CNS Summer Students 2024

Intermediate Project Results

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Million-level cell embedding Visualization

Start from visualization of two Lung cell datasets

hubmap id
HBM948.GXMD.986
HBM975.WQQQ.853

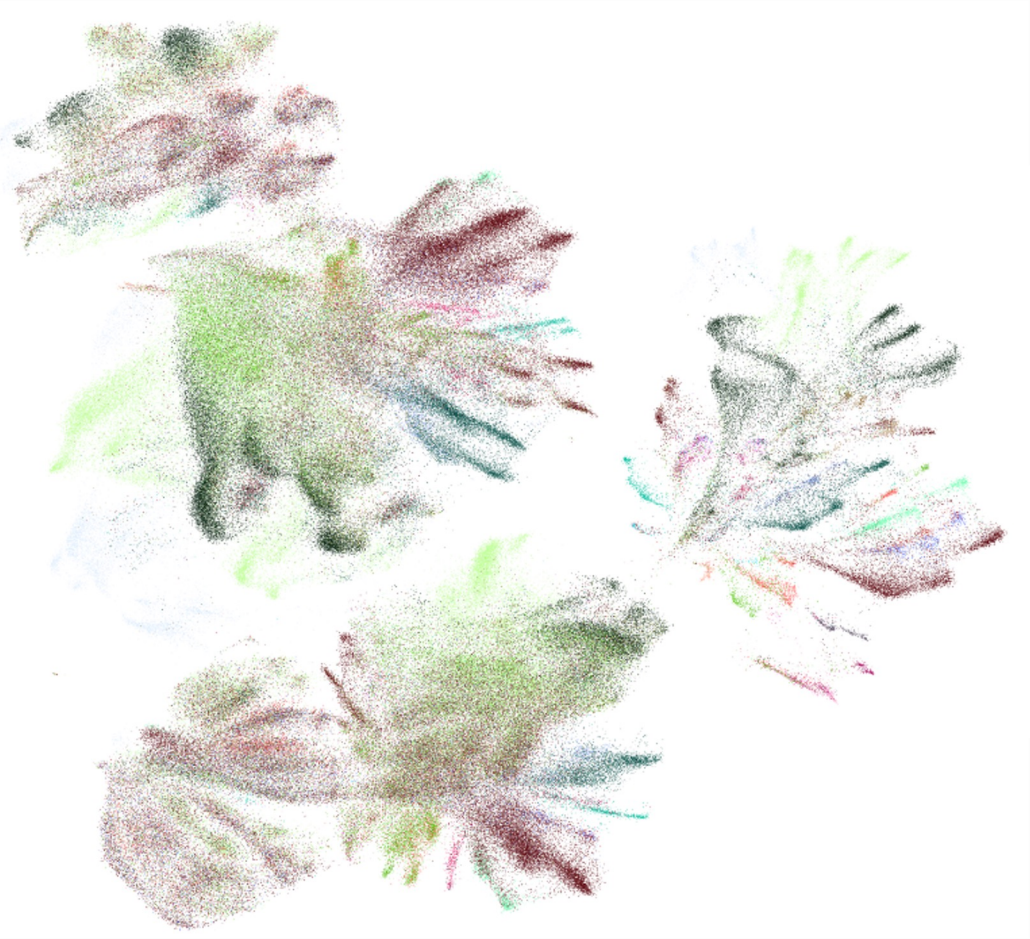
Cell number: 202k
Embedding space: 201905 × 60286

Special settings:
1. Utilize the correct matrix layer:

‘spliced_unspliced_sum’

2. Adopt two steps of normalization before conducting the UMAP function.

```
sc.pp.normalize_total(adata)
sc.pp.log1p(adata)
```



Same normalization settings for ALL 10 lung datasets

cell type number is: 48

CL_label	
capillary endothelial cell	119435
type I pneumocyte	94120
type II pneumocyte	51142
effector memory CD8-positive, alpha-beta T cell	42042
alveolar type 1 fibroblast cell	25102
multi-ciliated epithelial cell:non-nasal	24346
CD4-positive helper T cell	20184
ionocyte	14056
endothelial cell of venule	11042
endothelial cell of artery	10208
lung pericyte	6133
alveolar capillary type 2 endothelial cell	4925
alveolar type 2 fibroblast cell	4324
respiratory basal cell:resting	3358
smooth muscle cell	3066
endothelial cell of lymphatic vessel:mature	2203
endothelial cell of venule:pulmonary	2065
monocyte	1983
CD1c-positive myeloid dendritic cell	1877
B cell	1624

Million-level cell embedding Visualization

Alternative Visualization Published work based on PCA + t-SNE

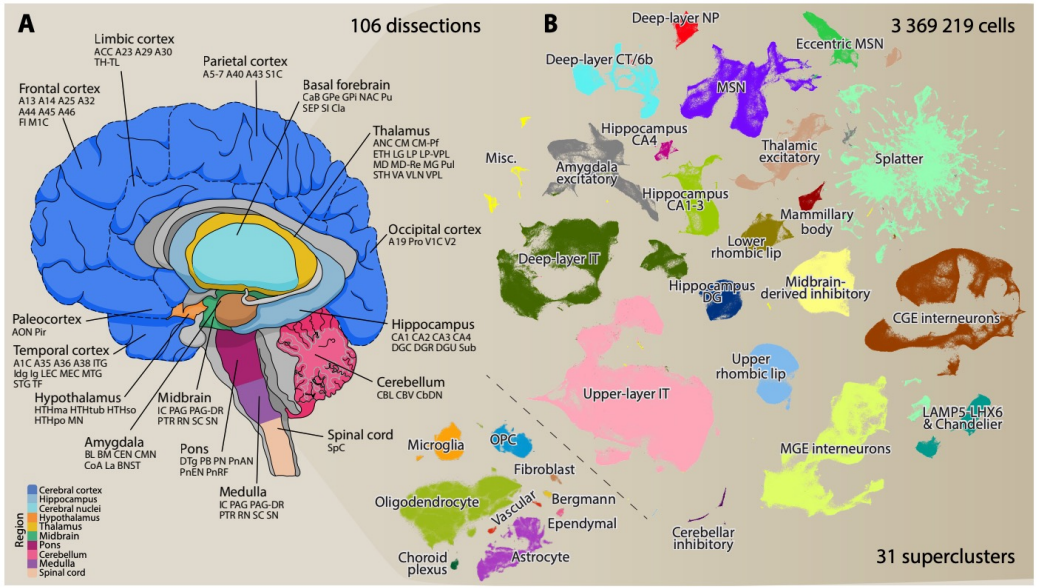
Transcriptomic diversity of cell types across the adult human brain

Kimberly Siletti, Rebecca Hodge, Alejandro Mossi Albiach, Lijuan Hu, Ka Wai Lee, Peter Lönnerberg, Trygve Bakken, Song-Lin Ding, Michael Clark, Tamara Caspe
Julie Nyhus, Herman Tung, Anna Marie Yanny, Ernest Arenas, Ed

doi: https://doi.org/10.1101/2022.10.12.511898

Now published in *Science* doi: 10.1126/science.add7046

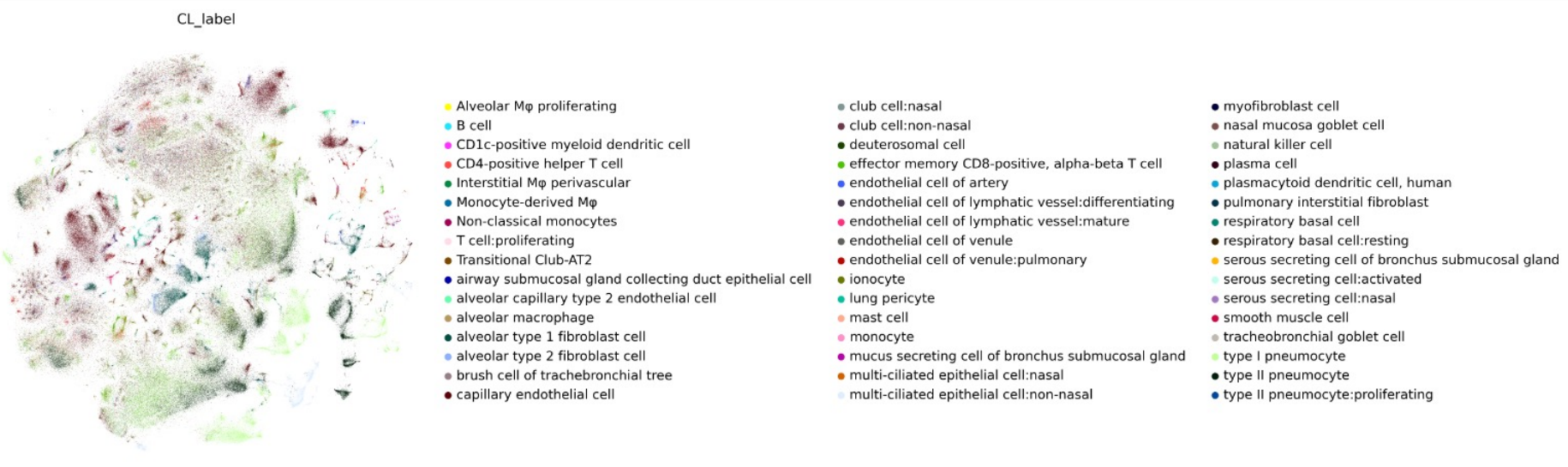
Open-sourced github repository, code for this figure: https://github.com/linnarsson-lab/adult-human-brain/blob/main/notebooks/Preprint/Figure1.ipynb

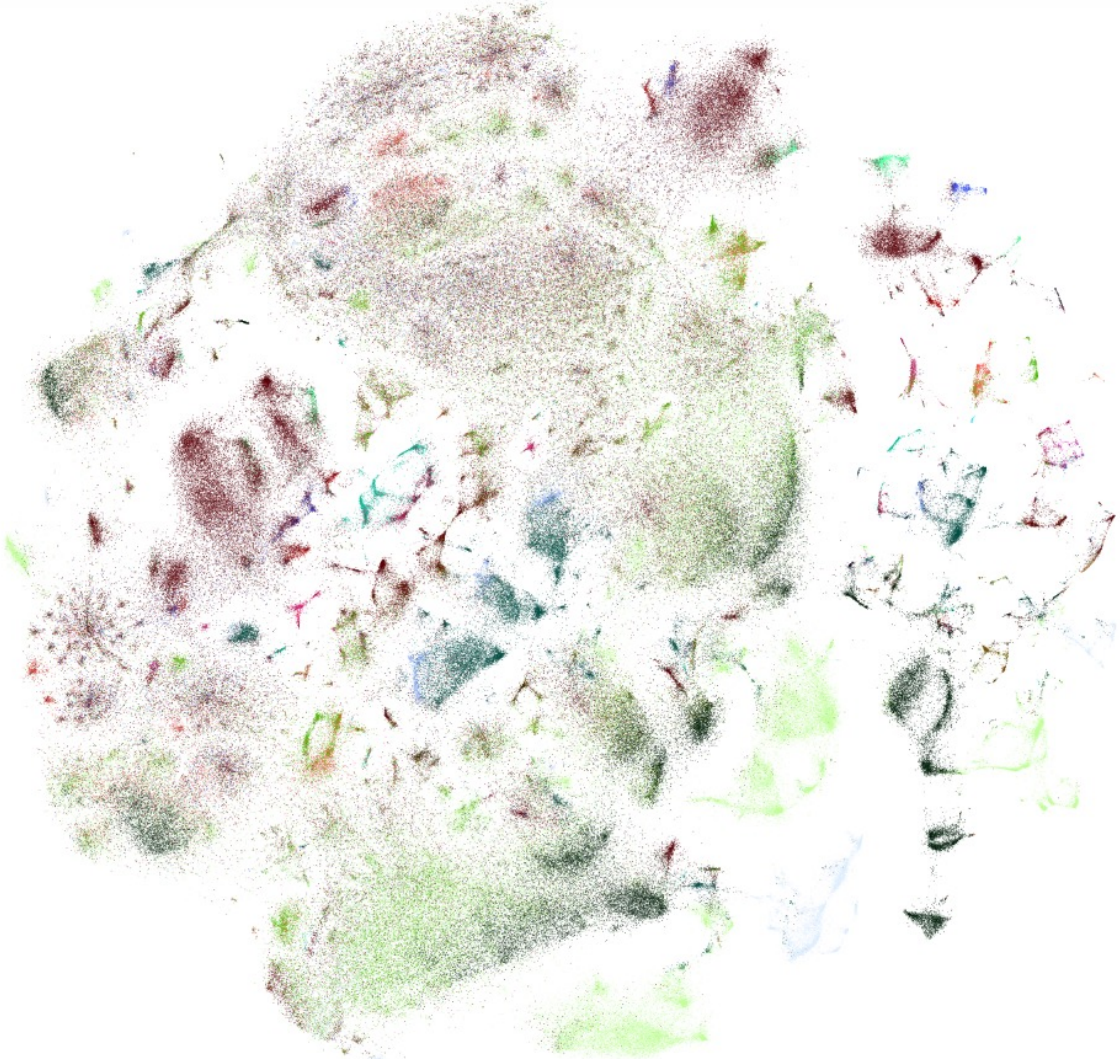


Million-level cell embedding Visualization

Alternative Visualization Published work based on PCA + t-SNE

We firstly adopt PCA for 200 dimensions, then use the t-SNE algorithm for final dimension reduction. We find the result is good, but the clusters are still not so perfectly separated.





453k cell embedding vis:

- PCA, dimension to 200
- T-SNE, dimension to 2 (scanpy default T-SNE)
- Two-step normalizaiton

Million-level cell embedding Visualization

Alternative Visualization Published work based on PCA + t-SNE

We then try the openTSNE (paralleled version) to obtain more t-SNE algorithm parameter settings.

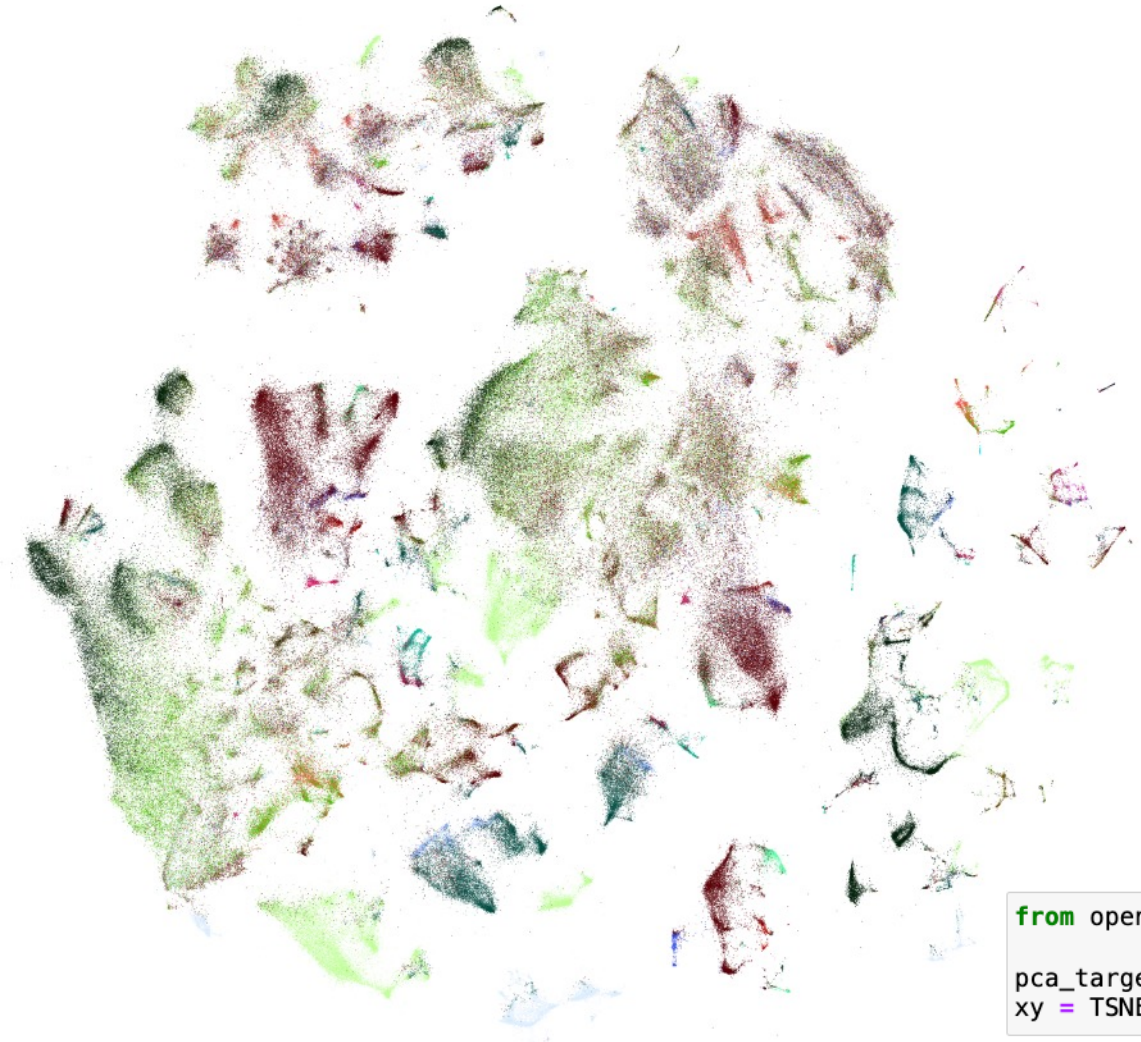
```
sc.tl.pca(adata_concat, n_comps=200)
```

```
from openTSNE import TSNE

pca_target = adata_concat.obsm['X_pca']
xy = TSNE(perplexity=20, exaggeration=1.5).fit(pca_target)
```

```
from openTSNE import TSNE

pca_target = adata_concat.obsm['X_pca']
xy = TSNE(perplexity=20, exaggeration=2.8).fit(pca_target)
```



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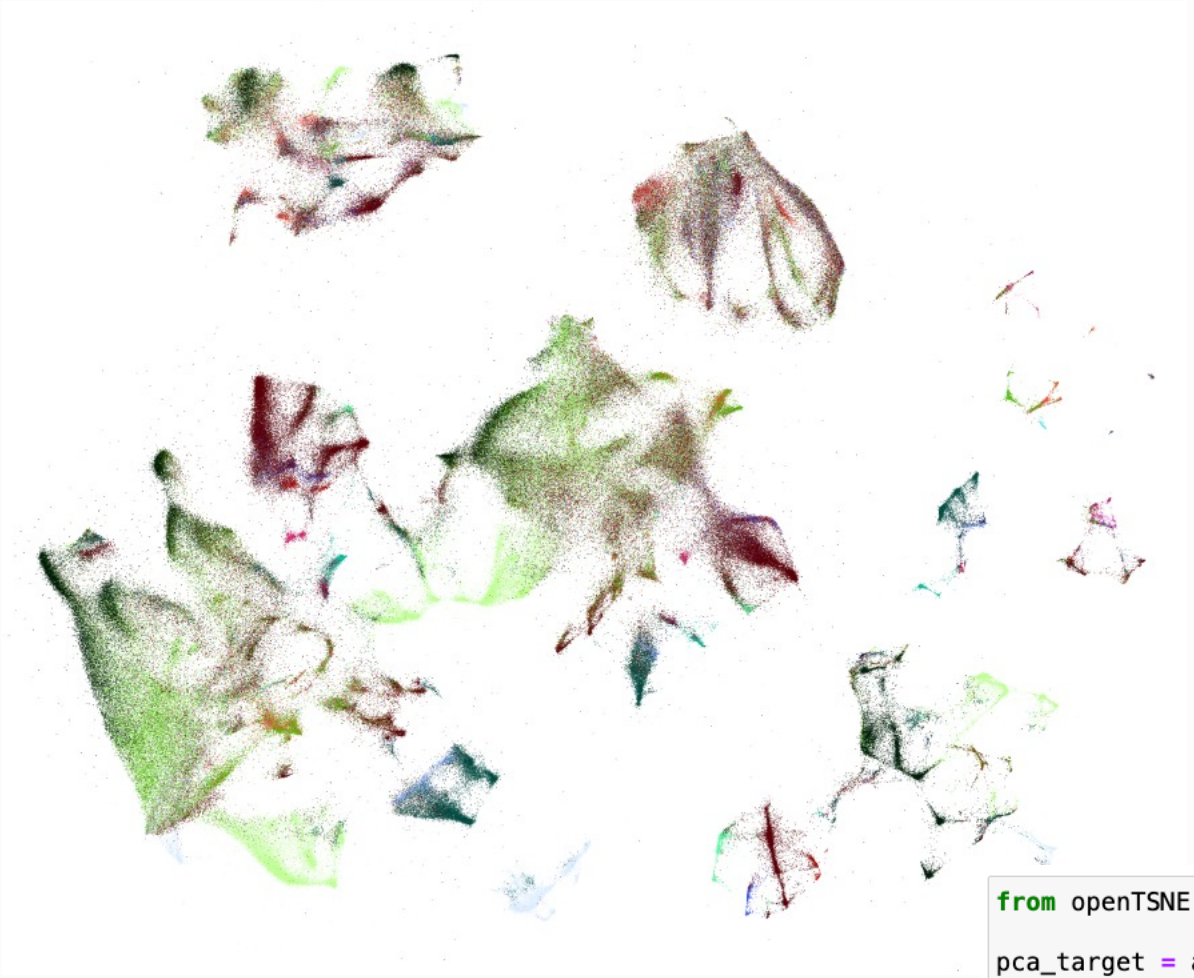

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Million-level cell embedding Visualization

Alternative Visualization Published work based on PCA + t-SNE

We then try the openTSNE (paralleled version) to obtain more t-SNE algorithm parameter settings.

```
sc.tl.tsne(adata_concat, use_rep='X_pca')
```

```
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pca_target = adata_concat.obsm['X_pca']
xy = TSNE(perplexity=20, exaggeration=1.5).fit(pca_target)
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As the ***exaggeration*** rate is higher, the clusters are more clearly separated in the visualization.

Million-level cell embedding Visualization

Alternative Visualization Published work based on PCA + t-SNE

To avoid the bias introduced by PCA re-processing dimension settings, we test 1000 dimension setting. Theoretically, the higher dimension number chosen should reflect better original embedding information.

```
sc.tl.pca(adata_concat, n_comps=1000)
```

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from openTSNE import TSNE

pca_target = adata_concat.obsm['X_pca']
xy = TSNE(perplexity=20, exaggeration=1.5).fit(pca_target)
```

```
from openTSNE import TSNE

pca_target = adata_concat.obsm['X_pca']
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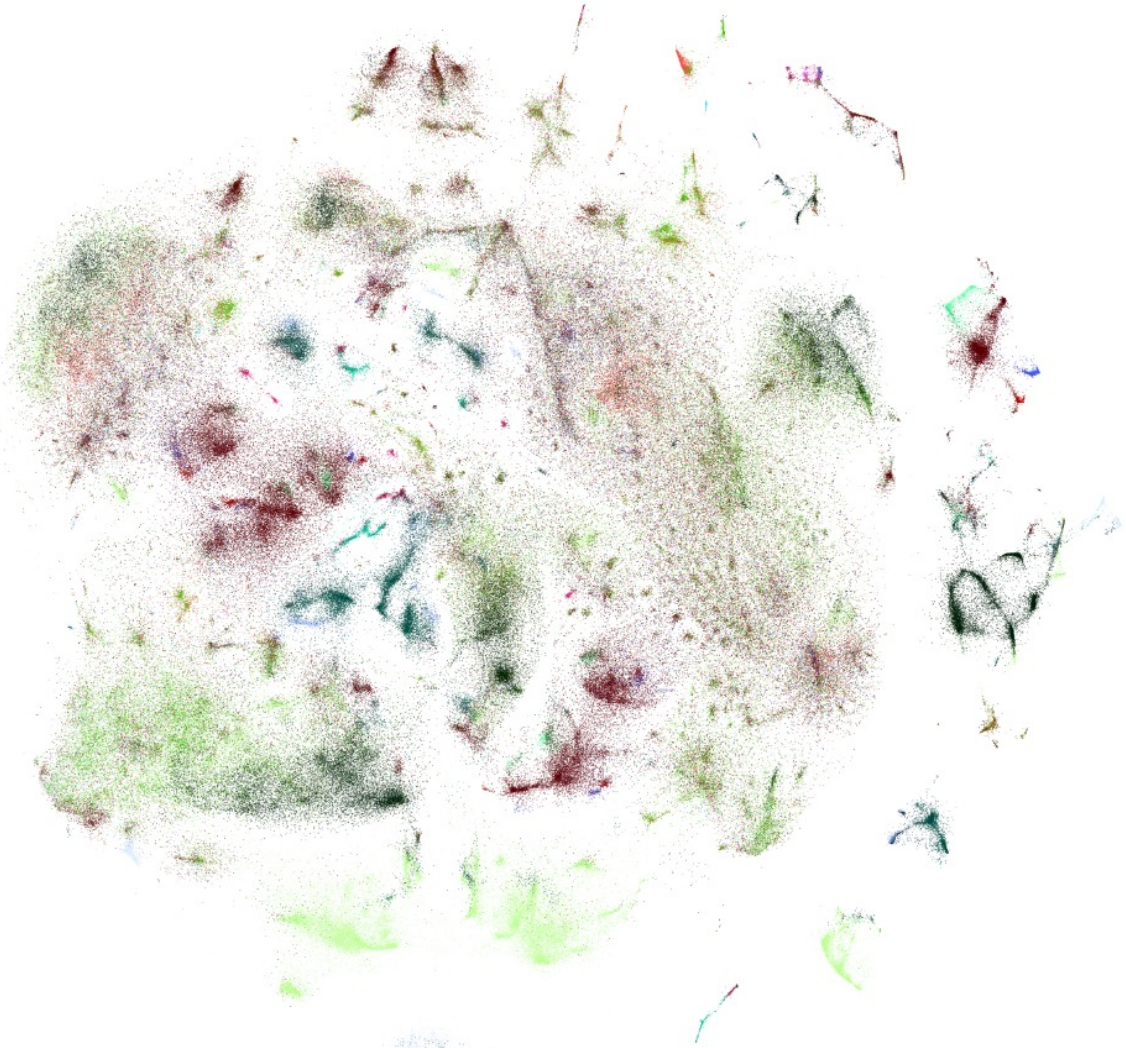

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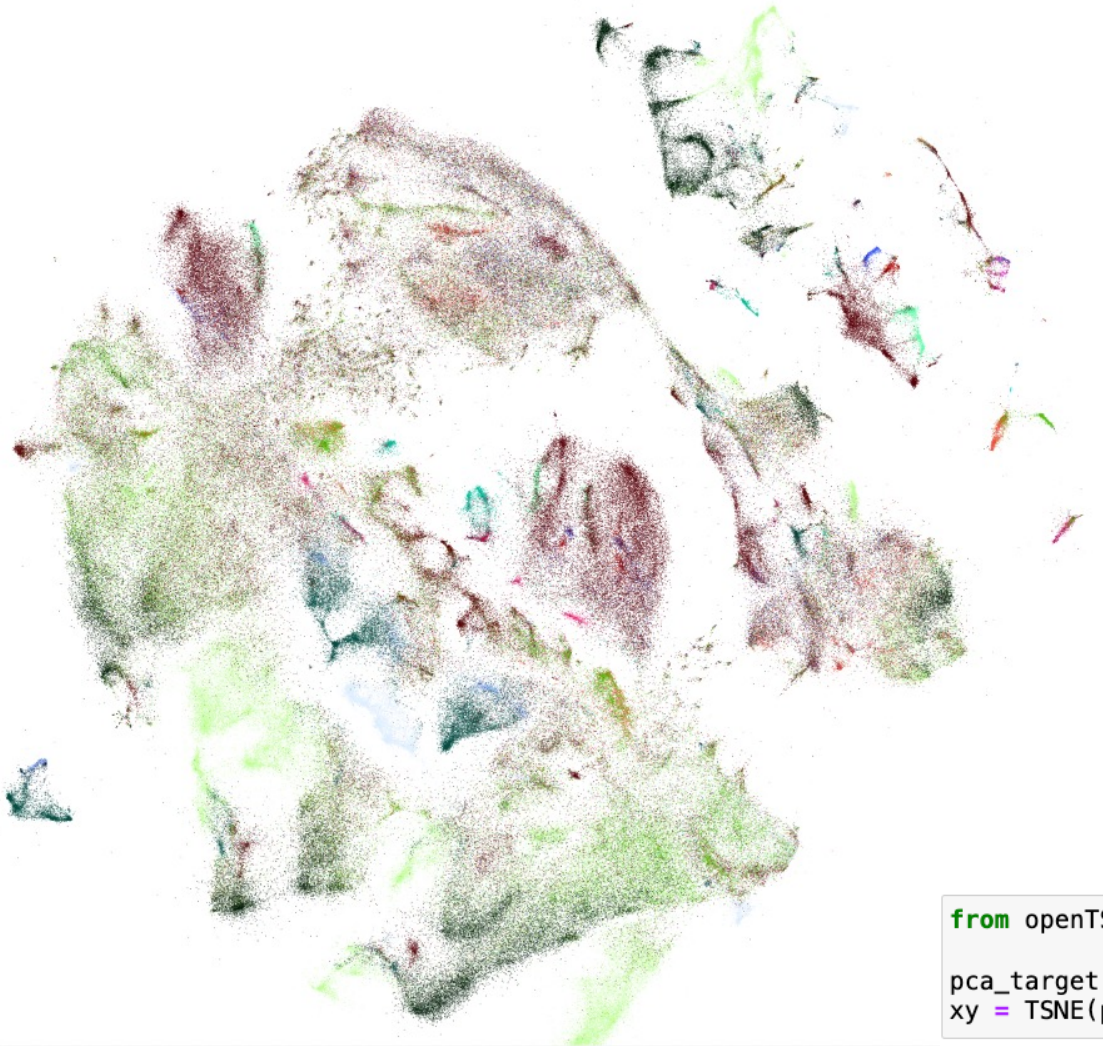

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Million-level cell embedding Visualization

Alternative Visualization Published work based on PCA + t-SNE

For 1000 PCA setting, to better understand the most prevalent cell types, we filtered out the cell categories that is less than 1k in the whole embedding. *453k → 448k*

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```

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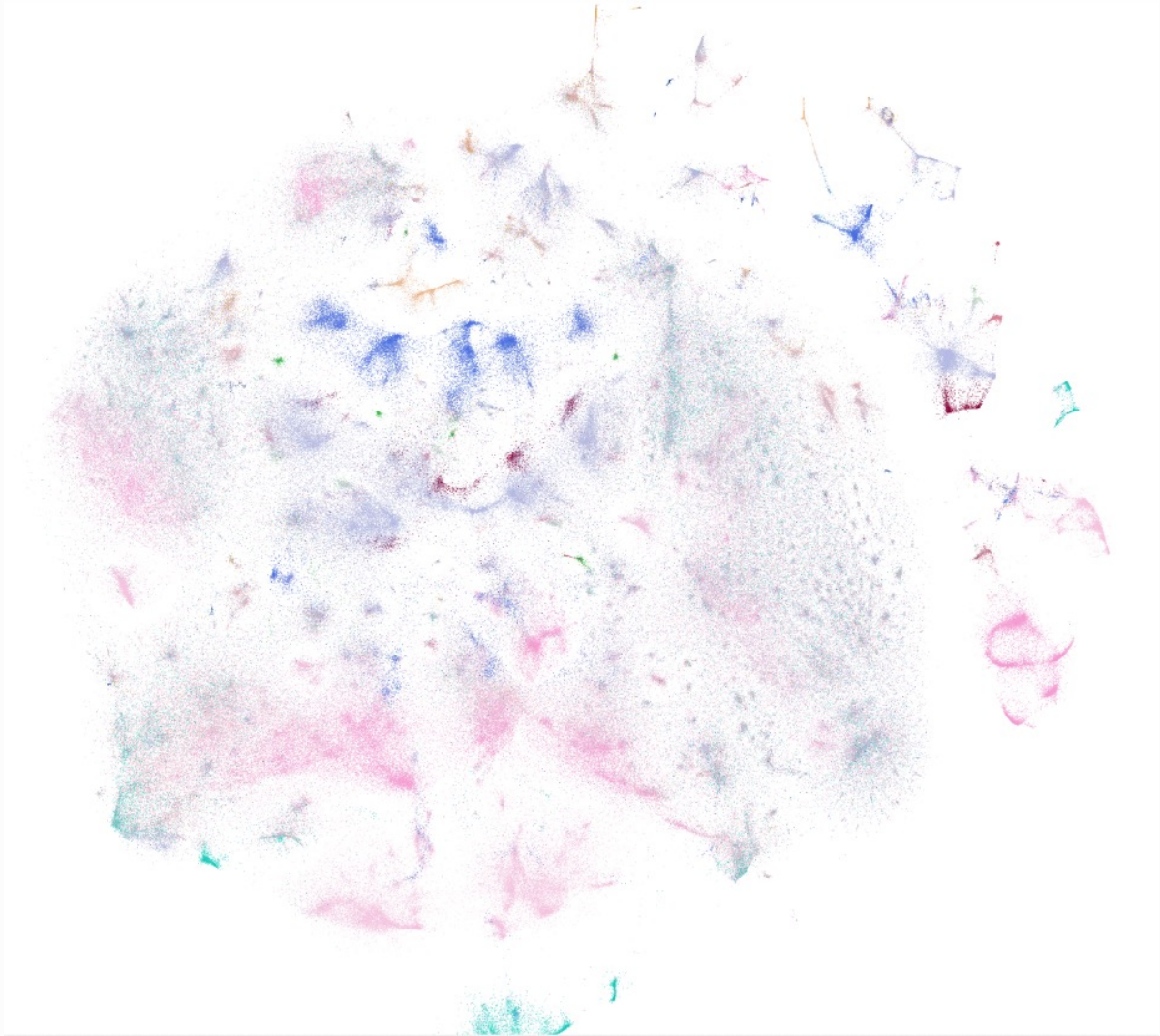

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- PCA, dimension to **1000**
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- Two-step normalizaiton
- Filtered cells that less than 1k of its type

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