immune_markers_peak_results_merged_conditions

March 26, 2021

[1]:
```
pwd
```

[1]:
```
'/data2/mito_lineage/Analysis/peaks_expression/jan21_2021'
```

[2]:
```
config_f = "config.yaml"
```

[3]:
```
from src.utils.parse_config import read_config_file
from os.path import join, dirname
import pandas as pd
from glob import glob
config = read_config_file(config_f)
import seaborn as sns
import numpy as np
import matplotlib.pyplot as plt
config

import mplh.cluster_help as ch


%load_ext autoreload
%autoreload 2
```

```
fig_utils from mplh
here
```

0.0.1 Load:

1. Raw cells barcode w qc info
2. filtered cells barcode list
3. Peak-by-cell sparse matrix
4. Peak gene annotations
5. Graph clustering filtered cells cluster label
6. TF-by-cell sparse count matrix
7. TF labels

[4]:
```
#curr_in = join(config["indir"], config['samples'][0], 'outs')

```

```
curr_in = join(config["indir"], "reanalysis_aggr", "outs")
samples = pd.read_csv(join(config["indir"], "barcodes_conditionInfo.csv"),␣
 ↪header=None)

### Create 1-based cell barcode index

cells = pd.read_csv(join(curr_in,"singlecell.csv"))
cells

good_cells = cells[~(cells["cell_id"]=="None")].copy()
good_cells["ID"] = np.arange(1,len(good_cells)+1)
good_cells

cell_inds = pd.read_csv(join(curr_in, "filtered_peak_bc_matrix","barcodes.
 ↪tsv"),header=None)
cell_inds

## Load peak annotations

peak_annotations = pd.read_csv(join(curr_in,'peak_annotation.tsv'),sep='\t')
peak_annotations["gene"] = peak_annotations["gene"].str.upper()
peak_annotations["Peak"]= peak_annotations.index+1
# peak_annotations


### Load clusters

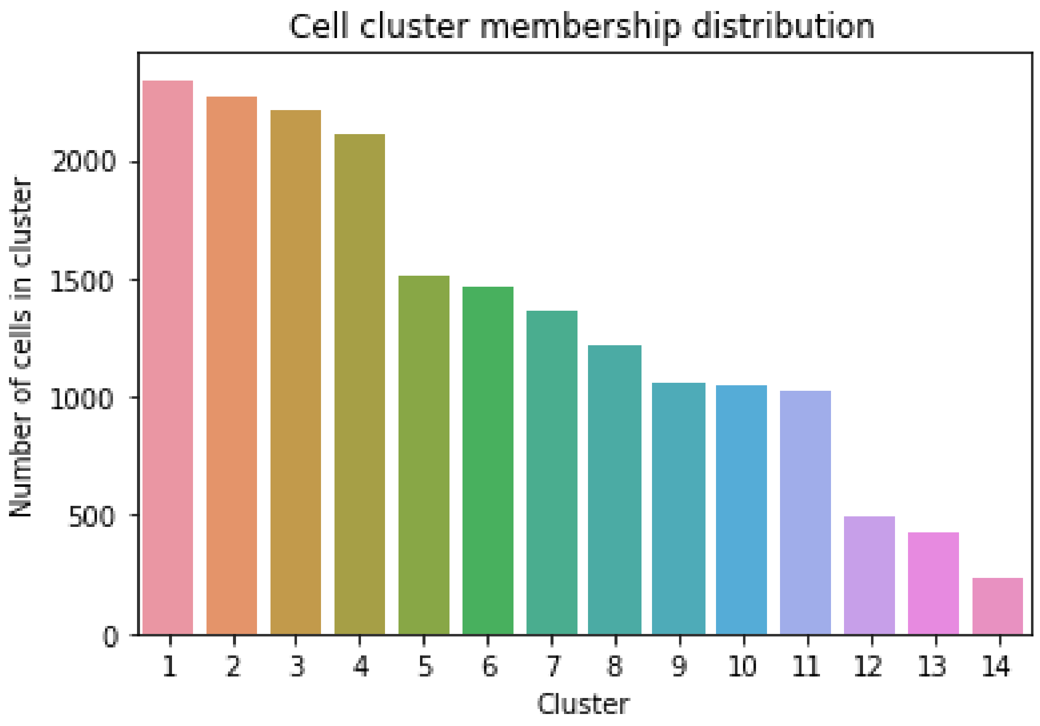
cluster_f = glob(join(curr_in,'analysis',"clustering", "graphclust","clusters.
 ↪csv"))
clusters_df = pd.read_csv(cluster_f[0])
#clusters_df

%matplotlib inline
sns.countplot(clusters_df["Cluster"])
plt.title("Cell cluster membership distribution")
plt.ylabel("Number of cells in cluster")
plt.xlabel("Cluster")
## Load peaks matrix

filename = join(curr_in,'filtered_peak_bc_matrix/matrix.mtx')
peaks_sparse_mtx = pd.read_csv(filename,sep=' ',skiprows=2, header=None).iloc[1:
 ↪].reset_index(drop=True)
peaks_sparse_mtx.columns = ["Peak", "Cell", "Count"]



#peaks_sparse_mtx
```



0.1 Look at marker genes from Lin et al

[5]:
```
immune_markers ={"cKit":"KIT",
                 "Sca1": "LY6E", #"Ly6a",  #?? LY6K LY6E LY6H
                 "CD11c": "ITGAX",
                 "CD150": "SLAMF1",
                 "CD34": "CD34",
                 "CD16/32":"FCGR3A",
                 "CD45.1": "PTPRC",
                 "CD45.2":"PTPRC",
                 "CD48": "SLAMF2", # Other SLAMFs
                 "IL7Ra":"IL7R",
                 "CD11b":"ITGAM"}
immune_genes = list(immune_markers.values())
immune_genes
```

[5]:
```
['KIT',
 'LY6E',
 'ITGAX',
 'SLAMF1',
 'CD34',
 'FCGR3A',
 'PTPRC',
 'PTPRC',
 'SLAMF2',
 'IL7R',
 'ITGAM']
```

[6]:
```
samples = samples.drop(0,axis=1).rename({1:"Condition"}, axis=1)
samples=pd.concat((samples,clusters_df), axis=1)
samples.index=samples.index+1
samples.head()
```

[6]:
```
  Condition             Barcode  Cluster
1     Flt3l  AAACGAAAGAGCTCCC-1        4
2      Ctrl  AAACGAAAGAGGTCCA-2        5
3     Flt3l  AAACGAAAGCGATACG-1        1
4      Ctrl  AAACGAAAGCGATACG-2        6
5     Flt3l  AAACGAAAGGCTTCGC-1        5
```

0.2 Filter for the immune genes

Uses peak_annotations

[7]:
```
imm_anno = peak_annotations[peak_annotations["gene"].isin(immune_genes)].
 ↪set_index("Peak", drop=True)
imm_anno.head()
```

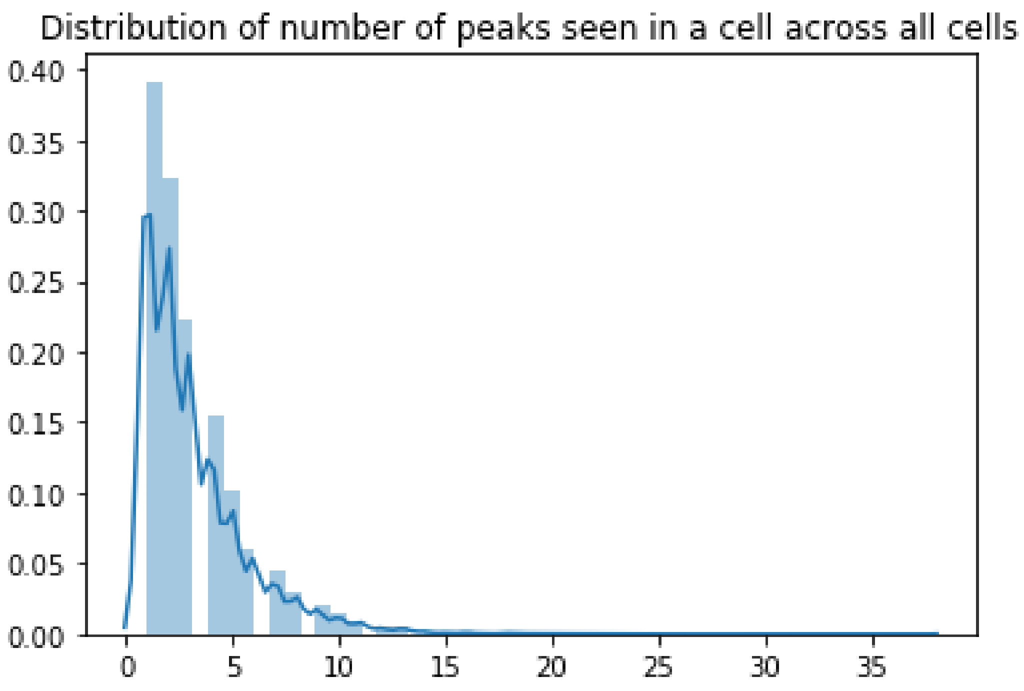
[7]:
```
                          peak    gene distance peak_type
Peak
7214  chr1_160618791_160619091  SLAMF1    27867    distal
7215  chr1_160625647_160627586  SLAMF1    19372    distal
7216  chr1_160629765_160630958  SLAMF1    16000    distal
7217  chr1_160640326_160640793  SLAMF1     6165    distal
7218  chr1_160641360_160641784  SLAMF1     5174    distal
```

[8]:
```
peaks_sparse_mtx = peaks_sparse_mtx.loc[peaks_sparse_mtx["Peak"].isin(imm_anno.
 ↪index)]
peaks_sparse_mtx["gene"] = peaks_sparse_mtx["Peak"].apply(lambda x:␣
 ↪peak_annotations.loc[x-1,"gene"])
peaks_sparse_mtx["Cluster"] = peaks_sparse_mtx["Cell"].apply(lambda x:␣
 ↪clusters_df.loc[x-1,"Cluster"])
peaks_dense = peaks_sparse_mtx.pivot(index="Peak",␣
 ↪columns="Cell",values="Count").fillna(0)
```

0.3 The number of cells within a peak and number of peaks within a cell.

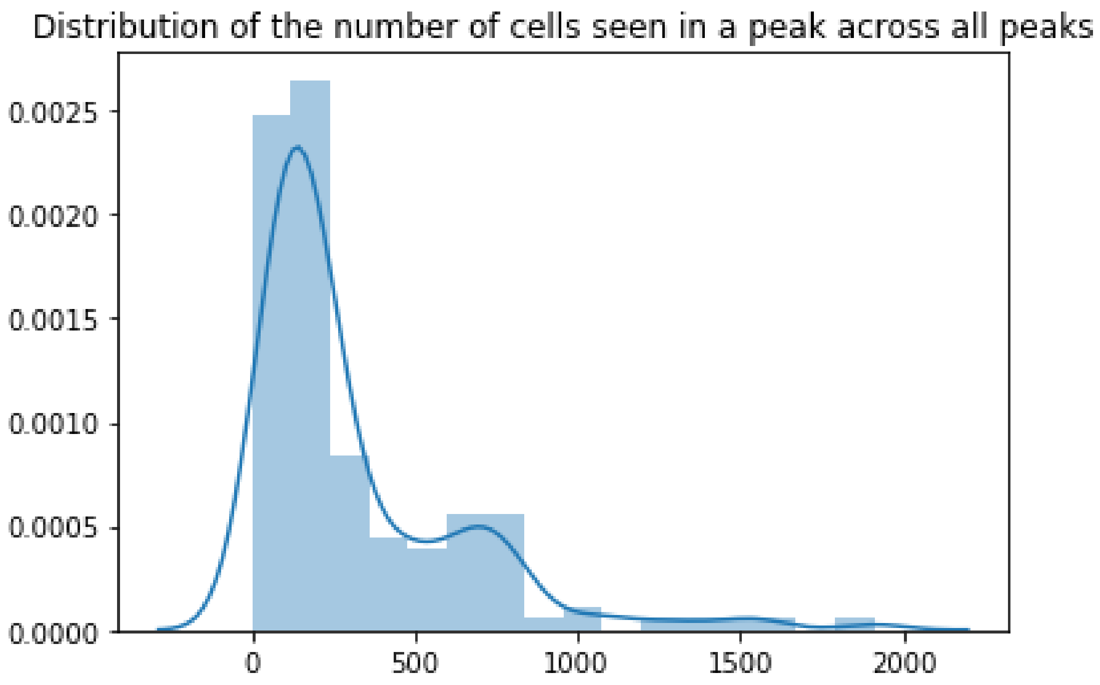
[9]:
```
sns.distplot((peaks_dense>0).sum(axis=0))
plt.title("Distribution of number of peaks seen in a cell across all cells")
```

[9]:
```
Text(0.5, 1.0, 'Distribution of number of peaks seen in a cell across all
cells')
```



[10]:
```
sns.distplot((peaks_dense>0).sum(axis=1))
plt.title("Distribution of the number of cells seen in a peak across all peaks")
```

[10]:
```
Text(0.5, 1.0, 'Distribution of the number of cells seen in a peak across all
peaks')
```



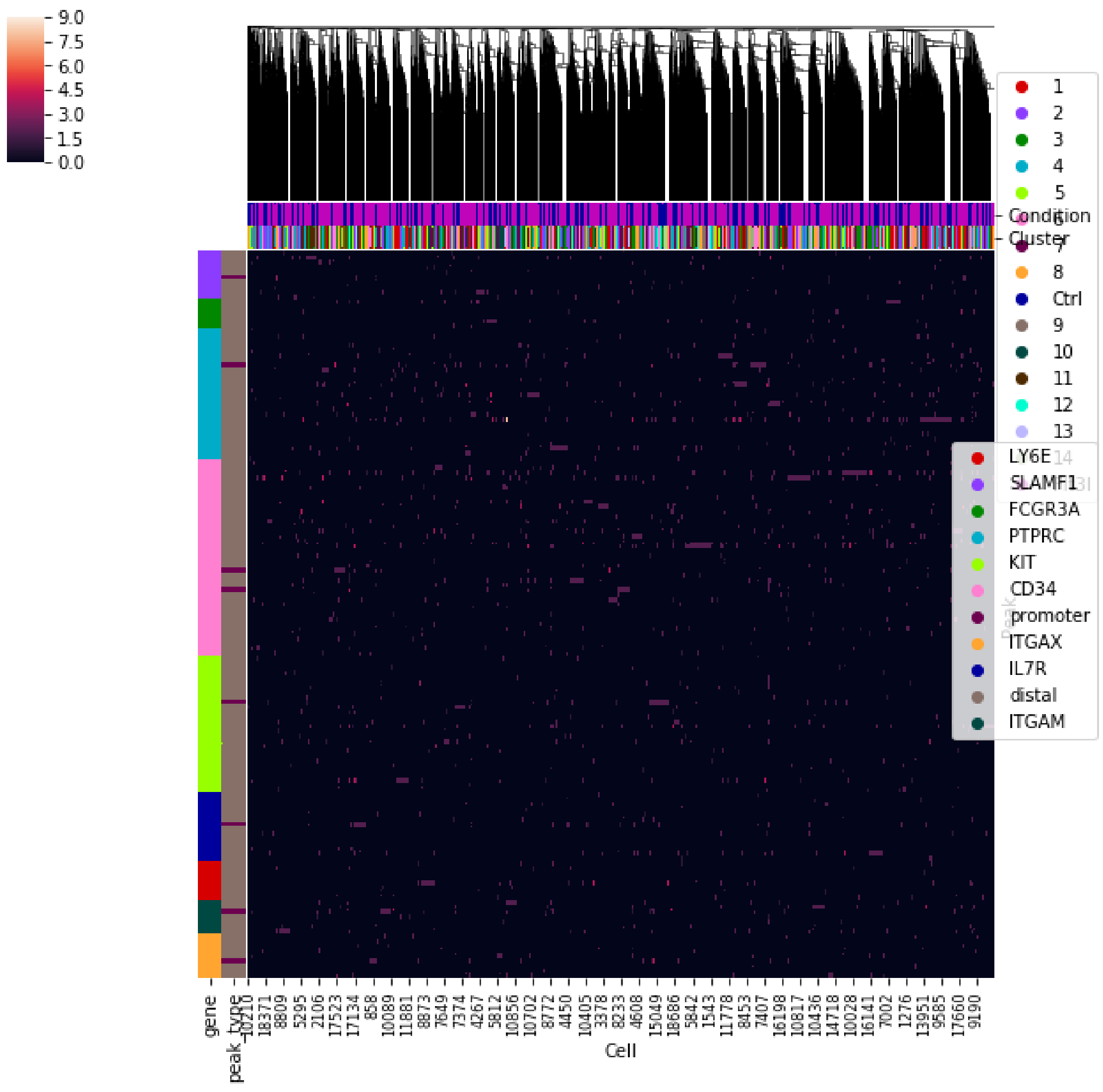
```
[11]: #imm_anno = peak_annotations[peak_annotations["gene"].isin(immune_genes)].copy()
      #imm_inds = peak_annotations.index
      imm_anno["ID"] = imm_anno.apply(lambda x:␣
       ↪x["gene"]+"_"+x["peak_type"]+"_"+x["distance"],axis=1)
      imm_peaks_dense = peaks_dense.loc[imm_anno.index].copy()
      #imm_peaks_dense = imm_peaks_dense.rename(imm_anno.set_index("Peak").
       ↪apply(lambda x: x["gene"]+"_"+x["peak_type"],axis=1), axis=0)
      #imm_peaks_dense
```

0.4 Cluster based on IM genes

0.4.1 a. Dont cluster the immune peaks

```
[12]: #rand_df = imm_peaks_dense.sample(n=500,axis=1)
      ch.plot_cluster(imm_peaks_dense.fillna(0), row_meta=imm_anno[["gene",␣
       ↪"peak_type"]],
                      col_meta=samples.drop("Barcode",axis=1), to_row_clust=False,␣
       ↪row_names=False,
                      metric='jaccard', to_legend=True, white_name=None )
```

```
[12]: <seaborn.matrix.ClusterGrid at 0x7ff9951ce320>
```

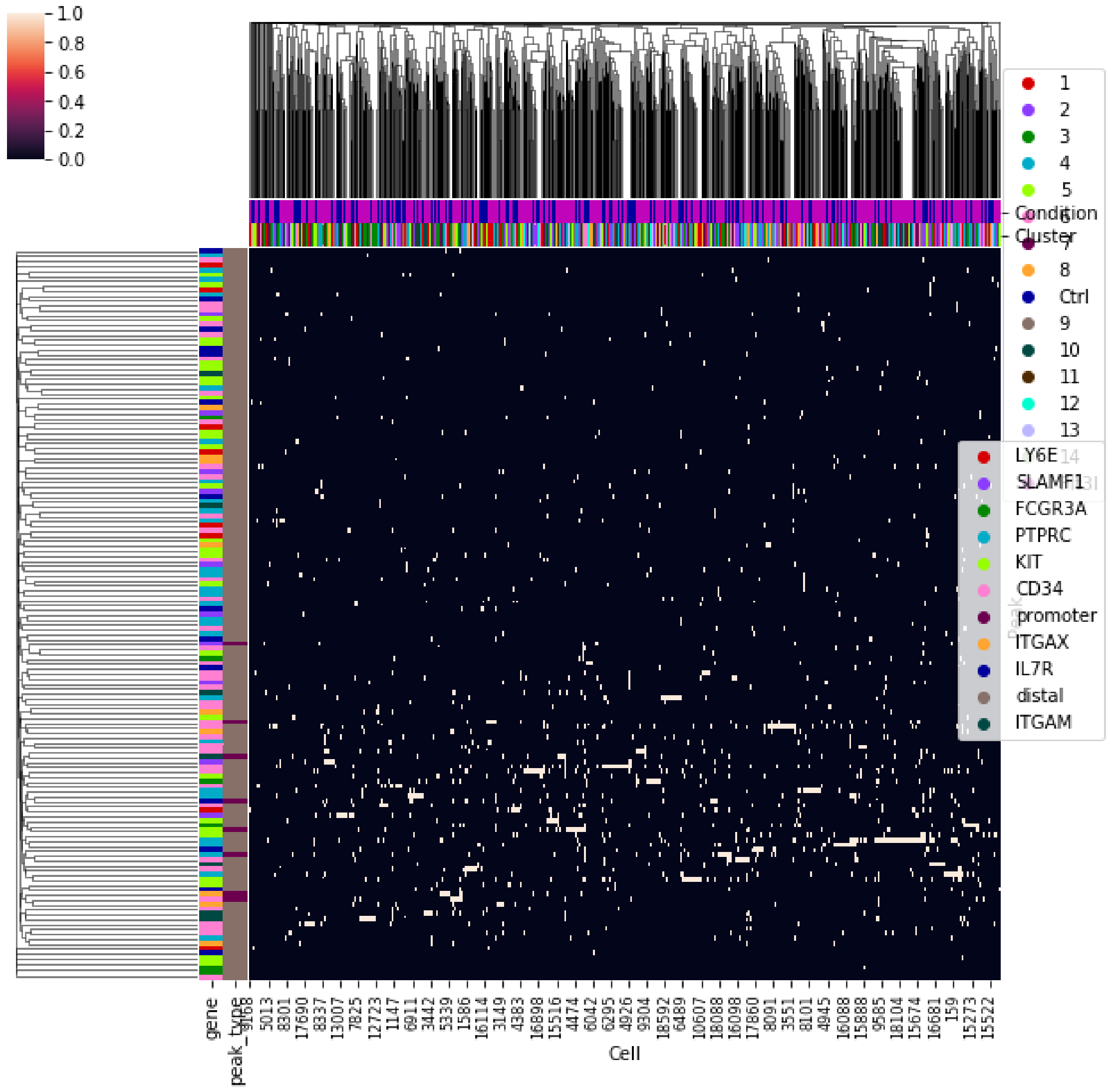


0.4.2 b. Clustering the immune peaks as well

and binarizing the results as 0 or >0

[13]:
```
#geneType_imm_anno = imm_anno.rename(imm_anno.apply(lambda x:␣
↪x["gene"]+"_"+x["peak_type"],axis=1), axis=0)
rand_df = imm_peaks_dense.sample(n=1000,axis=1)
ch.plot_cluster(rand_df.fillna(0), row_meta=imm_anno[["gene", "peak_type"]],
                col_meta=samples.drop("Barcode",axis=1).loc[rand_df.columns],␣
↪to_row_clust=True,
                metric='jaccard', vmax=1, to_legend=True, white_name=None,␣
↪row_names=False )
```

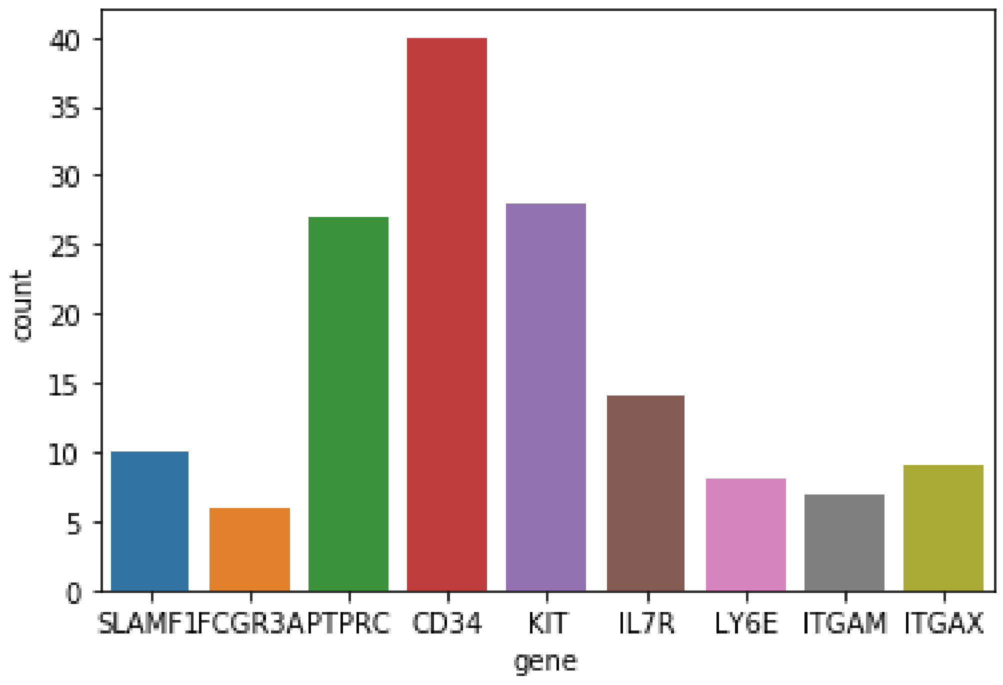
```
[13]: <seaborn.matrix.ClusterGrid at 0x7ff910f6e6a0>
```



0.5 Countplots for each gene and peak type

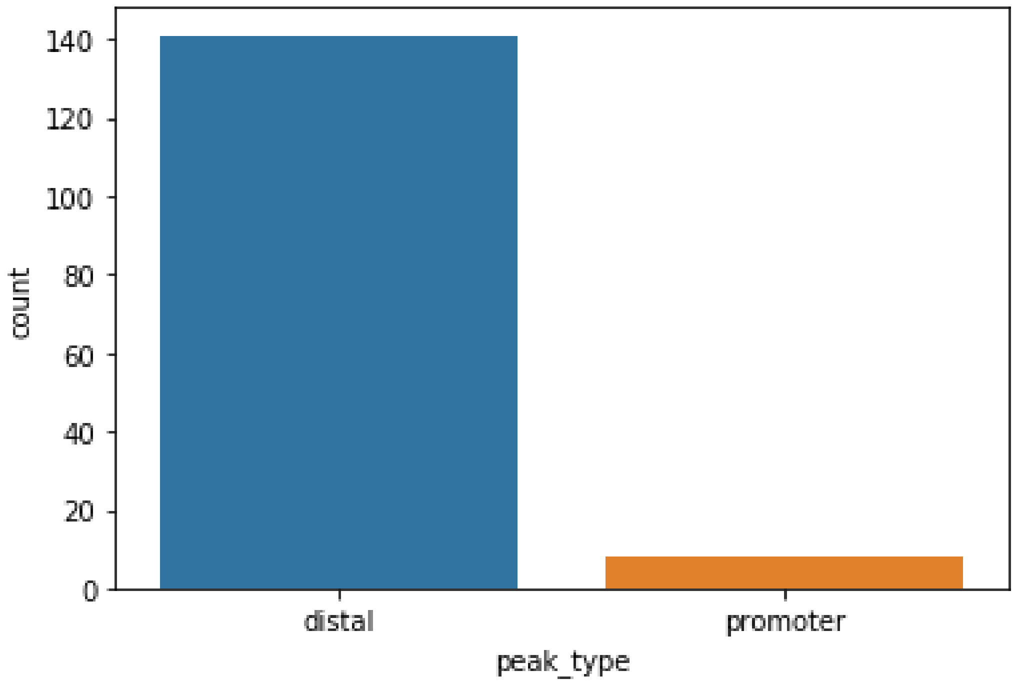
```
[14]: sns.countplot(imm_anno["gene"])
```

```
[14]: <matplotlib.axes._subplots.AxesSubplot at 0x7ff90c093e48>
```



```
[15]: sns.countplot(imm_anno["peak_type"])
```

```
[15]: <matplotlib.axes._subplots.AxesSubplot at 0x7ff90c054f60>
```



```
[ ]:
```

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[ ]:
```