BME-230B Spring 2019 HW 2 Question 2

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- ref
  - scanpy.tl.umap (https://icb-scanpy.readthedocs-hosted.com/en/stable/api/scanpy.tl.umap.html)
  - scanpy.api.pp.neighbors (https://icb-scanpy.readthedocs-hosted.com/en/stable/api/scanpy.api.pp.neighbors.html?highlight=neighbors)
  - scanpy.pl.umap (https://icb-scanpy.readthedocs-hosted.com/en/stable/api/scanpy.pl.umap.html#scanpy.pl.umap)
  - scanpy.tl.louvain (https://icb-scanpy.readthedocs-hosted.com/en/stable/api/scanpy.tl.louvain.html#scanpy.tl.louvain)
  - GSEAPY: Gene Set Enrichment Analysis in Python. pypi.org (https://pypi.org/project/gseapy/)
  - GSEAPY: Gene Set Enrichment Analysis in Python gseapy.readthedocs.io (https://gseapy.readthedocs.io/en/latest/introduction.html)
  - anndata (https://anndata.readthedocs.io/en/latest/anndata.AnnData.html)
    - "uns" stands for unstructured data
    - "obs" are panda data frame observations
    - "obsm key-indexed multi-dimensional observations
  - Hypergeometric_distribution (https://en.wikipedia.org/wiki/Hypergeometric_distribution)
  - Hypergeometric Tests for Gene Lists (http://users.unimi.it/marray/2007/material/day4/Lecture7.pdf)

In [1]:
```
from euclid_knn import KnnG
import gseapy as gp
import matplotlib.pyplot as plt
import numpy as np
import os

import pandas as pd
import scanpy.api as sc
import scanpy
print("scanpy.__version__:{}".format(scanpy.__version__))

import scipy.special
import scipy.stats as stats
```

```
scanpy.__version__:1.4
```

2.b. [5 pts]

Turn in a UMAP plot of the combined dataset as you did in question #1, but this time, color the cells by their Louvain cluster assignments determined for each cell within each batch as a different color in each plot [2 pts: UMAP]. Also report the modularity of the partition you obtained on the combined dataset [3 pts: Reporting Modularity].

In [2]:
```
%%time
anndata = sc.read("PBMC.merged.h5ad")
```

```
CPU times: user 4.2 s, sys: 134 ms, total: 4.33 s
Wall time: 1.53 s
```

In [3]:
```
# run our implementation of nearest neighboors and update anndata
KnnG(anndata, n_neighbors=12, runPCA=True, nPC=50)
```

Out[3]:
```
<euclid_knn.KnnG at 0x7f763c1d4b00>
```

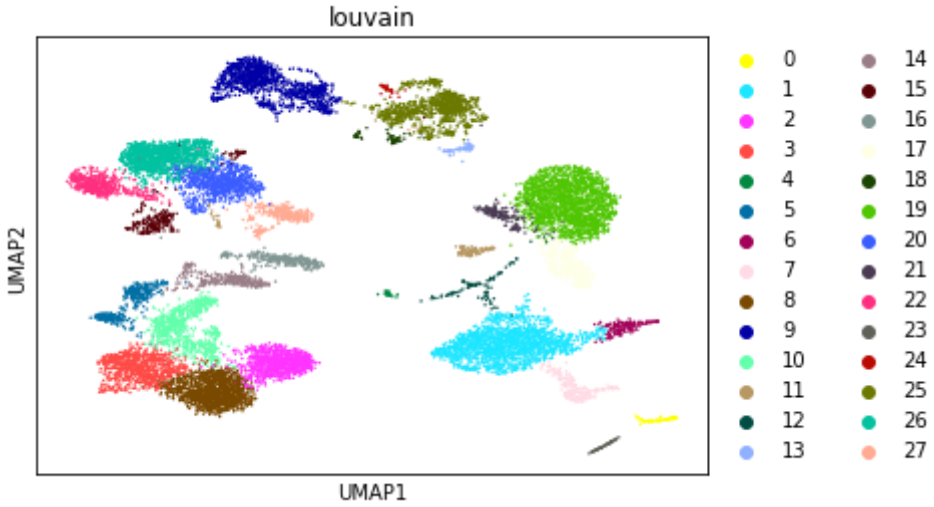
In [4]:
```
%%time
# running Scanpy's version of Louvian
scanpy.tl.louvain(anndata,
                  flavor='igraph',
                  directed=False,
                  use_weights=True)
```

```
CPU times: user 7.09 s, sys: 53.6 ms, total: 7.14 s
Wall time: 1.35 s
```

In [5]:
```
scanpy.tl.umap(anndata)
```

In [6]:
```
plt.figure(figsize=(10,10))
scanpy.pl.umap(anndata, color=["louvain"])
```

```
<Figure size 720x720 with 0 Axes>
```



2.c. [5 pts]

Turn in a table that lists each cluster and its best-matching cell type annotation. The table should contain the cluster number and its best matching cell-type annotation based on the hypergeometric analysis.

In [12]:
```
import hw2q2
```

In [13]:
```
cellCountsByClusterId, cellTypesInClusters = hw2q2.createCountsDict(anndata
```

In [16]:
```
annotations = hw2q2.bestAnnotation(anndata, cellTypesInClusters, cellCounts
annotations.sort_values(by=['clusterId'])
```

Out[16]:

	clusterId	Cell type	p-value
0	0	Plasmacytoid dendritic cell	0.000000e+00
1	1	Monocyte_CD14	0.000000e+00
2	2	CD8 T cell	3.223022e-11
3	3	CD4 T cell	0.000000e+00
4	4	Hematopoietic stem cell	0.000000e+00
5	5	CD8 T cell	1.956746e-11
6	6	Monocyte_FCGR3A	0.000000e+00
7	7	Monocyte_CD14	0.000000e+00
8	8	CD4 T cell	7.790102e-12
9	9	B cell	0.000000e+00
10	10	CD8 T cell	2.847245e-11
11	11	Monocyte_CD14	3.811396e-12
12	12	Megakaryocyte	1.195422e-11
13	13	B cell	0.000000e+00
14	14	NK cell	0.000000e+00
15	15	CD8 T cell	0.000000e+00
16	16	NK cell	0.000000e+00
17	17	Monocyte_FCGR3A	0.000000e+00
18	18	B cell	0.000000e+00
19	19	Monocyte_CD14	0.000000e+00
20	20	CD4 T cell	1.010458e-11
21	21	Monocyte_CD14	0.000000e+00
22	22	CD8 T cell	0.000000e+00
23	23	Plasmacytoid dendritic cell	0.000000e+00
24	24	B cell	0.000000e+00
25	25	B cell	0.000000e+00
26	26	CD4 T cell	1.193878e-11
27	27	CD8 T cell	5.398015e-12

2.d. [5 pts]

Turn in a list of top 5 pathways for each cluster in each dataset. You should use the gene expression signature of each cluster to find an associated pathway. A gene signature for a cluster represents the gene expression levels for a characteristic cell that is a member of the cluster. Use the centroid $\boldsymbol{\mu}$ i of the i th cluster as the signature. Compute the centroids for each cluster in each dataset. You will next derive a gene-signature based annotation for each cluster using these centroids. Use a list of Gene Ontology Biological Process categories (provided in the Resources section at the top of this homework) and your signatures to perform an all-against-all Gene Set Enrichment Analysis (GSEA). Turn in a table that lists the top 5 pathways for each cluster

In [18]:
```
clusterSigs = hw2q2.calculateClusterSignatures(anndata)
```

In [19]:
```
%%time
pathways = hw2q2.rankPathWays(anndata, clusterSigs, topN=5)
```

```
CPU times: user 2min 46s, sys: 8.38 s, total: 2min 54s
Wall time: 9min 33s
```

In [20]:
```
# https://stackoverflow.com/a/35693013/4586180
# display data frame with out index column
from IPython.display import display, HTML
display(HTML(pathways.to_html(index=False)))
```

Term	nes	cluster id
cellular protein metabolic process (GO:0044267)	1.333285	0
Fc-epsilon receptor signaling pathway (GO:0038...	1.323810	0
antigen processing and presentation of exogeno...	1.325697	0
antigen processing and presentation of peptide...	1.352975	0
positive regulation of phosphorylation (GO:004...	1.361454	0
neutrophil degranulation (GO:0043312)	2.059123	1
neutrophil activation involved in immune respo...	2.078532	1
neutrophil mediated immunity (GO:0002446)	2.107963	1
cellular protein metabolic process (GO:0044267)	1.827333	1
granulocyte chemotaxis (GO:0071621)	1.694330	1
T cell activation (GO:0042110)	2.452248	2
complement activation, classical pathway (GO:0...	1.699152	2
transmembrane receptor protein tyrosine kinase...	1.610634	2
enzyme linked receptor protein signaling pathw...	1.477404	2
response to cytokine (GO:0034097)	1.284005	2
regulation of protein kinase B signaling (GO:0...	1.595247	3
T cell receptor signaling pathway (GO:0050852)	1.599044	3
tumor necrosis factor-mediated signaling pathw...	1.802821	3
positive regulation of GTPase activity (GO:004...	1.579765	3
positive regulation of protein kinase B signal...	1.567524	3
positive regulation of gene expression (GO:001...	1.605392	4
positive regulation of nucleic acid-templated ...	1.758239	4
positive regulation of cellular biosynthetic p...	1.674370	4
negative regulation of nucleic acid-templated ...	1.671713	4
regulation of transcription, DNA-templated (GO...	1.664383	4
protein oligomerization (GO:0051259)	1.430223	5
cellular defense response (GO:0006968)	1.400160	5
regulation of immune response (GO:0050776)	1.720955	5
positive regulation of hydrolase activity (GO:...	1.436126	5
inflammatory response (GO:0006954)	1.403699	5
neutrophil degranulation (GO:0043312)	1.616853	6
neutrophil mediated immunity (GO:0002446)	1.622177	6
neutrophil activation involved in immune respo...	1.653849	6

Term	nes	cluster id
antigen processing and presentation of peptide...	1.571275	6
antigen processing and presentation of exogeno...	1.577903	6
antigen processing and presentation of exogeno...	1.855222	7
antigen processing and presentation of peptide...	1.814602	7
cellular response to interferon-gamma (GO:0071...	1.794303	7
interferon-gamma-mediated signaling pathway (G...	1.770483	7
antigen processing and presentation of exogeno...	1.757600	7
T cell activation (GO:0042110)	2.312814	8
complement activation, classical pathway (GO:0...	1.697793	8
positive regulation of protein kinase B signal...	1.363084	8
regulation of acute inflammatory response (GO:...	1.272369	8
humoral immune response mediated by circulatin...	1.323808	8
B cell receptor signaling pathway (GO:0050853)	2.208003	9
positive regulation of B cell activation (GO:0...	2.012516	9
positive regulation of lymphocyte activation (...	2.003641	9
antigen receptor-mediated signaling pathway (G...	2.173027	9
humoral immune response mediated by circulatin...	2.036609	9
regulation of GTPase activity (GO:0043087)	1.827392	10
positive regulation of GTPase activity (GO:004...	1.794914	10
positive regulation of hydrolase activity (GO:...	1.781682	10
T cell activation (GO:0042110)	1.671555	10
regulation of immune response (GO:0050776)	1.576390	10
antigen processing and presentation of exogeno...	1.784130	11
antigen processing and presentation of exogeno...	1.792342	11
T cell receptor signaling pathway (GO:0050852)	1.593702	11
interferon-gamma-mediated signaling pathway (G...	1.539432	11
antigen processing and presentation of peptide...	1.751178	11
response to molecule of bacterial origin (GO:0...	1.388828	12
positive regulation of leukocyte chemotaxis (G...	1.406809	12
platelet degranulation (GO:0002576)	1.433550	12
muscle contraction (GO:0006936)	1.454998	12
regulated exocytosis (GO:0045055)	1.519884	12
regulation of immune effector process (GO:0002...	1.737337	13
interferon-gamma-mediated signaling pathway (G...	1.728635	13
regulation of complement activation (GO:0030449)	1.712084	13
antigen processing and presentation of exogeno...	1.698735	13

Term	nes	cluster id
regulation of B cell activation (GO:0050864)	1.699109	13
positive regulation of hydrolase activity (GO:...	1.609459	14
positive regulation of ERK1 and ERK2 cascade (...	1.568272	14
regulation of GTPase activity (GO:0043087)	1.565476	14
positive regulation of GTPase activity (GO:004...	1.551697	14
regulation of immune response (GO:0050776)	1.640011	14
T cell activation (GO:0042110)	1.695259	15
negative regulation of cytokine production (GO...	1.488782	15
regulation of GTPase activity (GO:0043087)	1.562767	15
positive regulation of GTPase activity (GO:004...	1.595306	15
positive regulation of hydrolase activity (GO:...	1.605712	15
positive regulation of hydrolase activity (GO:...	1.529969	16
positive regulation of GTPase activity (GO:004...	1.494077	16
response to interferon-gamma (GO:0034341)	1.414437	16
regulation of GTPase activity (GO:0043087)	1.538661	16
cellular defense response (GO:0006968)	1.458698	16
neutrophil mediated immunity (GO:0002446)	1.625153	17
antigen processing and presentation of exogeno...	1.510699	17
regulated exocytosis (GO:0045055)	1.472575	17
neutrophil degranulation (GO:0043312)	1.611826	17
neutrophil activation involved in immune respo...	1.624471	17
regulation of protein processing (GO:0070613)	1.774148	18
Fc-gamma receptor signaling pathway involved i...	1.743001	18
Fc-epsilon receptor signaling pathway (GO:0038...	1.743197	18
regulation of complement activation (GO:0030449)	1.753462	18
humoral immune response mediated by circulatin...	1.806558	18
extracellular matrix organization (GO:0030198)	1.516295	19
positive regulation of NF-kappaB transcription...	1.551196	19
neutrophil degranulation (GO:0043312)	1.796621	19
neutrophil mediated immunity (GO:0002446)	1.819045	19
neutrophil activation involved in immune respo...	1.837040	19
positive regulation of gene expression (GO:001...	1.616762	20
regulation of transcription, DNA-templated (GO...	1.638333	20
regulation of transcription from RNA polymeras...	1.693520	20
regulation of programmed cell death (GO:0043067)	1.712579	20
tumor necrosis factor-mediated signaling pathw...	1.716800	20

Term	nes	cluster id
neutrophil degranulation (GO:0043312)	1.710914	21
neutrophil activation involved in immune respo...	1.706809	21
neutrophil mediated immunity (GO:0002446)	1.696067	21
cellular response to lipopolysaccharide (GO:00...	1.500996	21
regulation of cytokine production (GO:0001817)	1.481354	21
regulation of cellular macromolecule biosynthe...	1.459647	22
T cell activation (GO:0042110)	2.110025	22
regulation of cell cycle (GO:0051726)	1.500324	22
transmembrane receptor protein tyrosine kinase...	1.597967	22
enzyme linked receptor protein signaling pathw...	1.713461	22
interferon-gamma-mediated signaling pathway (G...	1.337556	23
regulation of B cell proliferation (GO:0030888)	1.345112	23
regulation of protein phosphorylation (GO:0001...	1.357005	23
positive regulation of phosphorylation (GO:004...	1.408704	23
Fc receptor signaling pathway (GO:0038093)	1.381194	23
regulation of B cell activation (GO:0050864)	2.035837	24
complement activation, classical pathway (GO:0...	1.948536	24
phagocytosis, engulfment (GO:0006911)	1.922344	24
positive regulation of lymphocyte activation (...	1.886077	24
B cell receptor signaling pathway (GO:0050853)	1.918055	24
regulation of protein activation cascade (GO:2...	1.937766	25
regulation of immune effector process (GO:0002...	1.954491	25
regulation of complement activation (GO:0030449)	1.973642	25
humoral immune response mediated by circulatin...	1.981524	25
regulation of humoral immune response (GO:0002...	2.039666	25
regulation of transcription from RNA polymeras...	1.395068	26
T cell activation (GO:0042110)	1.466689	26
protein complex assembly (GO:0006461)	1.678697	26
regulation of cell cycle (GO:0051726)	1.349217	26
regulation of cellular macromolecule biosynthe...	1.411476	26
regulation of transcription, DNA-templated (GO...	1.473158	27
negative regulation of transcription, DNA-temp...	1.490239	27
positive regulation of nucleic acid-templated ...	1.535971	27
regulation of transcription from RNA polymeras...	1.599694	27
negative regulation of transcription from RNA ...	1.725175	27