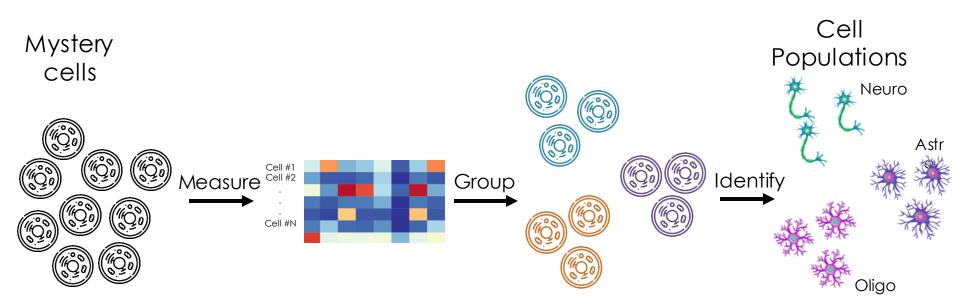
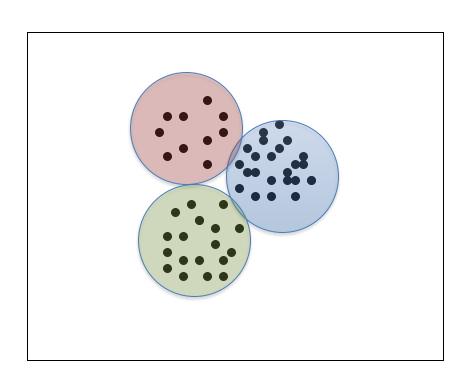


Clustering: Cell Identity





Clustering: Cell Identity

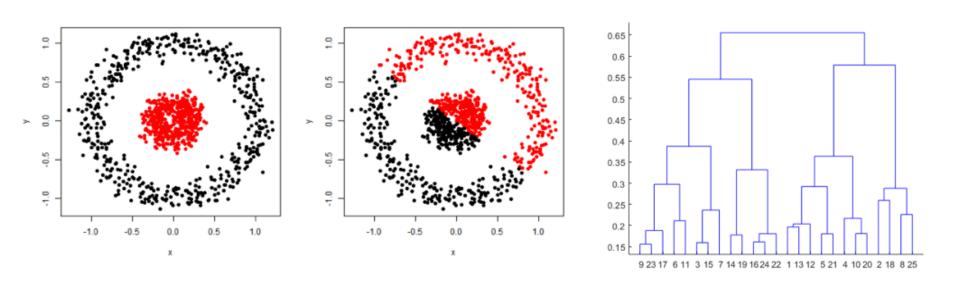


Clustering

Clustering method are divided into two categories*:



Hierarchical clustering



*Handbook of cluster analysis, Hennig C. et al.

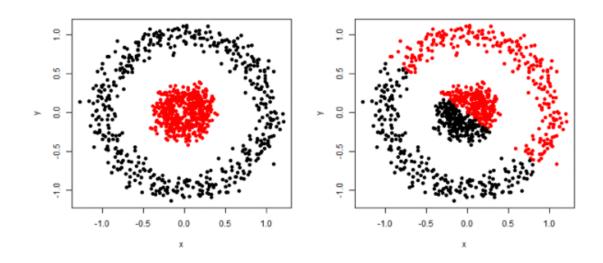
Partitioning clustering

Convex partitioning. Example: K-means

Density based approaches. Example: DBSCAN

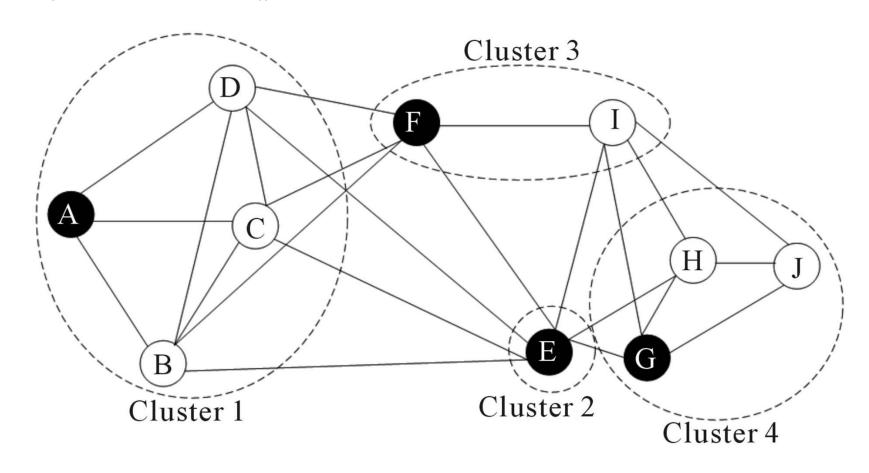
Model-based approaches. Example: Mclust

Graph based approaches: Example to follow



Graph-based

- Nodes -> cells
- Edges -> similarity ()



Graph-based: types

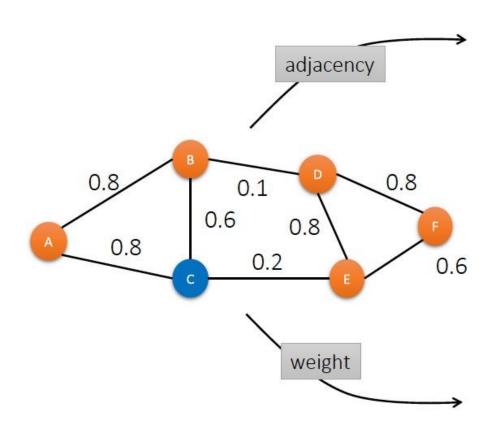
• k-Nearest Neighbor (kNN) graph

A graph in which two vertices p and q are connected by an edge, if the distance between p and q is among the k-th smallest distances from p to other objects from P.

• Shared Nearest Neighbor (SNN) graph

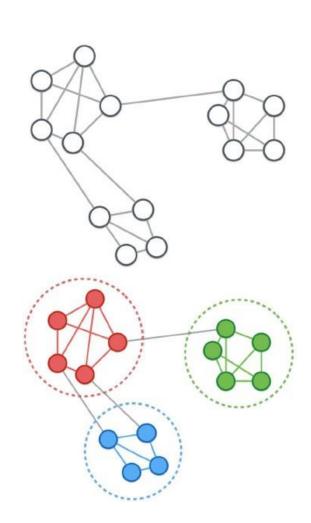
A graph in which weights define proximity, or similarity between two nodes in terms of the number of neighbors (i.e., directly connected nodes) they have in common.

Graph-based: types



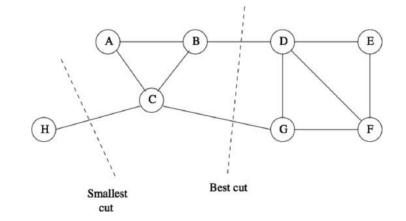
Graph-based: communities

- Communities (clusters):
 - roups of nodes with higher probability of being connected to each other than to members of other groups
- Community detection:
 - Find a group (community) of nodes with **more edges inside** the group than edges linking nodes of the group with the rest of the graph.



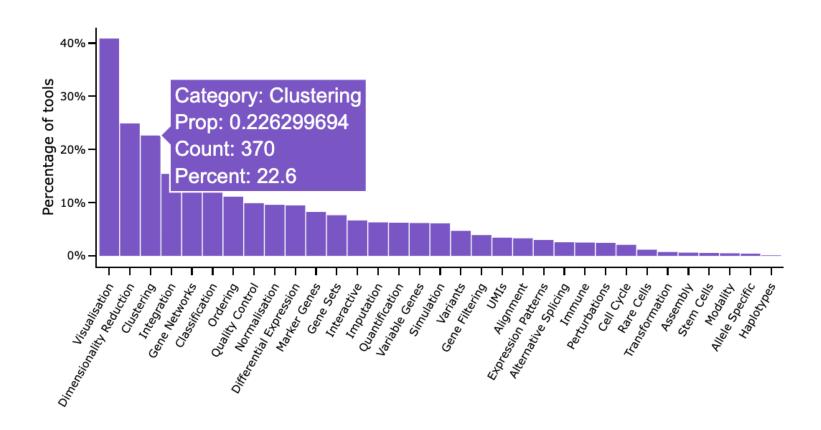
Graph-based: Cuts

- Graph cut partitions a graph into subgraphs
- Cut size is the number of cut edges
- Clustering by graph cuts: find the smallest cut that bi-partitions the graph
- The smallest cut is not always the best cut
- NP-hard
 - Heuristic methods applied e.g. Louvain



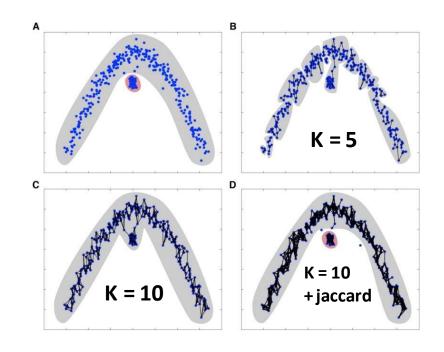
scRNA-seq clustering methods





Seurat

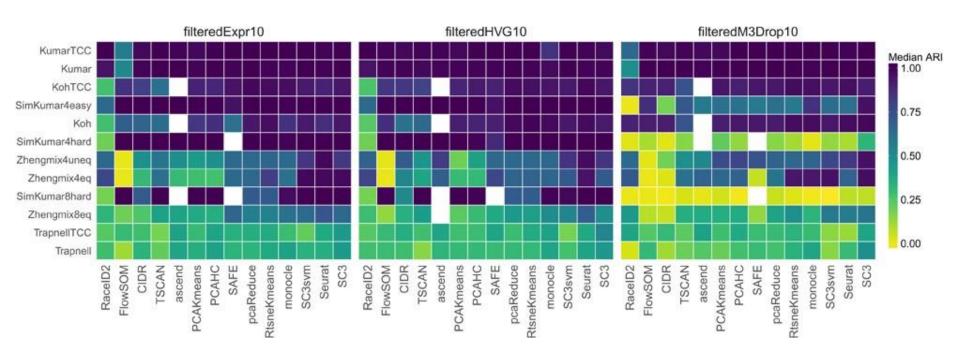
- Construct SNN graph based on the Euclidean distance in PCA space.
 (Default, but could be also kNN)
- Refine the edge weights between any two cells based on the shared overlap in their local neighborhoods (Jaccard index).
- Cluster cells by optimizing for modularity (cuts) (Louvain algorithm) (Modularity is a cost function, resolution is a parameter used to calculate the modularity)



obj <- FindClusters(obj)</pre>

Benchmarking





Clustering: Challenges

- What is a cell type?
- What is the number of clusters k?
- Check QC after clustering to see if no biases are constituting your clusters
- Clustering is subjective No ground truth
- How stable are the clusters
- How dependent are the clusters on the surrounding cells
- •Scalability: in the last few years the number of cells in scRNA-seq experiments has grown by several orders of magnitude from $\sim 10^2$ to $\sim 10^6$