single-cell Panoramic View clustering (PanoView) Manual

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1. Introduction

2. Installation

PanoView is a python module that uses other common python libraries such as *numpy*, *scipy*, *pandas*, *scikit-learn*, etc. Prior installing *PanoView* from Github repository, please make sure that *Git* is probably installed or go to https://git-scm.com/ for the installation of *Git*.

To install PanoView at your local computer, open your command prompt and type the following

pip install git+https://github.com/mhu10/scPanoView.git#egg=scPanoView

It will install all the required python libraries for executing *PanoView*. To test the PanoView, open the python interpreter or your preferred IDE (Spyder, PyCharm, Jupyter, etc.) and type the following

from PanoramicView import scPanoView

There should not be any error message popping out.

3. Tutorial

- 3.1 Pollen data
- 3.1.1 Initialization and Input expression matrix

The format of the expression matrix should be comma-separated values (csv). The rows are the genes and the columns are the cells. For demonstration, we use expression data (i.e. "PollenRaw.csv") from [Pollen, etc, Nature Biotechnology 2014] as the input matrix. Please make sure that "PollenRaw.csv" is in your python working directory.

First, import PanoView module by from PanoramicView import scPanoView. Second, chose a job name ("Pollen" in this tutorial) and initialize PanoView by inputting the filename of the expression matrix, "PollenRaw". (Note: you need to do the initialization whenever starting a new job)

```
from PanoramicView import scPanoView
[1] Pollen = scPanoView.PanoView( "PollenRaw" )
```

You may check the raw expression values of the first three genes/cells from the input matrix. The raw expression value is stored at Pollen.raw exp

```
[2] Pollen.raw_exp.iloc[:3,:3]
Gene Symbol
                 Hi 2338 1
                               Hi_2338_2
                                             Hi_2338_3
A1BG
                 9.08
                               0.00
                                             0.00
A1BG-AS1
                 0.00
                               0.00
                                             3.47
A1CF
                 0.00
                               0.05
                                             0.00
```

3.1.2 Generate clusters

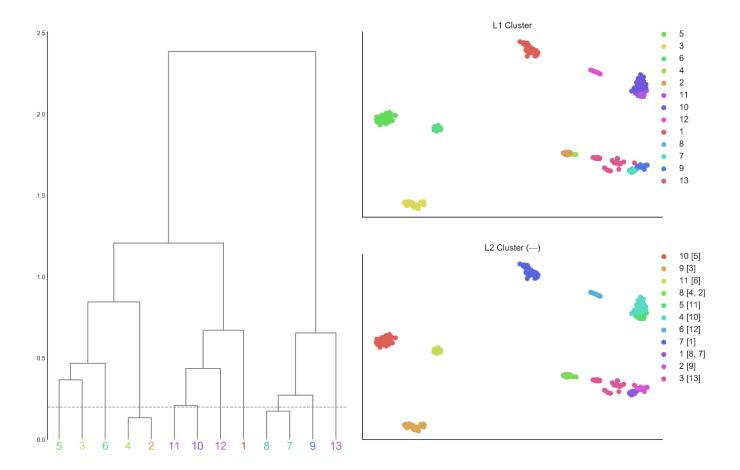
Use Pollen.RunPanoView() to identify single-cell clusters. It will show the progress of algorithm in percentage and display "RunPanoViw-Done" once the searching is finished.

```
[3] Pollen.RunPanoView()
14%
20%
33%
:
74%
86%
RunPanoView-Done
```

3.1.3 Output result

Use Pollen.OutputPanoView() to output the result of *PanoView* at your working directory. The result includes a table ("Cell_Membership.csv") storing clustering membership of cells and a figure ("PanoView_output.png") that consists of one cluster hierarchy and two TSNE plots.

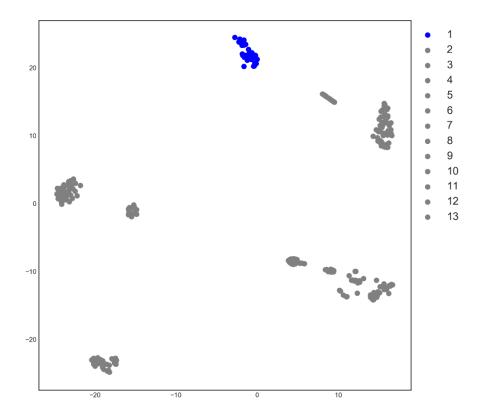
[4] Pollen.OutputPanoView()
Cell_Membership.csv
PanoView_output.png



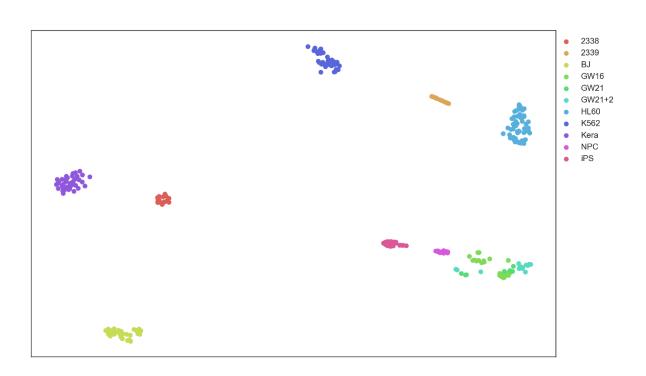
As shown in the figure, *PanoView* identified 13 clusters (Level-1 clusters) and used hierarchical tree to show the similarity of identified clusters. One intuitive parameter in *PanoView* is the height (fclust_dis = 0.2) of the hierarchical tree that would further merge nearby similar clusters. In this tutorial, the default value of 0.2 produced a total of 11 Level-2 clusters that merged cluster#2 and cluster#4 as cluster 8, and cluster#7 and cluster#8 as cluster 1.

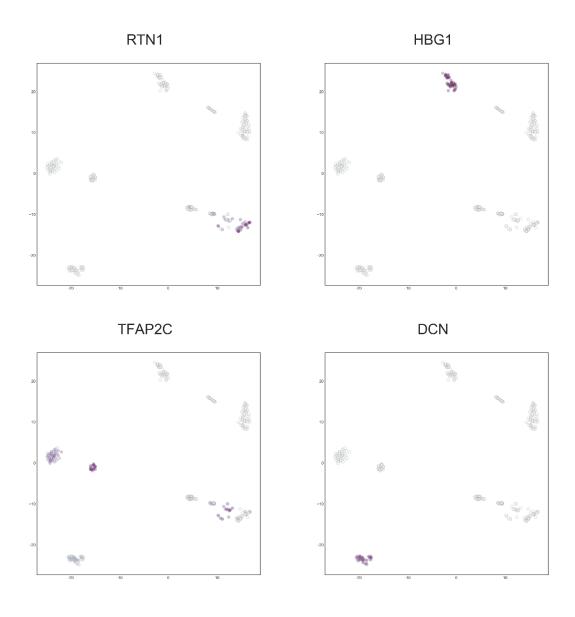
3.1.4 Visualization

[5] Pollen.VisCluster(clevel=1,cnumber=1)



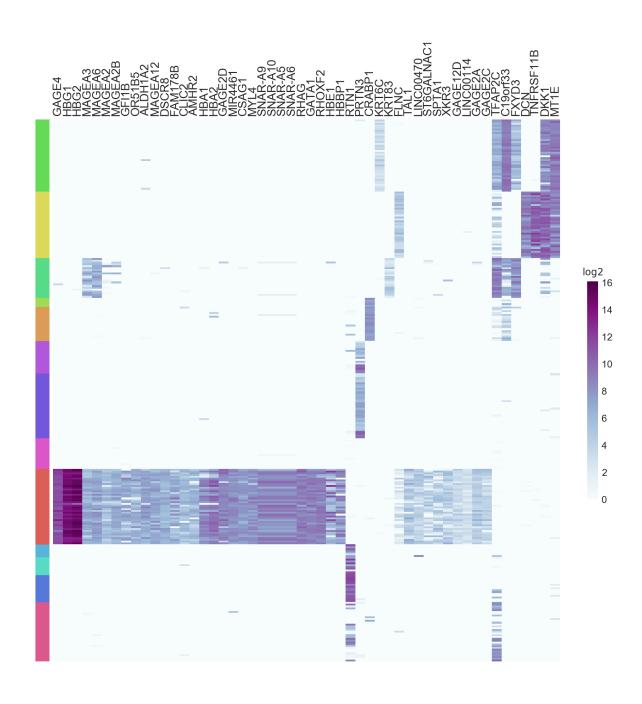
[6] Pollen.VisClusterAnno(annotation)





3.1.5 Variable Genes

[8] Pollen.VisGeneExp(pval=0.05,number=50,fd=2,clevel=1,genelist=['DCN','RTN1','HBG1','TFAP2C'])



4.	Fur	Functions	
	4.1	RunPanoView	
		RunPanoView(GeneLow='default',Zscore='default', Normal=True,Log2=True)	
	4.2	OutputPanoView	
		OutputPanoView(clust_merge = 'default', metric_dis = 'default', fclust_dis = 'default', init = 'default', n_PCs = 'default')	
	4.3	VisCluster	
		VisCluster(clevel,cnumber)	
	4.4	VisClusterAnno	
		VisClusterAnno(annotation)	
	4.5	VisGeneExp	
		VisGeneExp(genes)	
	4.6	RunVGs	
		RunVGs(clevel)	
	4.7	HeatMapVGs	

Heat Map VGs (pval, number, fd, clevel, gene list = 'none')

5. Others