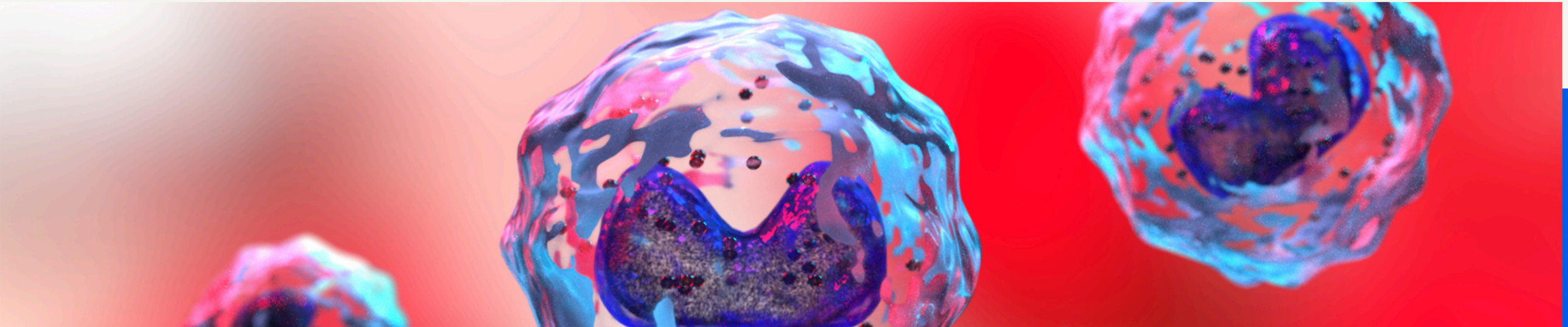
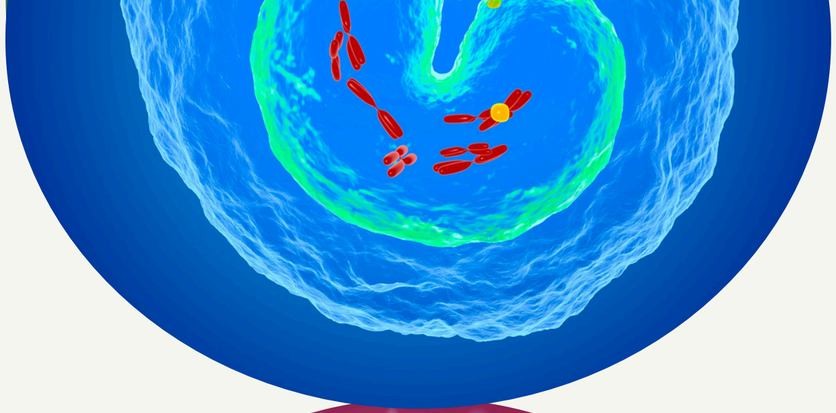
# CBS Project Presentation

Update on the ongoing analysis





# Outline

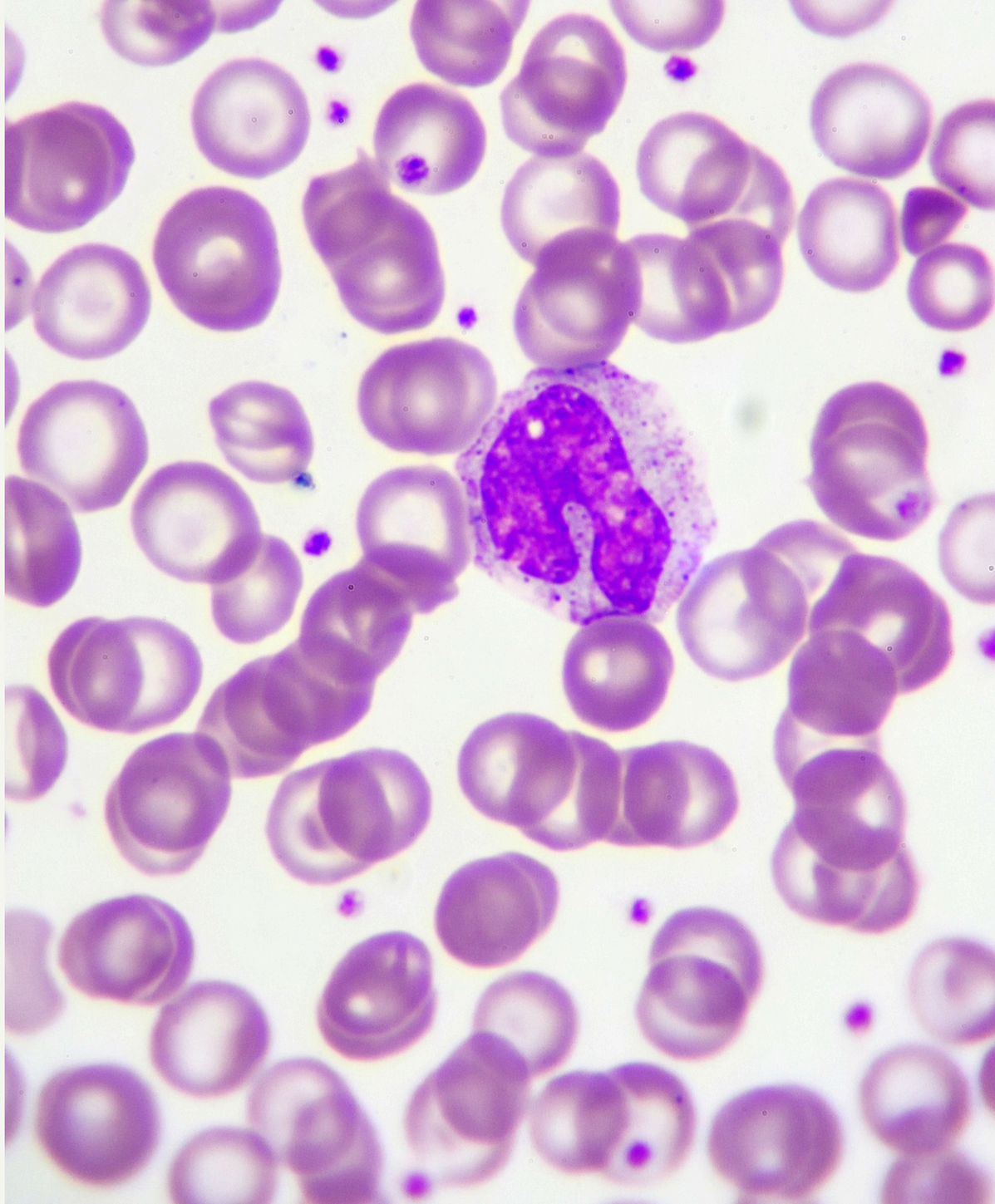
- reminder of the analysis objective (topic)
- comparison with previous results (from one month ago)
- remaining tasks

04.06.2024

# Topic

## — PART 1

MODELING OF COMPLEX BIOLOGICAL SYSTEMS



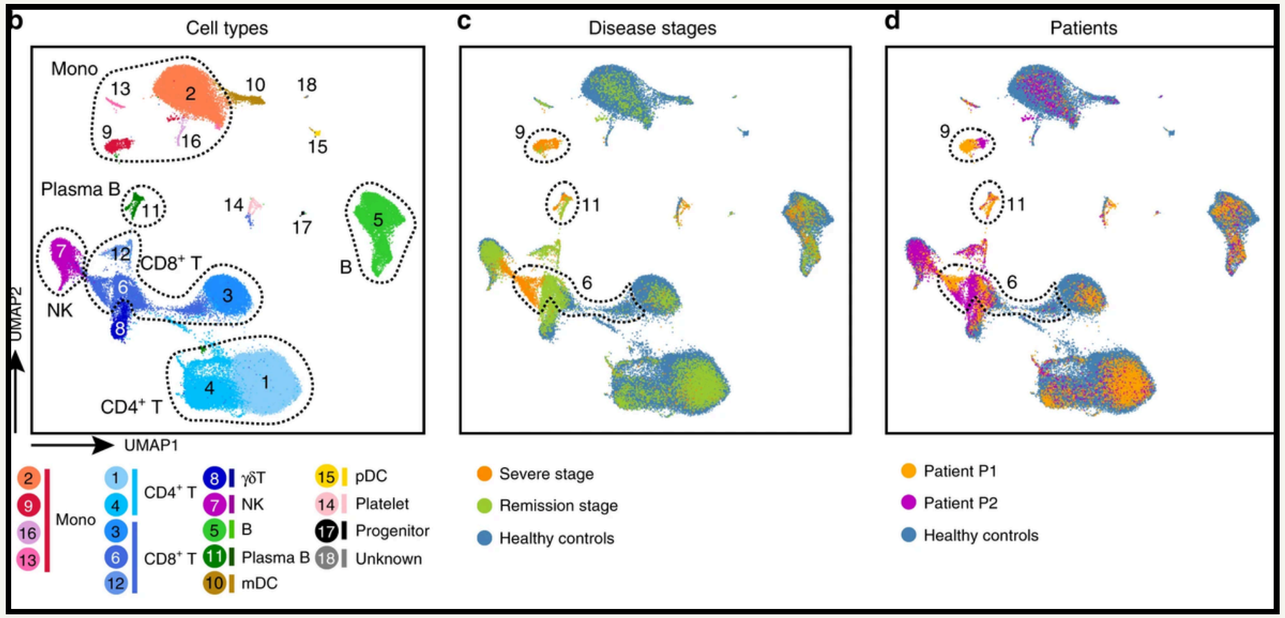
Guo, C., Li, B., Ma, H. et al. Single-cell analysis of two severe COVID-19 patients reveals a monocyte-associated and tocilizumab-responding cytokine storm. Nat Commun 11, 3924 (2020)



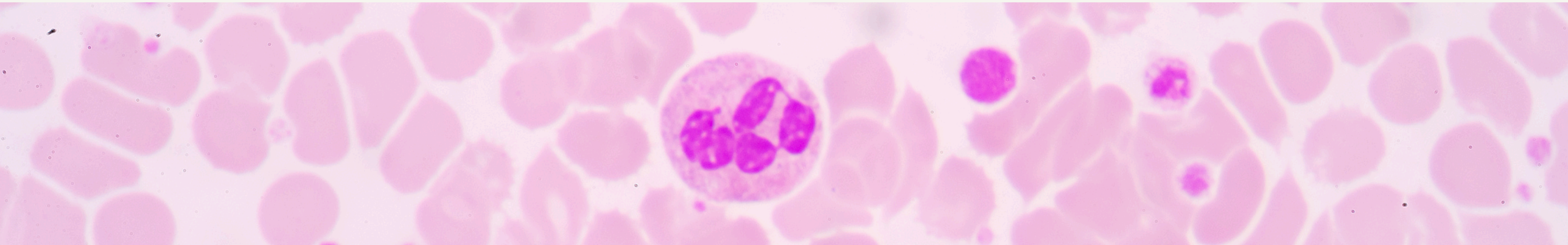
# Topic

## SEVERE STAGE SPECIFIC-MONOCYTE CLUSTER

- they identified the **monocyte subpopulation contributing** to the cytokine storm



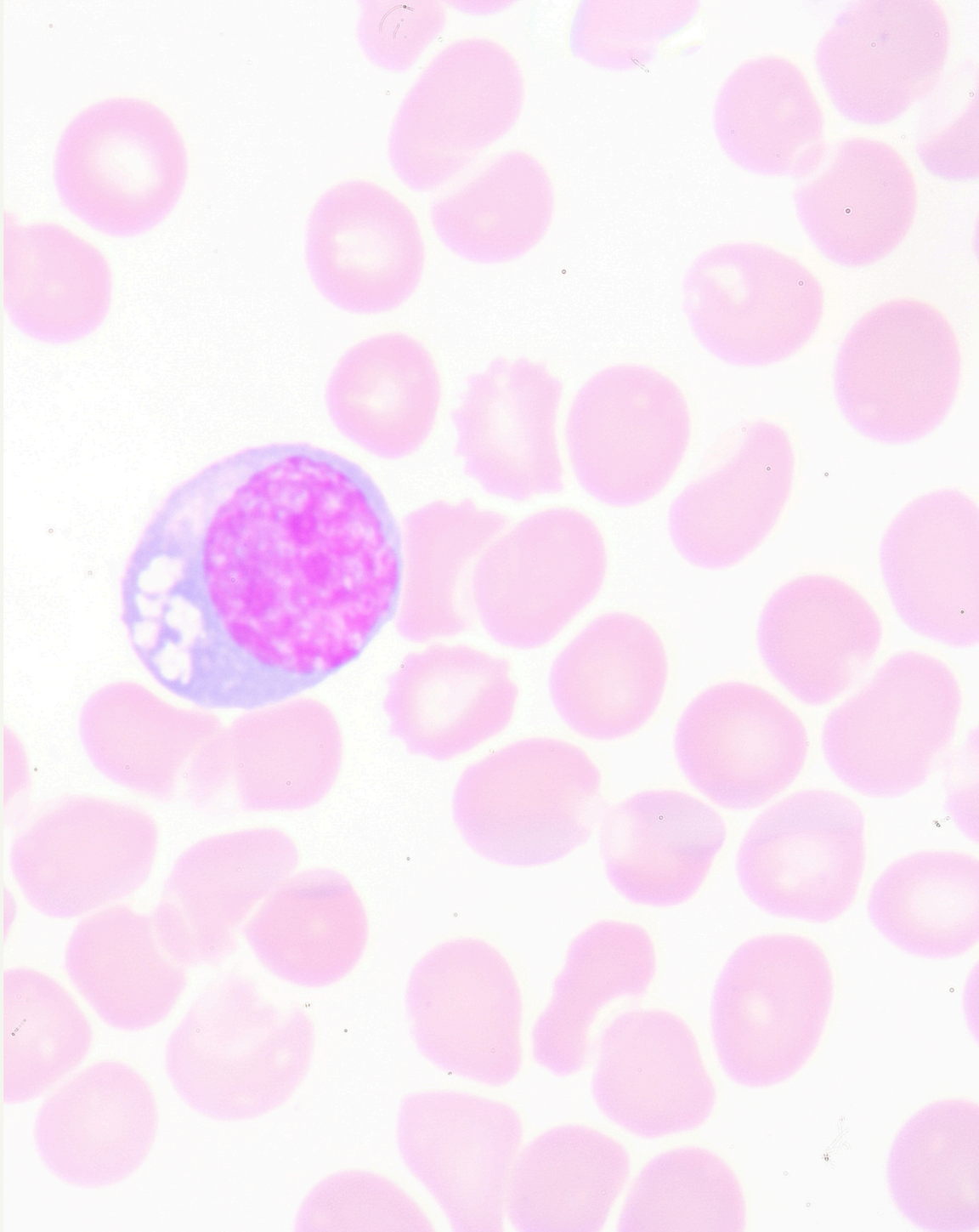

The figures from the original paper show 18 identified cell clusters (b) based on specific marker expression. Cell distribution by disease stage (c) and by patient batch (d) is also presented.



04.06.2024

# Analysis

— PART 2



MODELING OF COMPLEX BIOLOGICAL SYSTEMS

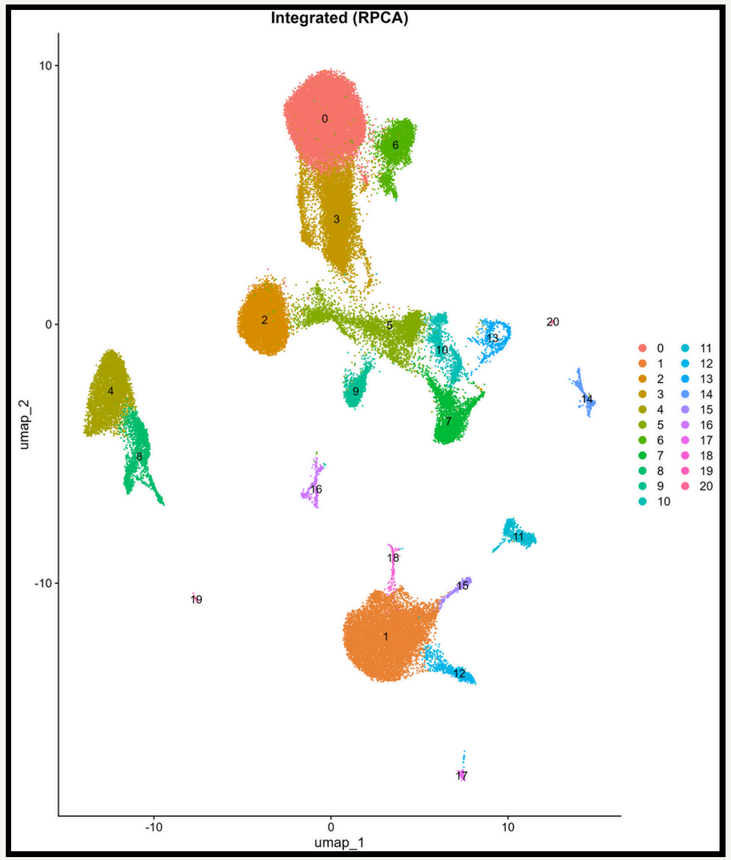
Guo, C., Li, B., Ma, H. et al. Single-cell analysis of two severe COVID-19 patients reveals a monocyte-associated and tocilizumab-responding cytokine storm. Nat Commun 11, 3924 (2020)



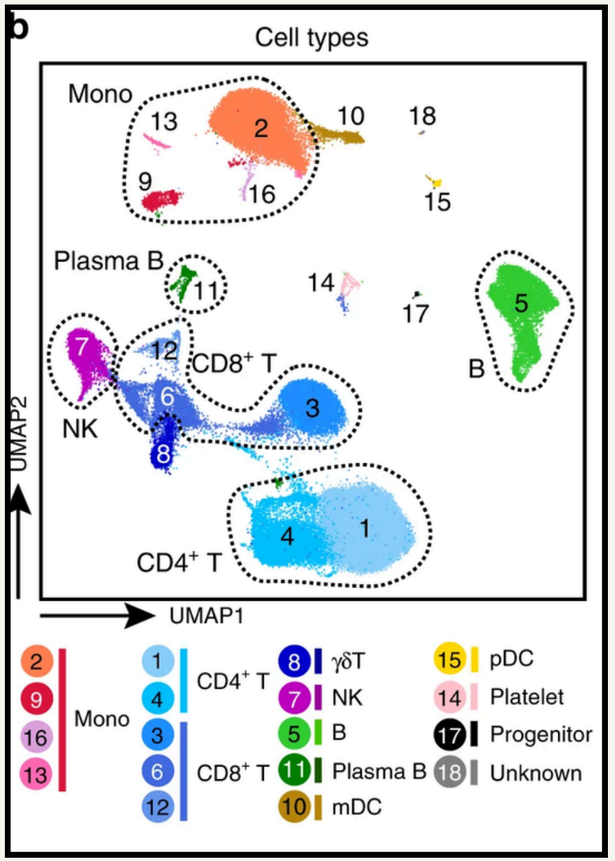
# Obtaining the same results

**I encountered difficulties in removing duplicates and integrating the control and experimental samples.**

- removing the doublets using Seurat was **challenging**, as I was working in R (had to use the reticulate package)
- integration was very time- and memory-consuming, so I used the **RPCA** method to handle it more efficiently.




Using the same or closely matching parameter values, I identified 20 distinct clusters.

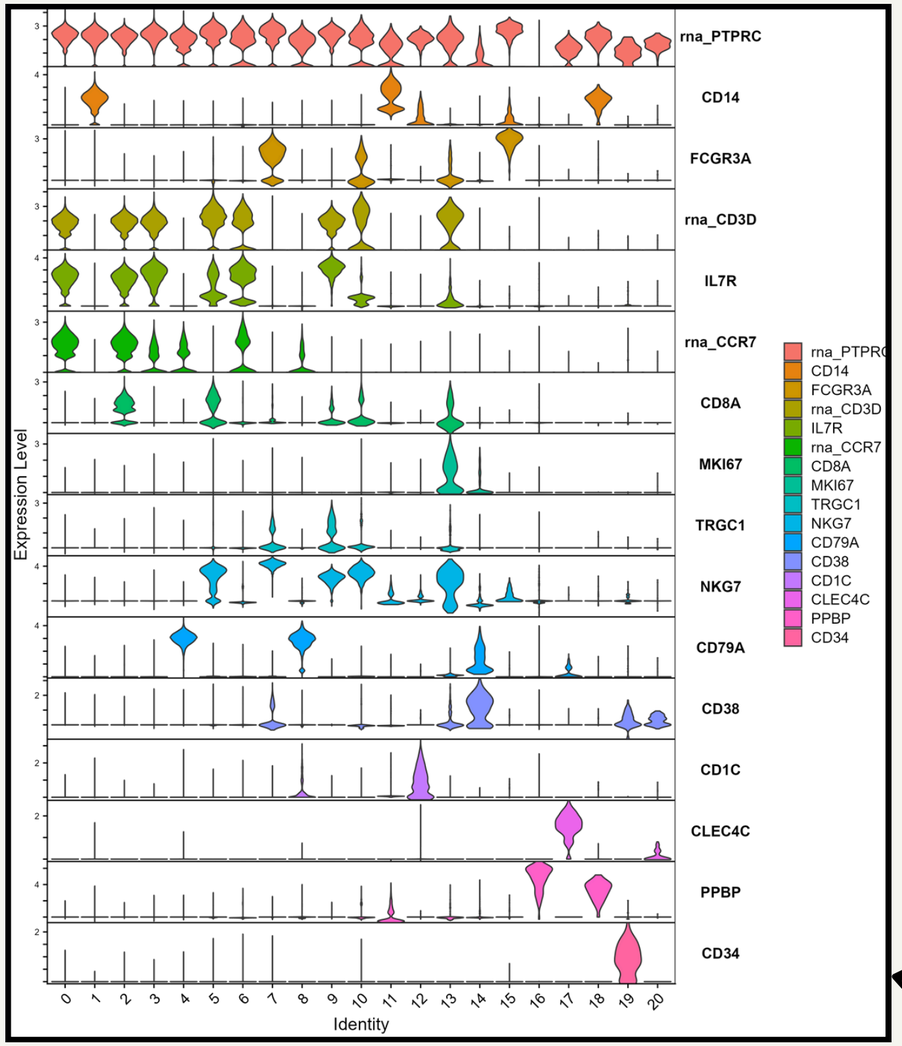


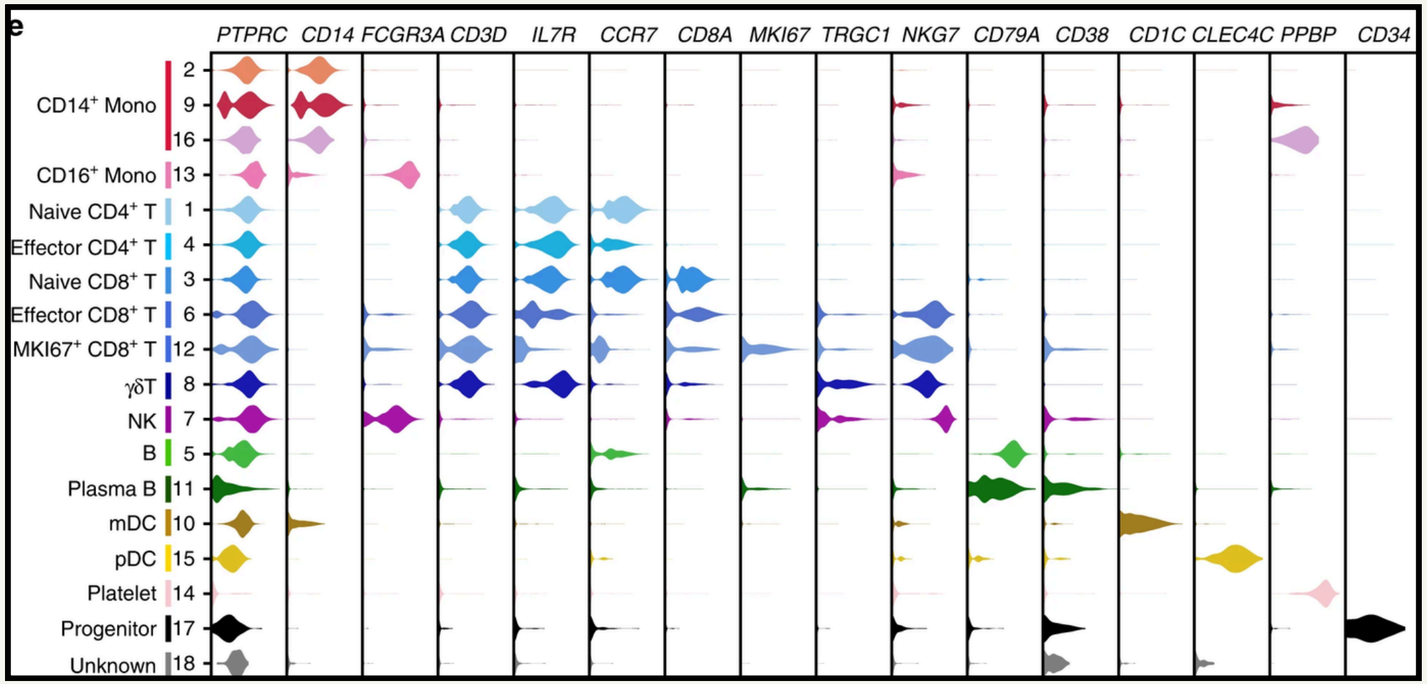

The figures from the original paper show 18 identified cell clusters.

Guo, C., Li, B., Ma, H. et al. Single-cell analysis of two severe COVID-19 patients reveals a monocyte-associated and tocilizumab-responding cytokine storm. Nat Commun 11, 3924 (2020)



# Results: cluster identification based on violin plots




Violin plots of marker expression from the publication

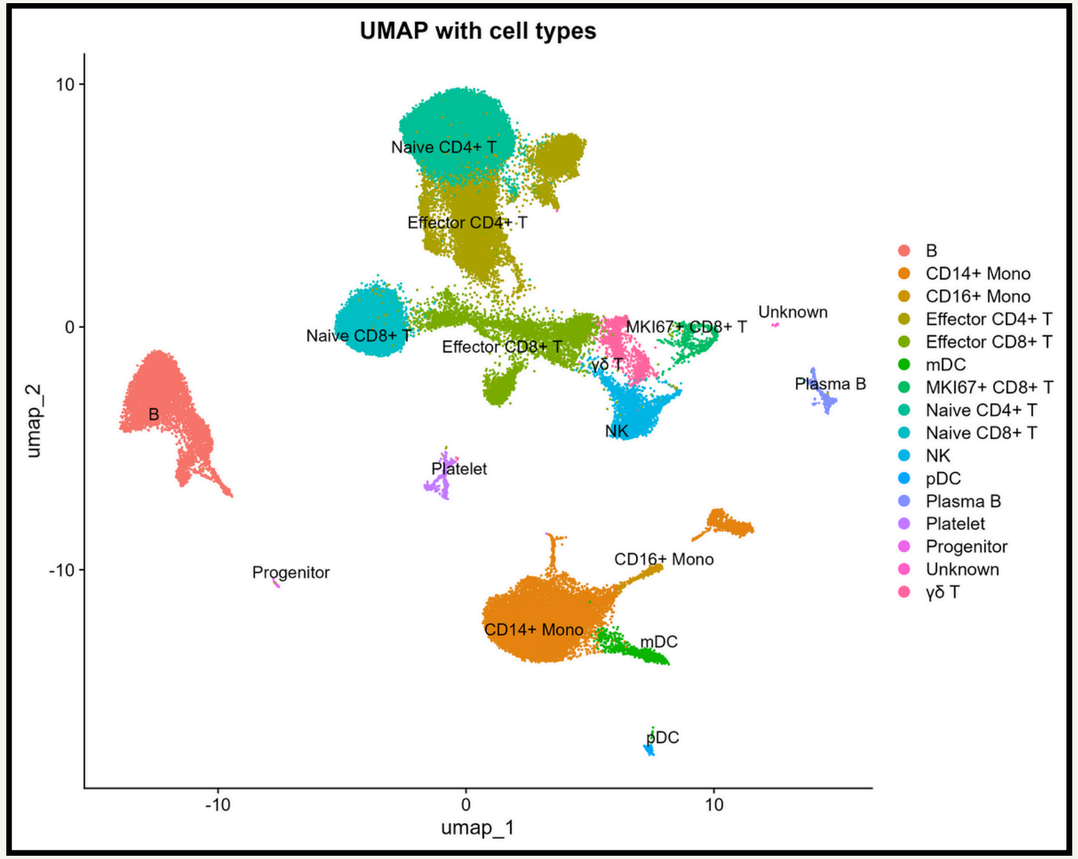
Violin plots of marker expression in my analysis

Guo, C., Li, B., Ma, H. et al. Single-cell analysis of two severe COVID-19 patients reveals a monocyte-associated and tocilizumab-responding cytokine storm. Nat Commun 11, 3924 (2020)

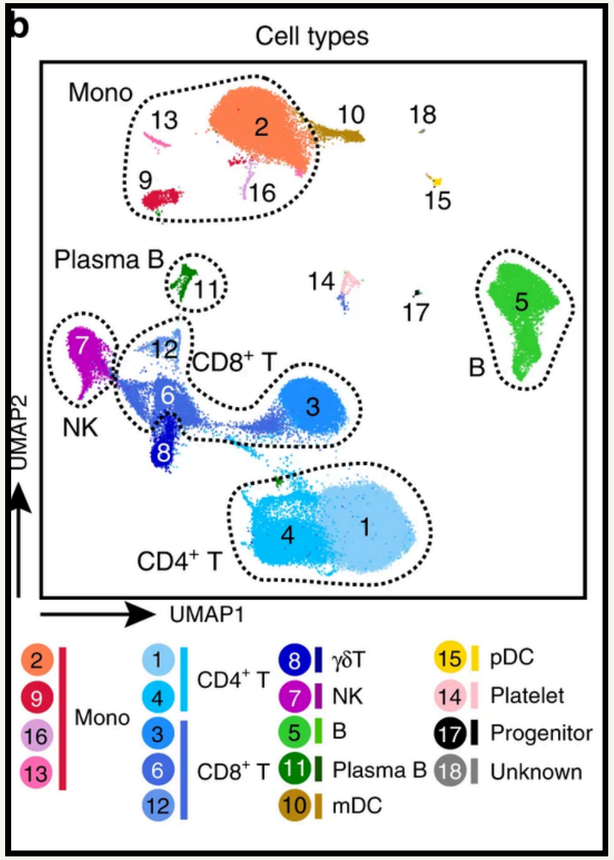


# Obtaining the same results

I struggled to remove the dublets and to integrate control and experimental samples

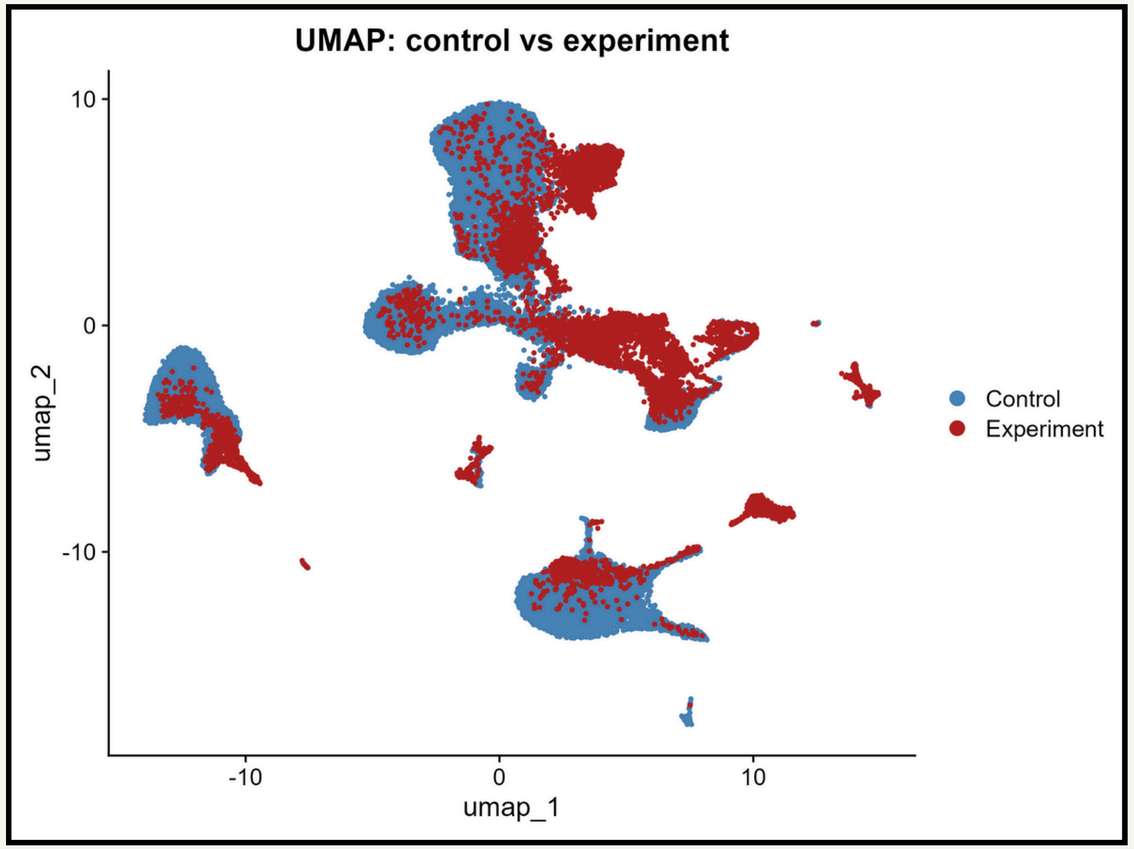



Assignment of cell subtypes to clusters in my work.

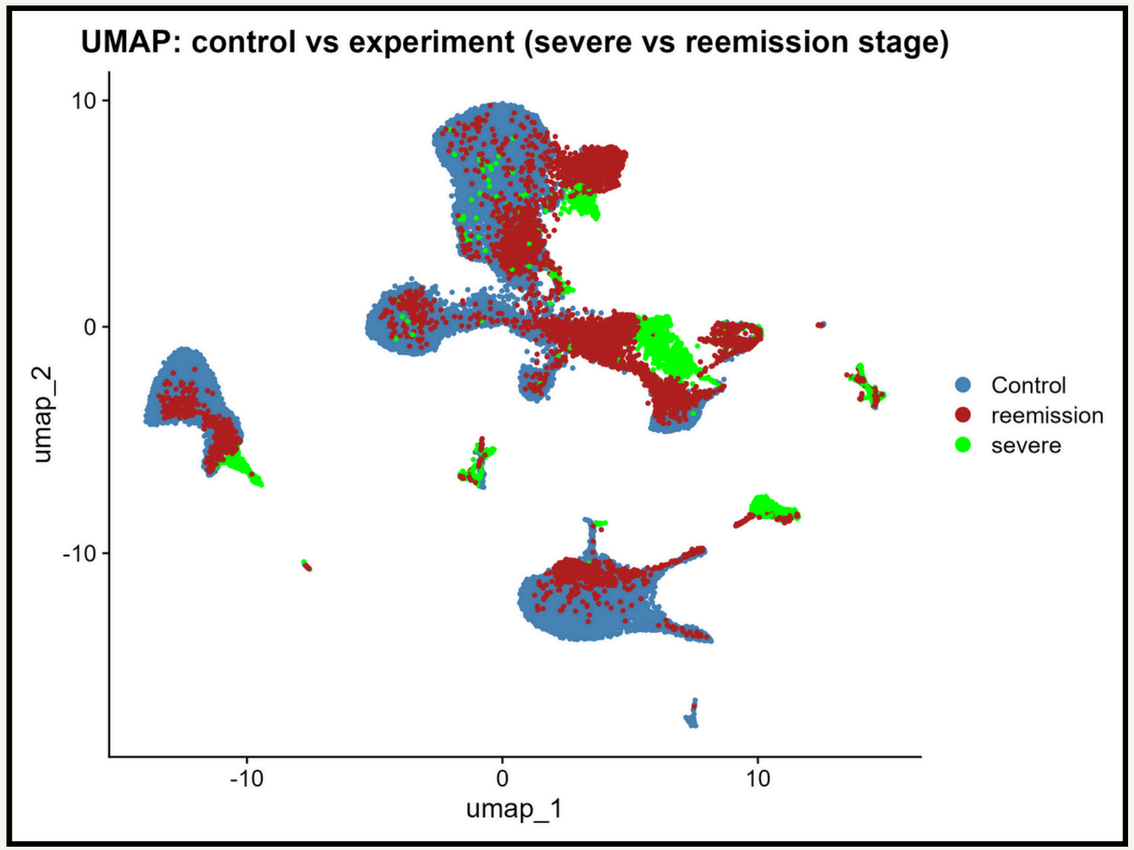



The figures from the original paper show 18 identified cell clusters.

# Results: UMAP plots




Analyzed cells depicted with respect to control and experimental groups.

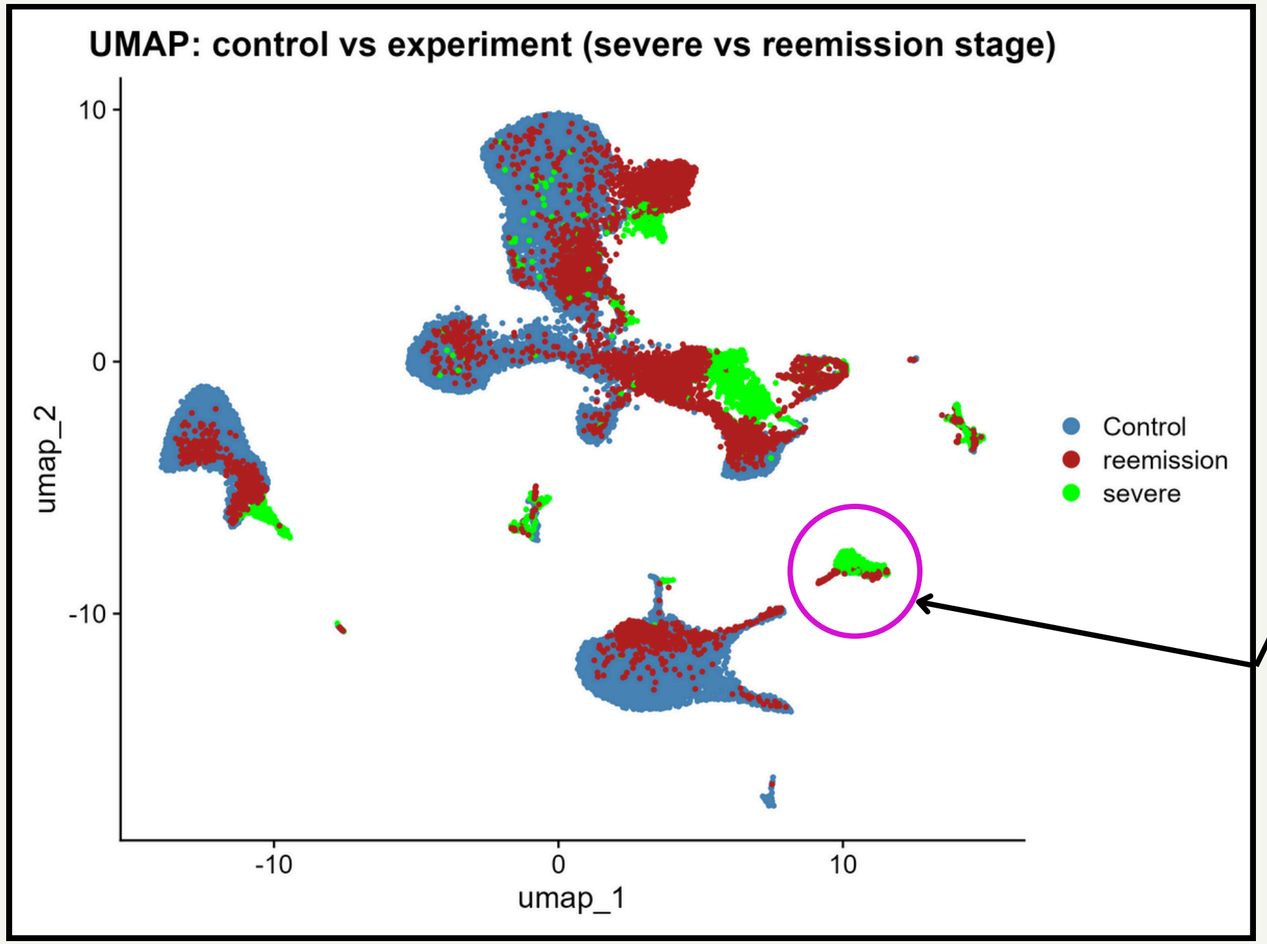



Analyzed cells shown with respect to disease stage.

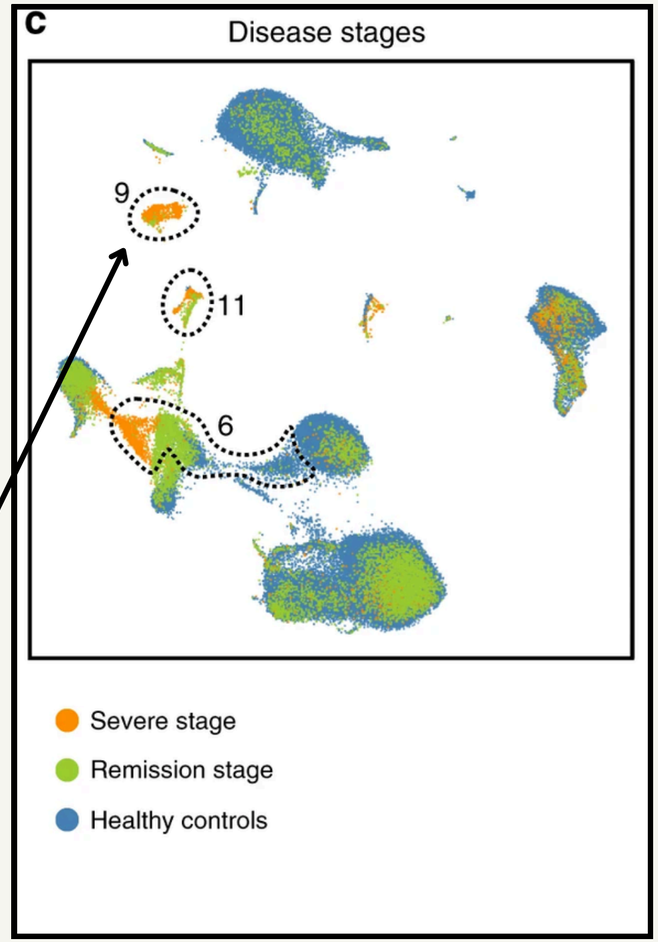
Guo, C., Li, B., Ma, H. et al. Single-cell analysis of two severe COVID-19 patients reveals a monocyte-associated and tocilizumab-responding cytokine storm. Nat Commun 11, 3924 (2020)



# Results: obtaining the monocyte cluster



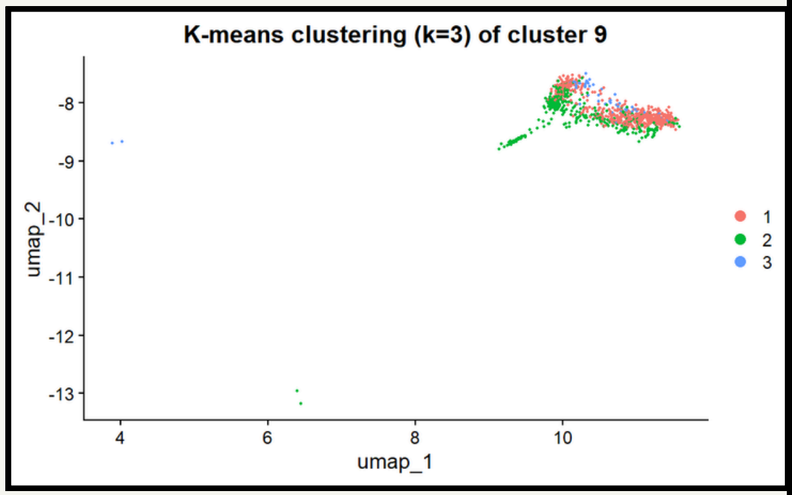

Analyzed cells shown with respect to disease stage.

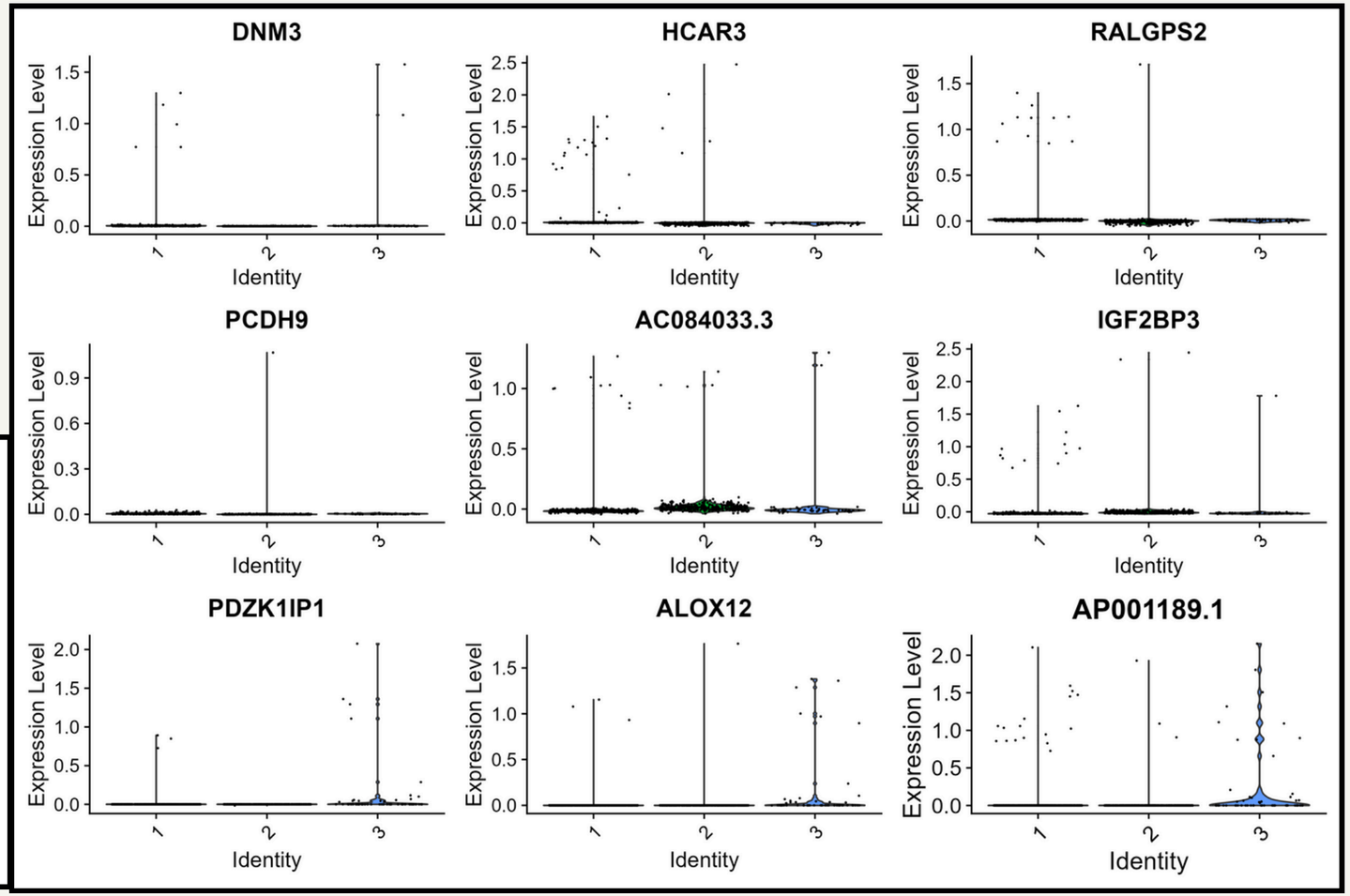



Cell distribution by disease stage from the original paper

# Results: monocyte cluster characterization

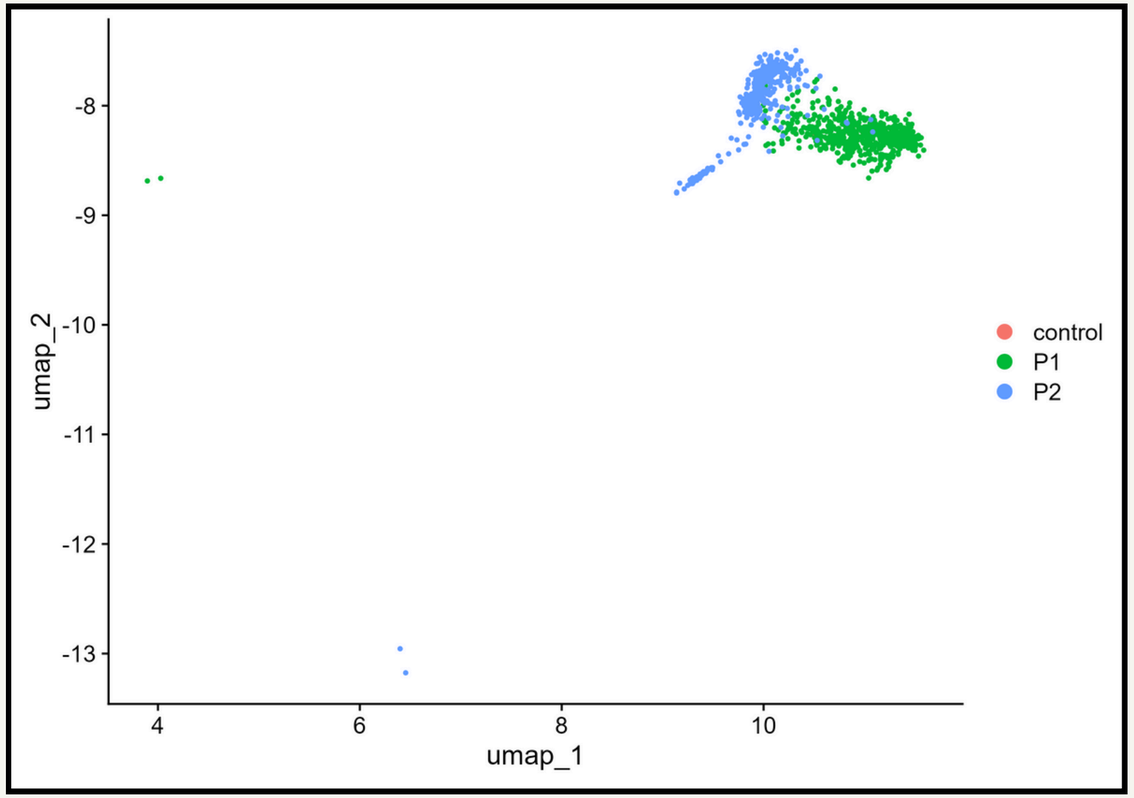
- cluster is **homogenous**
- small changes in gene expression
- AP001189.1 - unkown function, leucine rich repeat containing membrane protein



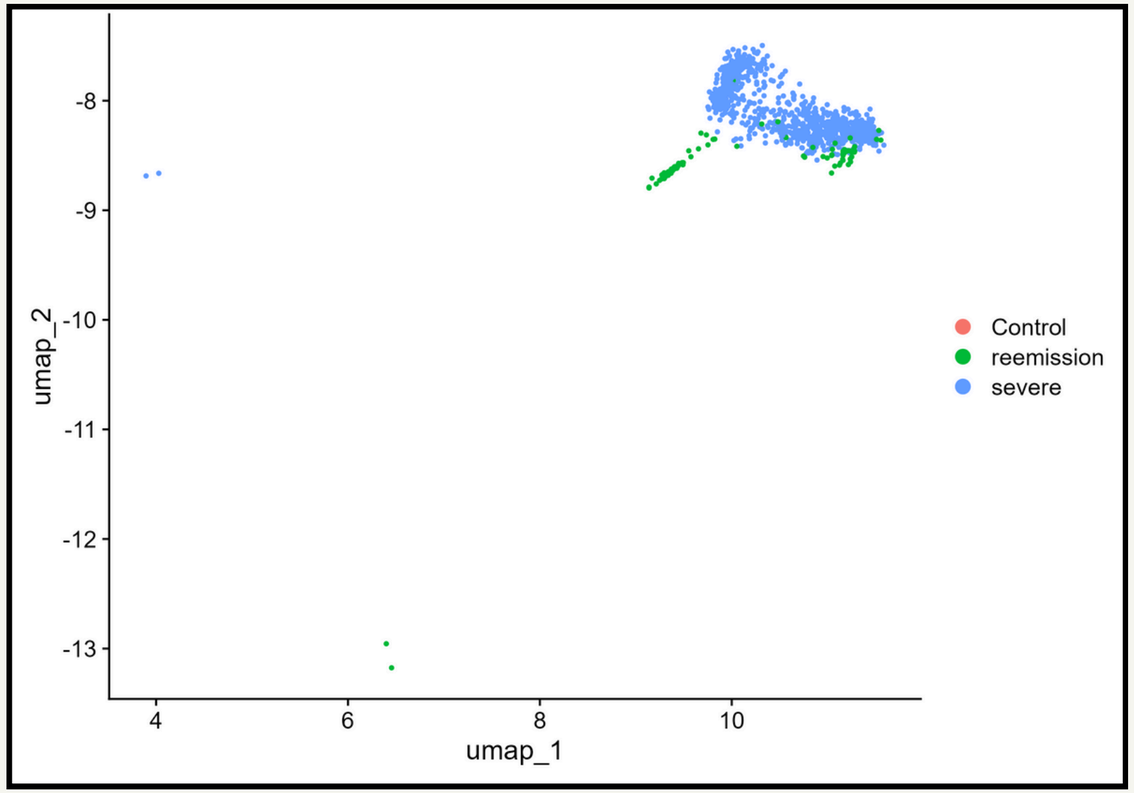




ViolinPlots of 9 most differentially expressed markers in each of 3 monocyte subclusters.

# Results: monocyte cluster characterization



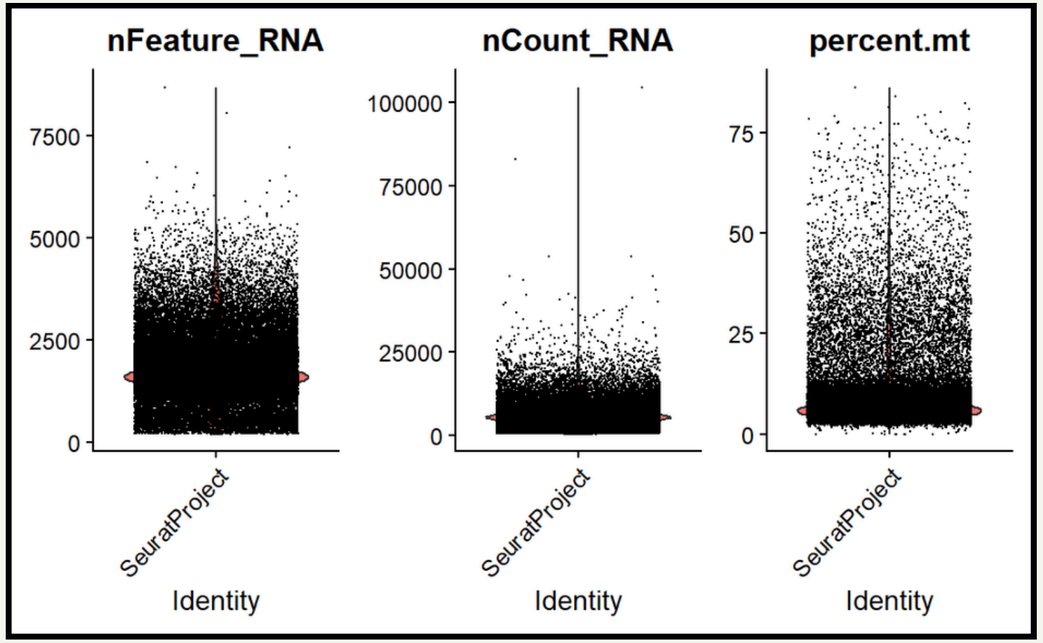

Monocyte cluster divided in respect to patient samples



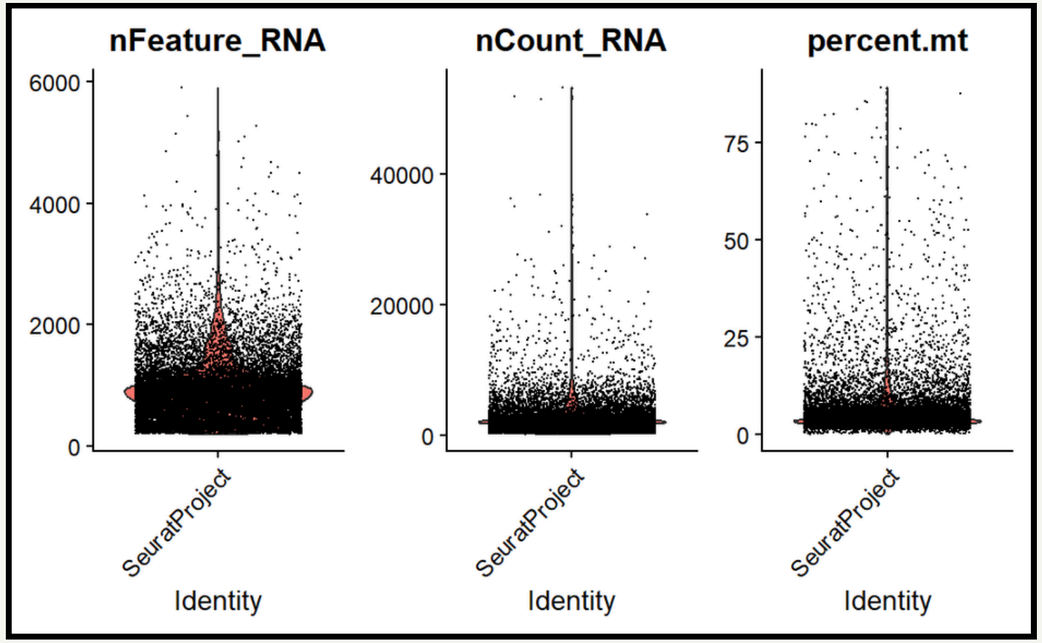

Monocyte cluster divided in respect to disease stage

# Results: clustering with different parameters

The mitochondrial threshold was adjusted to 25% for control samples and 15% for experimental samples.




FeatureScatter plots of chosen features
in the control samples

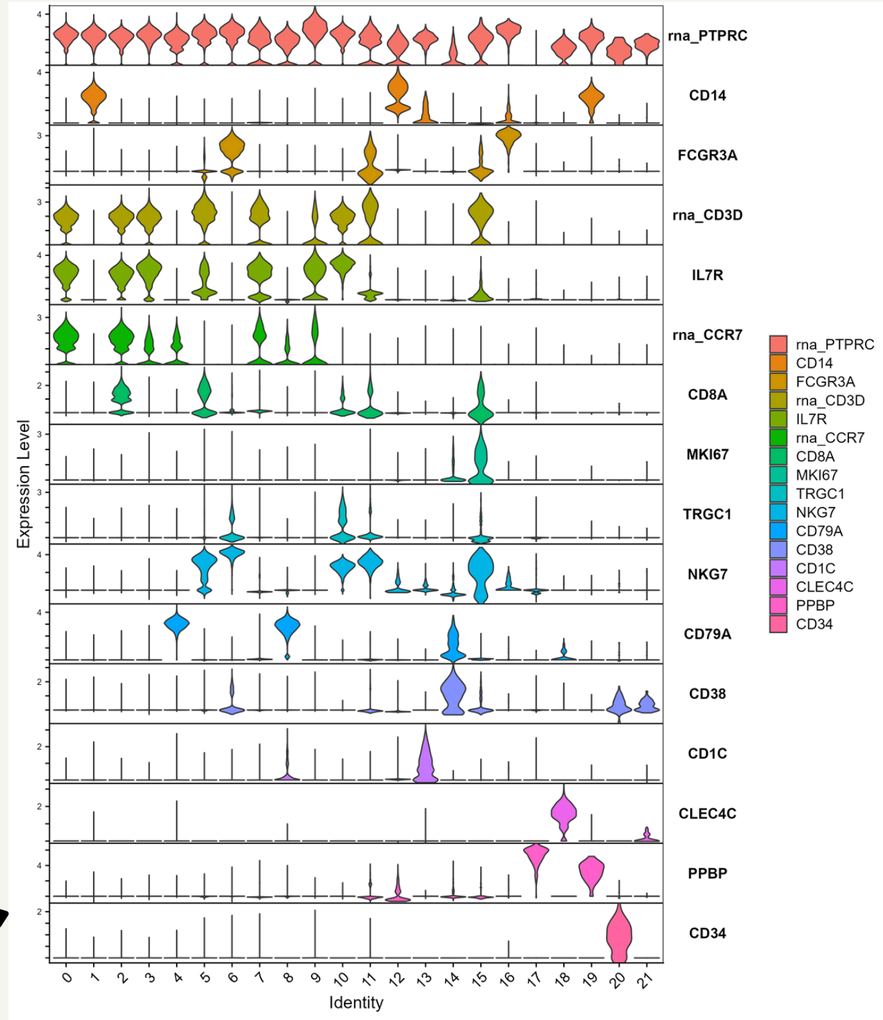



FeatureScatter plots of chosen features
in the experimental samples

# Results: clustering with different parameters

The mitochondrial threshold was adjusted to 25% for control samples and 15% for experimental samples.

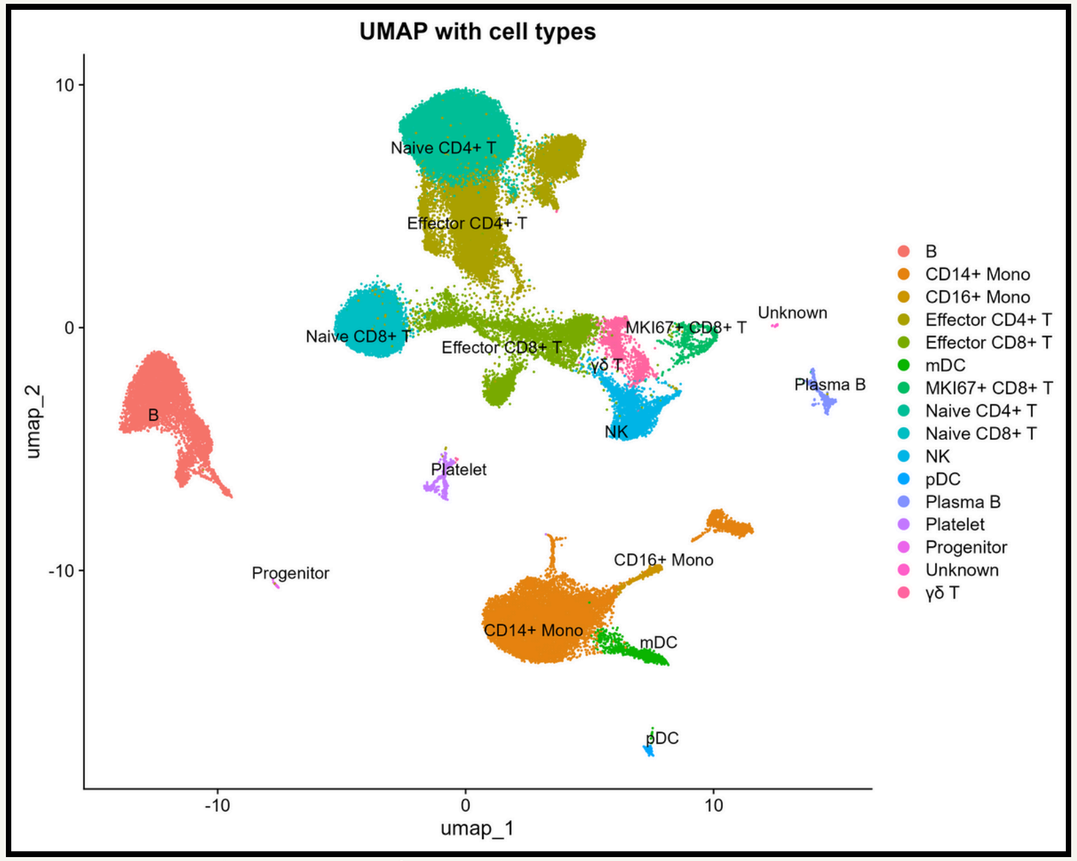
- 22 clusters were built
- again, the violing plots were build and **arduous** assignment of cells subtypes was conducted



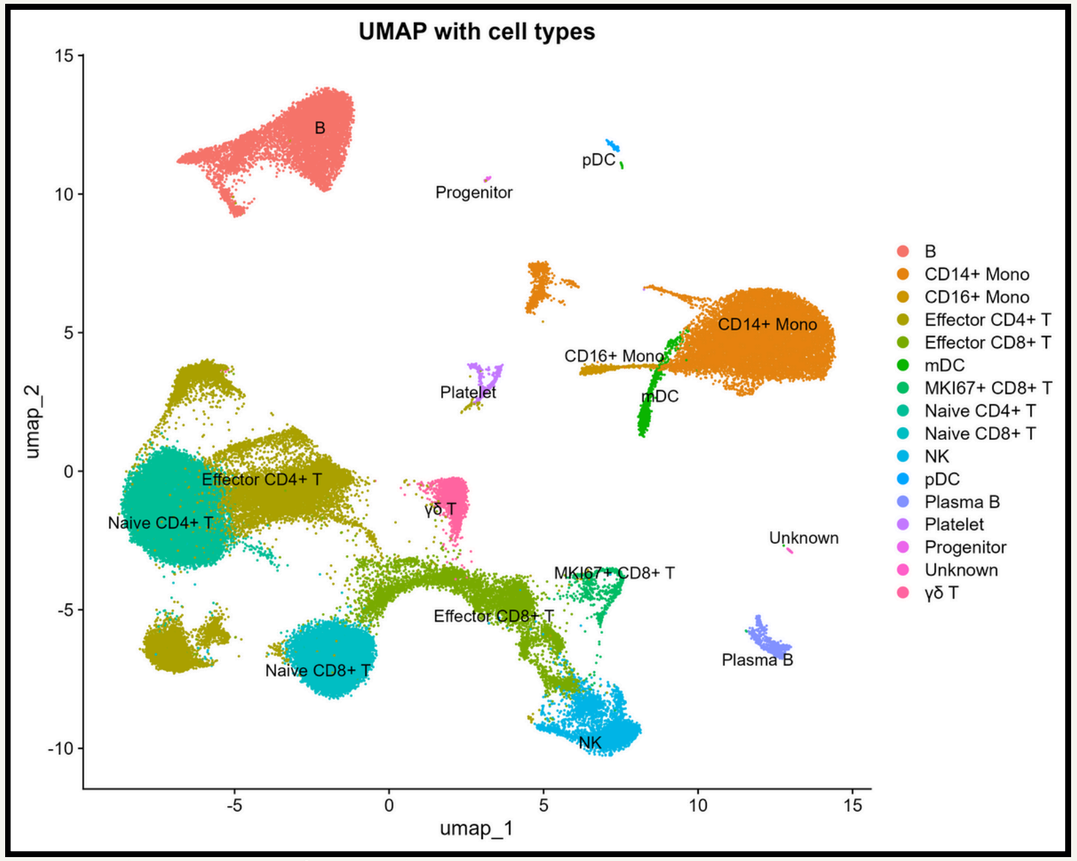

Violin plots of marker expression in my analysis for the new mitochondrial cutoff

# Results: clustering with different parameters

The mitochondrial threshold was adjusted to 25% for control samples and 15% for experimental samples.



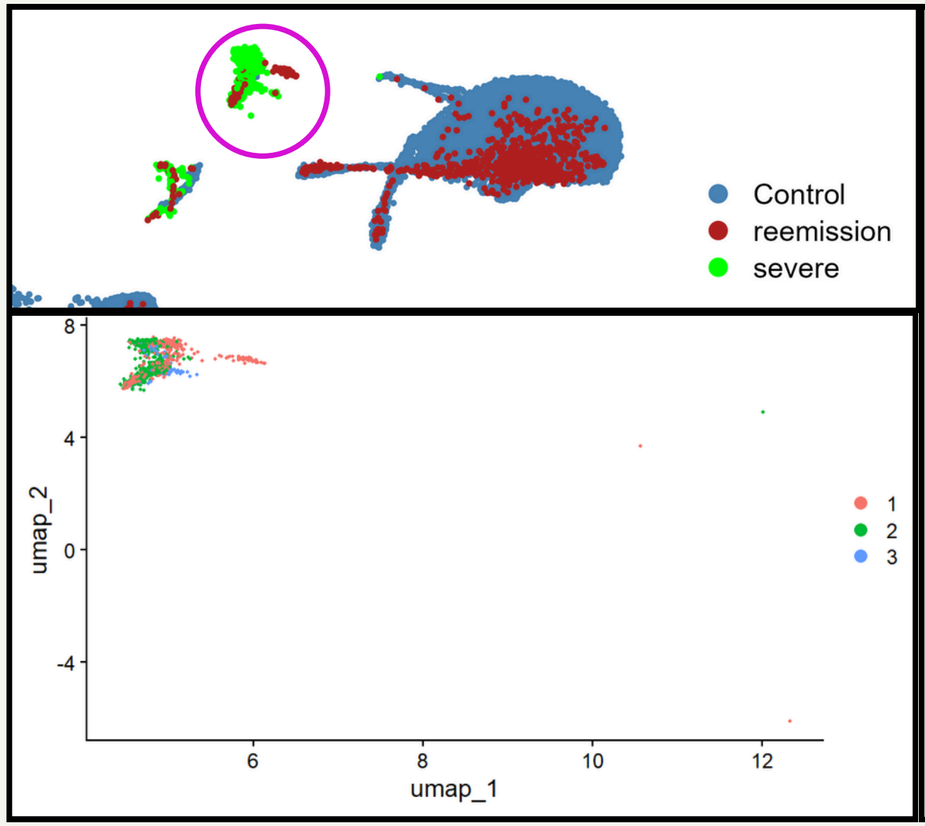

Assignment of cell subtypes to clusters based on the analysis using published values



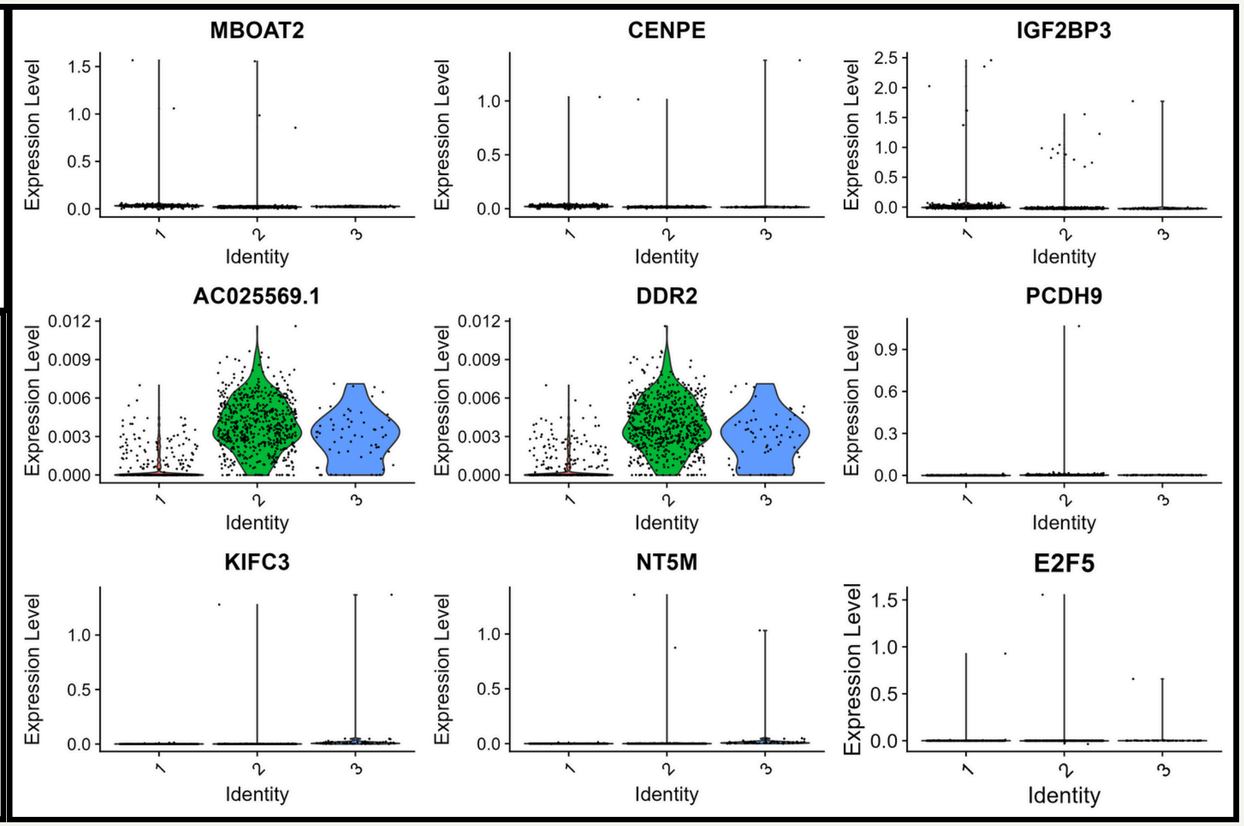

Assignment of cell subtypes to clusters based on my analysis

# Results: clustering with different parameters

The mitochondrial threshold was adjusted to 25% for control samples and 15% for experimental samples.




Top: the monocyte cluster
Bottom: Result of K-means clustering of the monocyste cluster




ViolinPlots of 9 most differentially expressed markers in each of 3 monocyte subclusters.

# To do

Remaining tasks

Further analysis of this monocyte subcluster and comparison to other publicly available datasets.

# Literature

Guo, C., Li, B., Ma, H. et al. Single-cell analysis of two severe COVID-19 patients reveals a monocyte-associated and tocilizumab-responding cytokine storm. Nat Commun 11, 3924 (2020). https://doi.org/10.1038/s41467-020-17834-w