Multi Omics Clustering

Outline

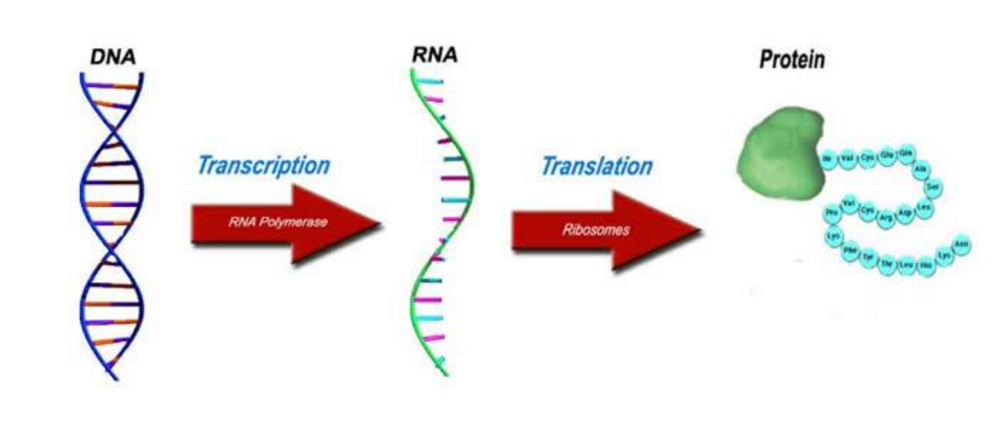
- Introduction
- Cluster of Clusters (COCA)
- iCluster
- Nonnegative Matrix Factorization (NMF)
- Similarity Network Fusion (SNF)
- Multiple Kernel Learning (MKL)

Outline

- Introduction
- Cluster of Clusters (COCA)
- iCluster
- Nonnegative Matrix Factorization (NMF)
- Similarity Network Fusion (SNF)
- Multiple Kernel Learning (MKL)

Omics

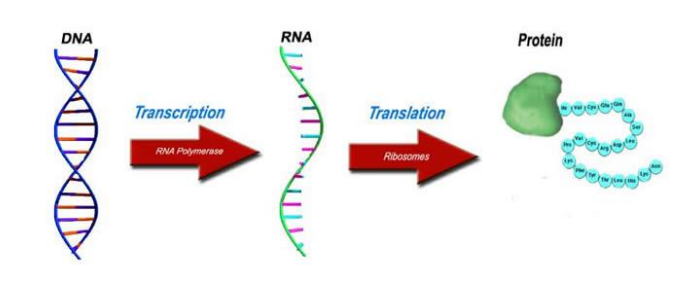
- "Basic dogma of biology":



- So far in the course – mainly RNA
- Can't we use DNA or protein data?

Omics

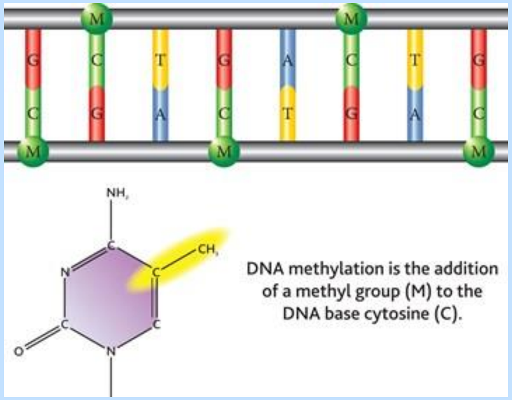
- "Basic dogma of biology":



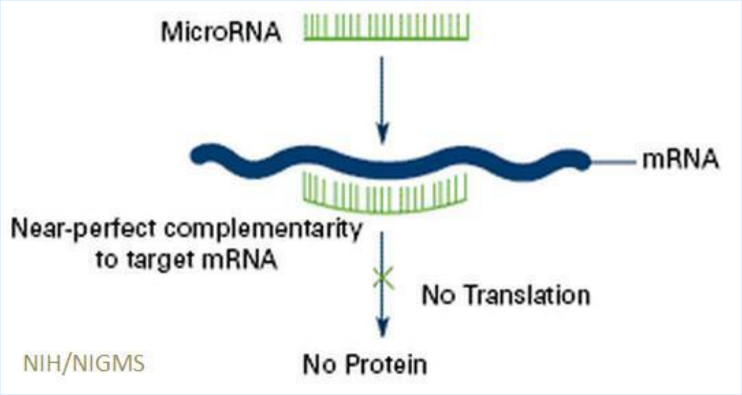
- Omics - characterization of specific type of biological data
- Anything that ends with –ome
- Genome, genomic (adjective), genomics (study of genome)
- Genome, transcriptome, proteome

Additional Omics

- All cells in the human body share (approximately) the same DNA
- However, different genes are expressed and in different abundance in different tissues
- Regulation that is present not only in the genome
- Methylation and microRNA

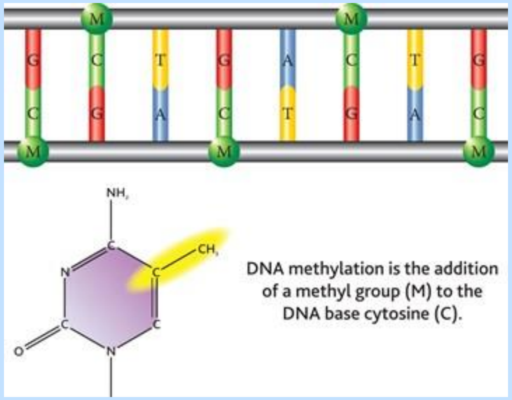


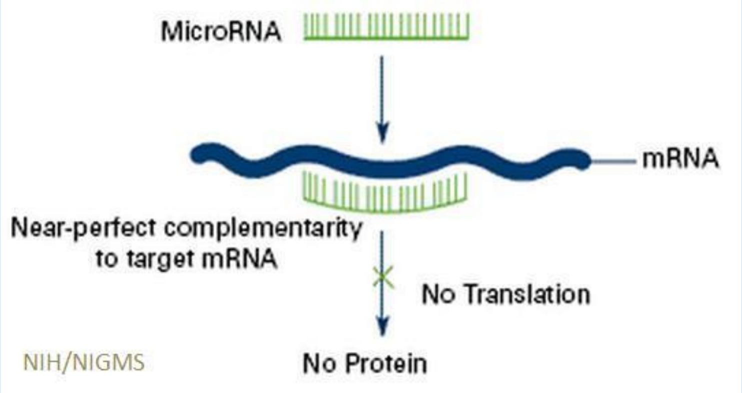
DNA methylation is the addition of a methyl group (M) to the DNA base cytosine (C).



Additional Omics

- Methylation and microRNA

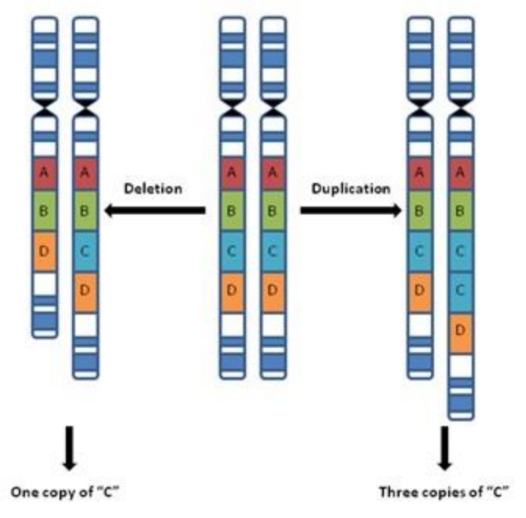

M
G C T G A C T G
C G A C T G A C
NH2
CH3
N
C
O
DNA methylation is the addition of a methyl group (M) to the DNA base cytosine (C).



- Methylation – "punctuation" for the genetic code
  - Methylation of promoters correlated with decreased expression
- MicroRNA – RNA molecules not coding for protein
  - Can stop RNA from being translated

Additional Omics

- Copy number variations



- Prevalent in cancer

Additional Omics

- Genome
- Transcriptome (expression)
- Proteome
- Methylome
- MicroRNA
- Copy number variations
- (Clinical parameters)

- All can be measured in a high throughput manner
- (Either arrays, sequencing, or mass spectrometry)

Additional Omics

- Genome
- Transcriptome (expression)
- Proteome
- Methylome
- MicroRNA
- Copy number variations
- (Clinical parameters)

- Can be used to answer different questions
  - Predict phenotype from genotype
  - Predict age from methylation

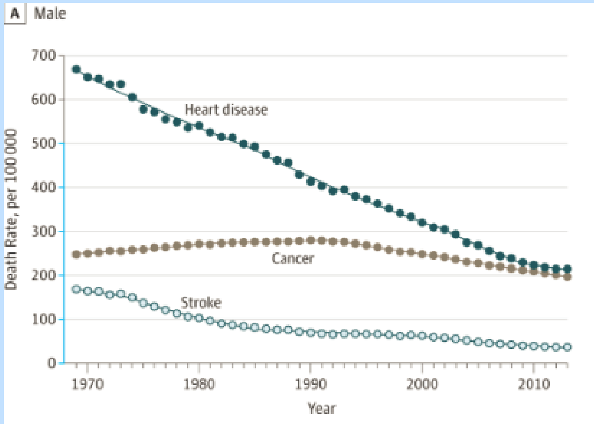
Multi Omics

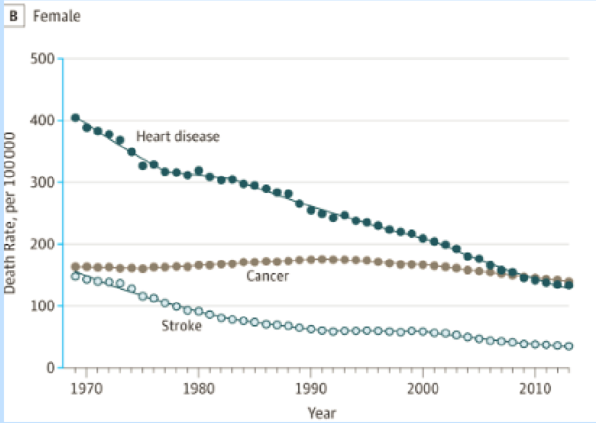
- Using several types of omics data
- Multi omics clustering
- Multi omics dimension reduction
- Multi omics predictions
- …

- This talk: multi omics clustering for cancer subtyping

Cancer Subtyping

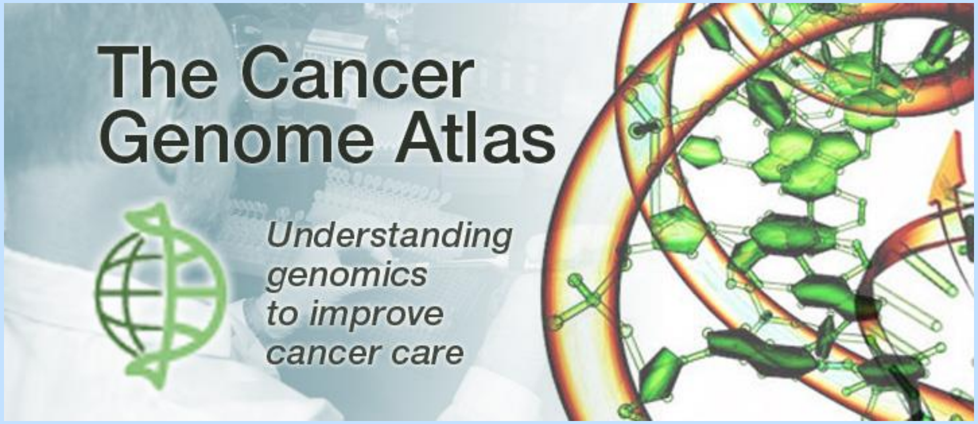
- Cancers are heterogeneous (even within a tissue)
- Therapeutic decisions based on pathologic parameters and biomarkers
- High throughput expression data used in recent years (PAM50, MammaPrint, Oncotype…)
- Copy number, methylation etc. has a known role in cancer prognosis





TCGA

- The Cancer Genome Atlas
- Collect and analyze data from cancer patients using high throughput technologies
- Samples from 11000 patients, more than 30 tumor types
- (Hundreds of millions of dollars)



Multi Omics Data

- Mutations – binary (or sequence)
- Copy number variations – counts
- Gene expression, micro RNA expression, protein arrays – numerical (hundreds miRNA, 20000 genes)
- DNA methylation – beta value (up to 450K sites)
- Clinical parameters – age, tumor size…

	Gene1 Exp	Gene2 Exp	Gene3 Exp
Patient1	323	643	50
Patient2	356	712	38
Patient3	344	680	58

	CpG 1	CpG 2	CpG 3
Patient1	0.2	0.3	0.12
Patient2	0.25	0.32	0.17
Patient3	0.23	0.35	0.09

Approaches

Early integration

- Concatenate matrices
- Dimensionality
- Data from different distributions

Intermediate integration

- Omics are different “views” of clusters
- Build model using all omics

Late integration

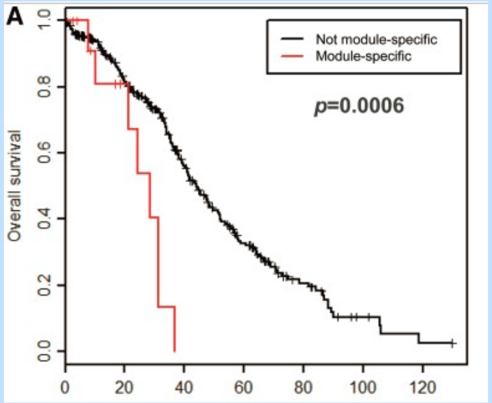
- Consensus clustering
- Dependencies between features from different omics
- Weak but consistent signals

Approaches

- Support for any omic data type
  - General
  - Loses knowledge of the biological role
  - (Continuous vs. discrete)
- Omic specific methods
  - For example – expression is increasing in copy number
- Omic specific feature representation
  - Replace genes with pathways

Comparing Clusterings

- Compare to “gold standard”
  - No gold standard for cancer subtypes
- Create synthetic data
- Compare prognosis or other clinical and genomic features
- Use homogeneity, separation, silhouette score…



Silhouette Score

- a(i) – average distance of i to points within its cluster
- b(i) – average distance of i to points within closest cluster it doesn't belong to
- Silhouette score for i:
- s(i) = $\frac{b(i)-a(i)}{\max(b(i),a(i))}$
- Between -1 and 1
- Silhouette score for clustering is average silhouette score across samples
- (Requires a definition of distance)

Introduction - Recap

- Omics
- Multi omics and how the datasets look
- Cancer subtyping
- TCGA
- Multi omics clustering approaches
- Comparing clusterings

Outline

- Introduction
- Cluster of Clusters (COCA)
- iCluster
- Nonnegative Matrix Factorization (NMF)
- Similarity Network Fusion (SNF)
- Multiple Kernel Learning (MKL)

Cluster of Clusters (COCA)

- Hoadley et. al (Cell, 2014)
  - (as part of The Cancer Genome Atlas Research Network)
- Late integration method

- Tissue of origin is heavily used for therapeutic decision making
- Cluster TCGA samples from multiple tissues
- What are the clusters? Do they match the tissue of origin?

Consensus Clustering Reminder

- The data $D=\{e_1, \ldots e_N\}$; e_i : GE profile of sample/patient #i
- Want a partition $\{P_1,\ldots P_k\}$ of the items
- $D^{(h)}$: resampled dataset #h
- $M^{(h)}$: result of clustering $D^{(h)}$
  - $M^{(h)}(i,j) = 1$ if i,j in same cluster, 0 o/w
- $I^{(h)}(i,j) = 1$ if i,j are both included in $D^{(h)}$
- $\mathcal{M}(i,j) = \Sigma_h M^{(h)}(i,j) / \Sigma_h I^{(h)}(i,j)$

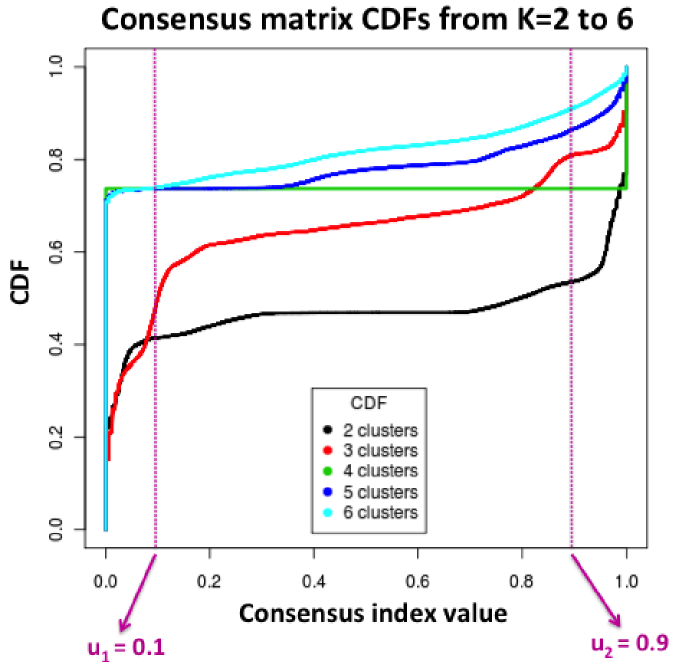
$\mathcal{M}$: consensus matrix

$\mathcal{M}(i,j)$ consensus index of i,j

- Change to distance: $\mathcal{D}(i,j) = 1 - \mathcal{M}(i,j)$
- Cluster $\mathcal{D}$ using a distance based method, e.g. HC

Consensus Clustering Reminder

- CDF(c) = $|\{(i,j)\ |\ i<j, \mathcal{M}(i,j) \leq c\}|\ /\ N(N-1)/2$



PAC for each K is $CDF_k(u_2)$ - $CDF_k(u_1)$.
According to this criterion, optimal K here is 4.

https://en.wikipedia.org/wiki/Consensus_clustering

COCA

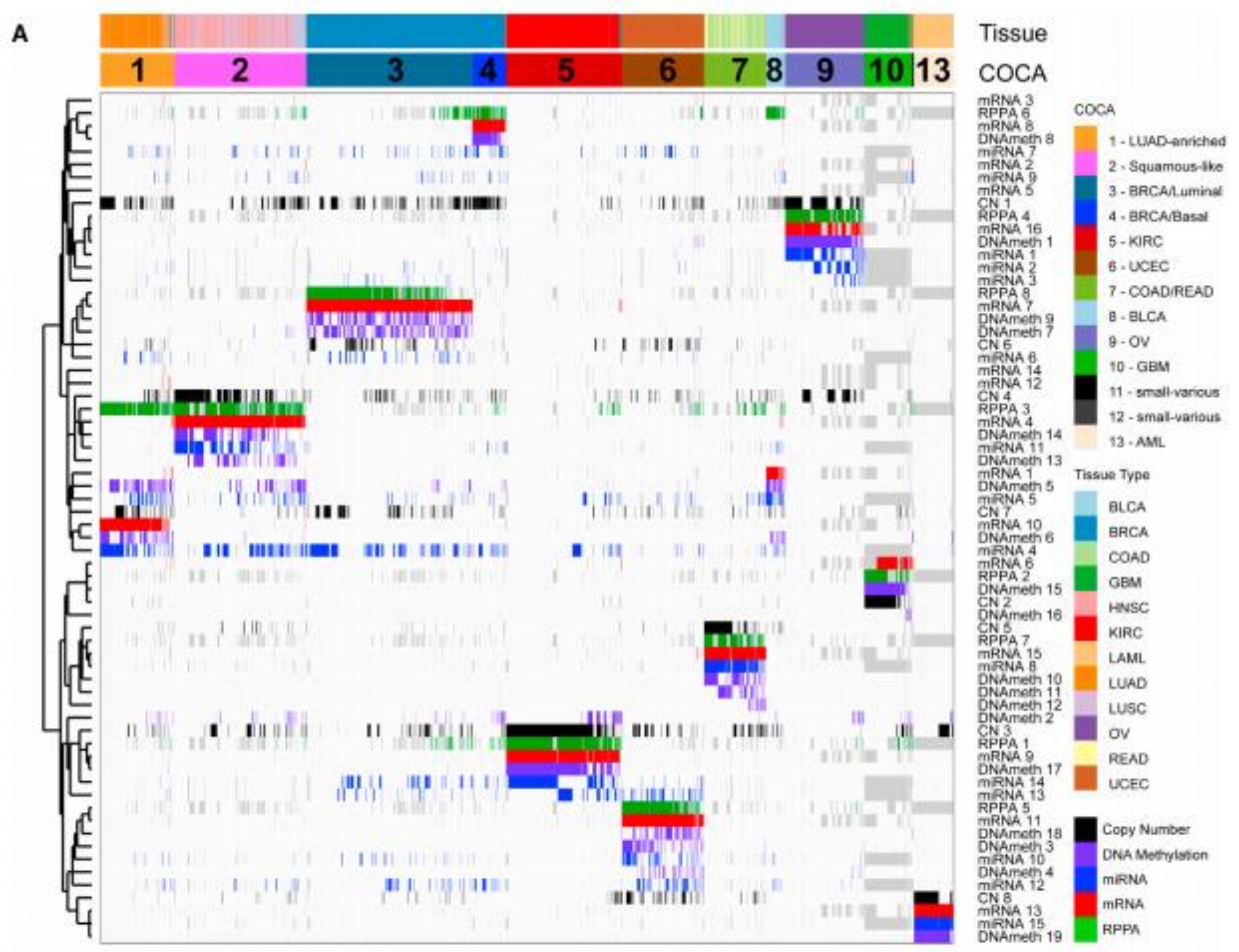
- Cluster each omic separately
  - Can use any clustering algorithm for each omic
  - different omics can use different k
- Represent each sample by an indicator vector of the single omic cluster memberships:
  - Sample is in cluster 3 out of 5 in omic 1: (0, 0, 1, 0, 0)
  - Sample is in cluster 2 out of 3 in omic 2: (0, 1, 0)
  - Sample representation: (0, 0, 1, 0, 0, 0, 1, 0)
- Run consensus clustering on the new representation (80% sampling, hierarchical clustering algorithm) for the samples and return its output

COCA - Results

- Run on all (3527) TCGA samples from 12 cancer tissues
- Use expression, methylation, miRNA, copy number, RPPA (protein arrays)
- Each with different clustering scheme - hierarchical, NMF, consensus clustering…

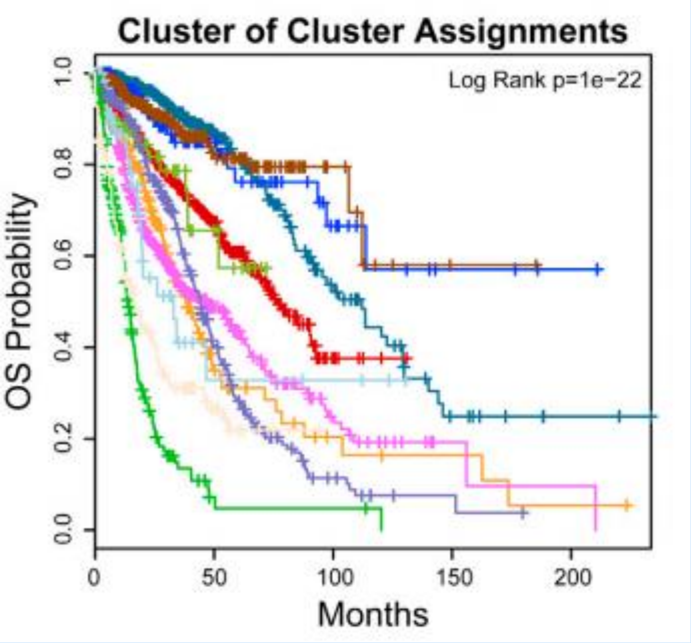
- 11 clusters, 5 nearly identical to tissue of origin
- Lung squamous, head and neck cluster together
- Bladder cancer split into 3 pan-cancer subtypes

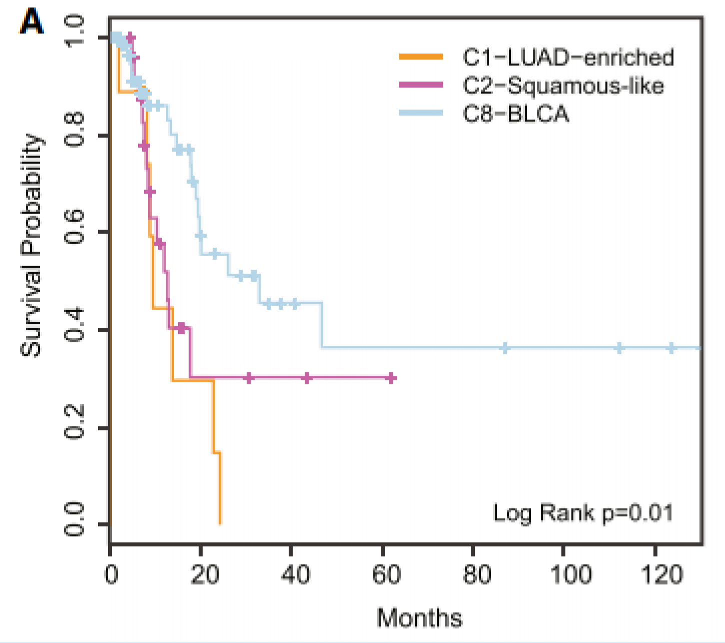
COCA - Results



COCA - Results

- Survival analysis of the different clusters
- Survival analysis of bladder cancers within different clusters





COCA - Recap

- Algorithm:
  - Cluster each omic separately
  - Cluster membership indicator representation
  - Consensus clustering on that representation
- Run on TCGA data from all available tissues
- Clusters generally match tissue of origin, with few exceptions (squamous cancers, bladder cancer)

Outline

- Introduction
- Cluster of Clusters (COCA)
- iCluster
- Nonnegative Matrix Factorization (NMF)
- Similarity Network Fusion (SNF)
- Multiple Kernel Learning (MKL)

iCluster

- Shen, Olshen, Ladanyi (Bioinformatics, 2009)
  - Memorial Sloan-Kettering Cancer Center, New York
- Dimension reduction
- m different omics, X_i observed matrices of dimension $p_i \; x \; n$
- Z – $k \; x \; n$ cluster membership binary matrix
- $\begin{matrix} 1 & 0 & 1 & 0 & 0 \\ 0 & 0 & 0 & 1 & 0 \\ 0 & 1 & 0 & 0 & 1 \end{matrix}$

iCluster

- m different omics, X_i observed matrices of dimension $p_i\ x\ n$
- Z – $k\ x\ n$ cluster membership binary matrix
- $X = WZ + \epsilon$
- ϵ is added per column
- It is normal with zero mean and diagonal covariance (each feature has different independent noise)
- Equal observed values for data coming from same cluster (up to Gaussian noise)
- (PCA with membership as low rank representation. Also, very similar to k-means)

iCluster

- $X = WZ + \epsilon$
- Multi omic version:
- $X_1 = W_1 Z + \epsilon_1$
- $X_2 = W_2 Z + \epsilon_2$
- …
- $X_m = W_m Z + \epsilon_m$

- Each ϵ_i is normal with zero mean and diagonal covariance ψ_i (again, added per column)

Bayesian Statistics

- First, Frequentist statistics
- Tossing a coin n times with probability p to heads
- Can estimate p by maximizing the likelihood: Pr(data | p)
- Why maximize Pr(data | p) and not Pr(p | data)?
- p is not a random variable, it is a number!

Bayesian Statistics

- In Bayesian statistics, parameters are random variables
- Pr(p) – prior probability for parameter p (e.g. uniform[0,1])
- Natural way to incorporate domain knowledge to the model
- Pr(p | data) = Pr(data | p) * Pr(p) / Pr(data)
- Can maximize Pr(p|data) or take E[p | data]

iCluster

- $X_1 = W_1 Z + \epsilon_1$
- $X_2 = W_2 Z + \epsilon_2$
- …
- $X_m = W_m Z + \epsilon_m$

- Instead of a discrete Z, use continuous Z*
- Assume prior distribution Z* ~ N(0, I)
- Note that W are numbers, and Z* random variables
- (Other formulations use Z*Z*'=I. Using normal Z* may lose interpretability).

iCluster

- $X_1 = W_1 Z + \epsilon_1$
- $X_2 = W_2 Z + \epsilon_2$
- …
- $X_m = W_m Z + \epsilon_m$

- $W = (W_1, \dots, W_m)'$
- $X = (X_1, \dots, X_m)' \sim N(0, WW' + \psi)$
- Can write the log likelihood and try to numerically optimize

Expectation Maximization

- Reminder: similar problem in de novo motif discovery
- Observed sequence is either a part of a motif or the background – denote the unknown data Z

Outline of EM algorithm:

- **Choose starting θ**
- **Repeat until convergence of θ:**
  - **E-step: Re-estimate Z from θ, X**
  - **M-step: Re-estimate θ, from X, Z**
- **Repeat all of the above for various starting values θ, λ …**

Expectation Maximization

Outline of EM algorithm:

- **Choose starting θ**
- **Repeat until convergence of θ:**
  - **E-step: Re-estimate Z from θ, X**
  - **M-step: Re-estimate θ, from X, Z**
- **Repeat all of the above for various starting values θ, λ …**
- $X_i = W_i Z + \epsilon_i$
- In our case, $\theta = (W, \psi)$

EM for iCluster

- $$l_c(W,\psi,Z) = -\frac{n}{2}\left\{\sum_{i=1}^{m} p_i ln(2\pi) + \ln\det(\Psi)\right\} - \frac{1}{2}\left\{tr((\mathbf{X}-\mathbf{W}\mathbf{Z}^*)'\Psi^{-1}(\mathbf{X}-\mathbf{W}\mathbf{Z}^*)) + tr(\mathbf{Z}^{*'}\mathbf{Z}^*)\right\}.$$ ($\det(2\pi\boldsymbol{\Sigma})^{-\frac{1}{2}}\, e^{-\frac{1}{2}(\mathbf{x}-\boldsymbol{\mu})'\boldsymbol{\Sigma}^{-1}(\mathbf{x}-\boldsymbol{\mu})}$)
- **Number of parameters =$\boldsymbol{O}(\boldsymbol{\Sigma}\mathbf{p_i} * \mathbf{k}) \gg \mathbf{n}$**
- Add regularization: encourage the model to use less parameters
- Lasso regularization (Tibshirani, 1996)
- $l(W,\psi) = l_c(W,\psi) - \lambda * \Sigma_\mathrm{i}\Sigma_\mathrm{j}\Sigma_k|w_{ijk}|$
- λ – tradeoff between likelihood and shrinkage
- Feature selection!

EM for iCluster

- **Repeat until convergence of *θ*:**
  - **E-step: Re-estimate *Z* from *θ*, *X***
  - **M-step: Re-estimate *θ*, from *X*, *Z***
- E-step (expected value of Z given current parameter estimates and data):

$$E[\mathbf{Z}^*|\mathbf{X}]=\mathbf{W}'\Sigma^{-1}\mathbf{X} \text{ and}$$
$$E[\mathbf{Z}^*\mathbf{Z}^{*'}|\mathbf{X}]=\mathbf{I}-\mathbf{W}'\Sigma^{-1}\mathbf{W}+E[\mathbf{Z}^*|\mathbf{X}]E[\mathbf{Z}^*|\mathbf{X}]'.$$

- M-step:

$$\mathbf{\Psi}^{(t+1)}=\frac{1}{n}\text{diag}\left\{\mathbf{X}\mathbf{X}'-\mathbf{W}^{(t)}E[\mathbf{Z}^*|\mathbf{X}]\mathbf{X}'\right\}$$

$$\mathbf{W}_{\text{lasso}}^{(t+1)}=\text{sign}(\mathbf{W}^{(t+1)})\left(|\mathbf{W}^{(t+1)}|-\lambda\right)_+.$$

- $W^{(t+1)}=(XE[Z^*|X]')(E[Z^*Z^{*\prime}|X])^{-1}$
- $\Sigma = WW' + \psi$

EM for iCluster

- E-step (expected value of Z given current parameter estimates and data):

$$E[\mathbf{Z}^*|\mathbf{X}] = \mathbf{W}'\Sigma^{-1}\mathbf{X} \text{ and}$$
$$E[\mathbf{Z}^*\mathbf{Z}^{*'}|\mathbf{X}] = \mathbf{I} - \mathbf{W}'\Sigma^{-1}\mathbf{W} + E[\mathbf{Z}^*|\mathbf{X}]E[\mathbf{Z}^*|\mathbf{X}]'.$$

- M-step:

$$\Psi^{(t+1)} = \frac{1}{n}\operatorname{diag}\left\{\mathbf{X}\mathbf{X}' - \mathbf{W}^{(t)}E[\mathbf{Z}^*|\mathbf{X}]\mathbf{X}'\right\} \qquad \mathbf{W}^{(t+1)}_{\text{lasso}} = \operatorname{sign}(\mathbf{W}^{(t+1)})\left(|\mathbf{W}^{(t+1)}| - \lambda\right)_+ ,$$

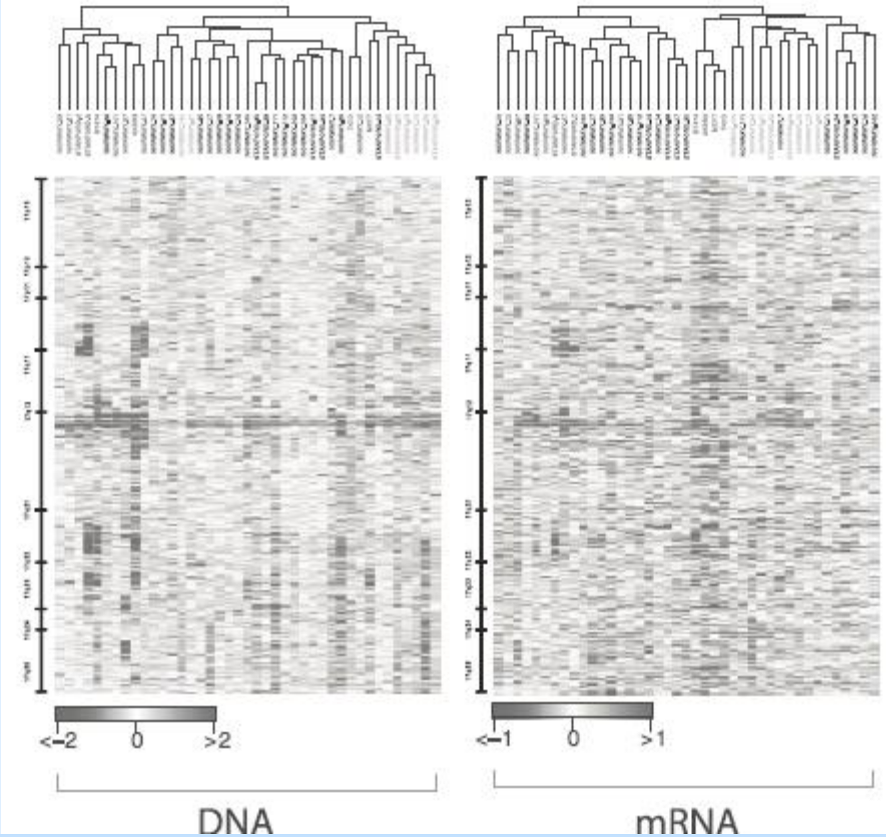
- $W^{(t+1)} = (XE[Z^*|X]')(E[Z^*Z^{*'}|X])^{-1}$
- $\Sigma = WW' + \psi$
- Finally, estimate for Z is given by *E*[Z*|X]
- Run k-means on *E*[Z*|X] to obtain final cluster assignments

iCluster Model Selection

- How to choose k and λ?
- $E[Z^*|X]' E[Z^*|X]$ is n x n matrix
- For cluster matrix Z, ordered by cluster membership, $E[Z|X]' E[Z|X]$ is a perfect 1-0 block matrix
- Measure distance of absolute values between observed normalized $E[Z^*|X]' E[Z^*|X]$, and perfect one
- Measures the posterior probability that two samples belong to the same cluster
- Choose k and λ that minimize the distance

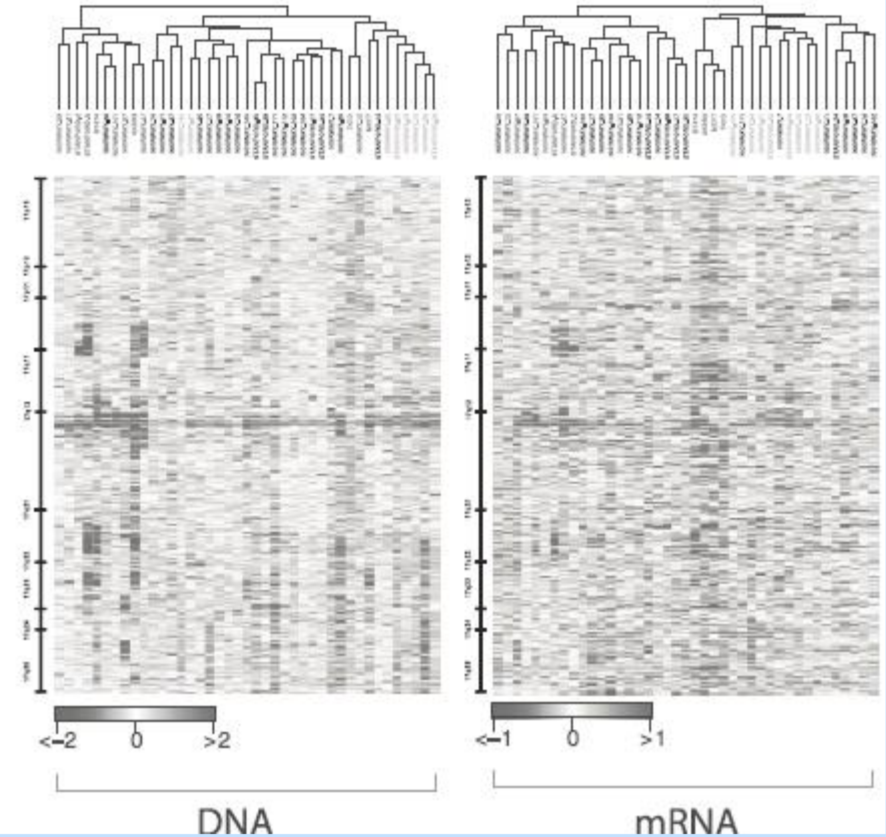
iCluster - Results

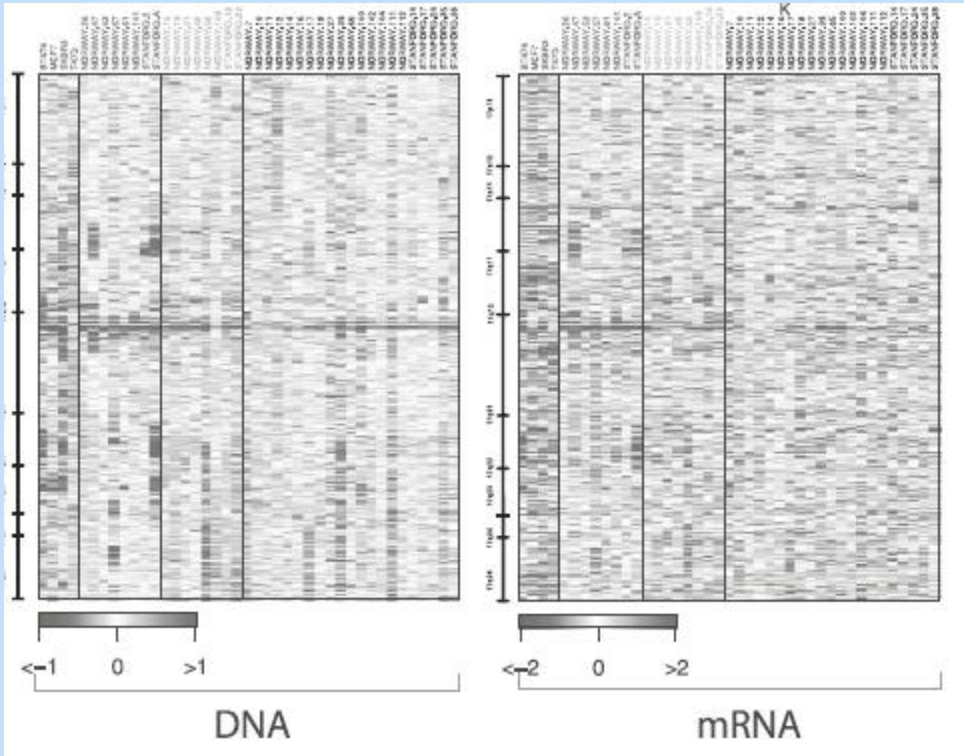
- Dataset: gene expression and copy number variation
  - 37 breast cancer + 4 cell lines samples
  - 91 lung adenocarcinoma
- Separate omic hierarchical clustering



iCluster - Results

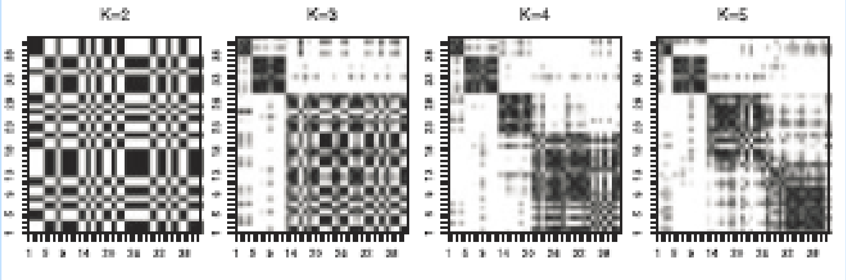
- Separate compared to integrative clustering



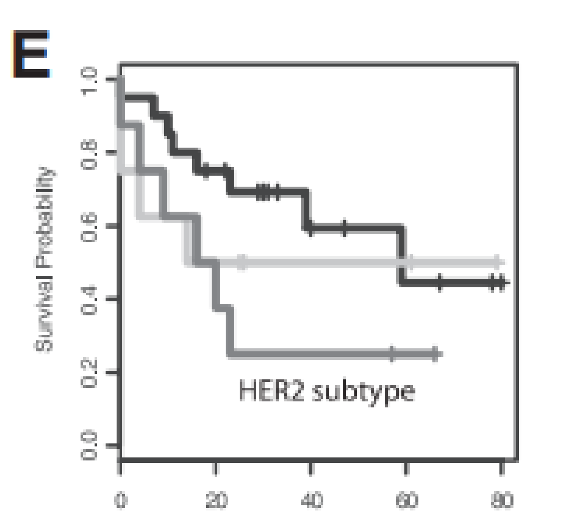


iCluster - Results

- $E[Z^*|X]' \, E[Z^*|X]$ matrix:

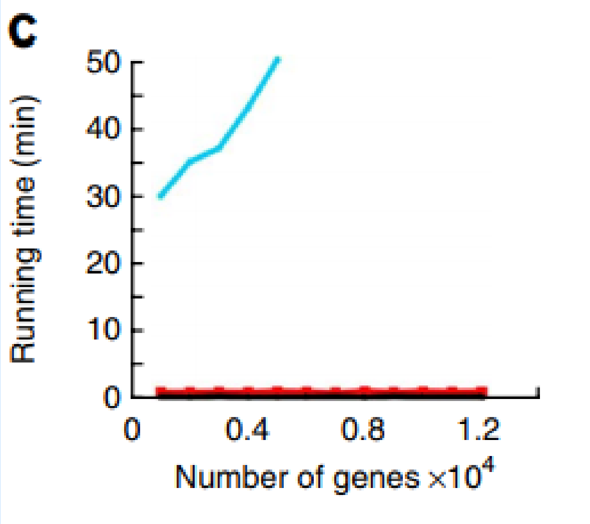


- Survival analysis:



iCluster - Results

- Runtime
- About an hour for ~200 samples, 4000 + 1300 + 500 features, and therefore requires gene preselection



iCluster - Recap

- Low dimension + probabilistic model
- $X_i = W_i Z + \epsilon_i$
- Z and ϵ_i have normal distribution
- Find parameters using EM with regularization for a sparse model
- Use deviation from perfect clustering matrix to determine k and λ
- Run on breast and lung adenocarcinoma samples

Outline

- Introduction
- Cluster of Clusters (COCA)
- iCluster
- Nonnegative Matrix Factorization (NMF)
- Similarity Network Fusion (SNF)
- Multiple Kernel Learning (MKL)

Joint NMF

- Shihua Zhang, …, Jasmine Zhou (2012, bioinformatics)
  - University of Southern California, now at UCLA
- NMF = Nonnegative Matrix Factorization
- Dimension reduction –basic idea similar to iCluster

- Model can be used for clustering, but in this work the main goal is to find "md-modules": (possibly overlapping) sets of features from all omics that define the patients' molecular profile

NMF

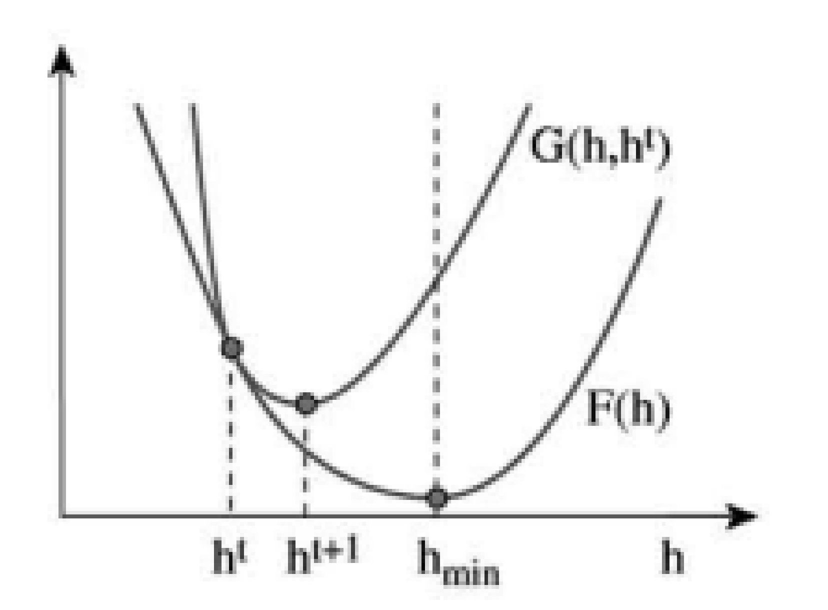
- NMF = Nonnegative Matrix Factorization
- Given a non negative matrix X, factorize it as $X = WH \; s.t \; W, H \geq 0$,
- $x_{.j} = \Sigma_k w_{.k} h_{kj} = W h_{.j}$
- Higher interpretability, makes sense where data is comprised of several parts
- Minimize $||X - WH||_F^2$ ($||A||_F = \sqrt{\Sigma_i \Sigma_j a_{ij}^2}$)
- Often optimized using multiplicative update rule:
- $H_{ab} = H_{ab} \frac{(W^T X)_{ab}}{(W^T W H)_{ab}}, W_{ab} = W_{ab} \frac{(X H^T)_{ab}}{(X H H^T)_{ab}}$

NMF - Proof Sketch

- Lee and Seung, NIPS 2000
- Minimize $||X - WH||_F^2$
- Will show proof sketch for H update rule
- Minimize $\mathrm{F(h)} = \ ||x - Wh||_F^2$
- Definition: G is an auxiliary function for F(h) if: G(h, h') $\geq$ F(h), G(h, h)= F(h)
- Lemma: if G is an auxiliary function, then F is non increasing under the update:$h^{t+1} = argmin_h G(h, h^t)$
- Proof: $F(h^{t+1}) \leq G(h^{t+1}, h^t) \leq G(h^t, h^t) = F(h^t)$

NMF – Proof Sketch

- Definition: G is an auxiliary function for F(h) if: G(h, h') ≥ F(h), G(h, h)= F(h)
- Lemma: if G is an auxiliary function, The F is non increasing under the update:$h^{t+1} = argmin_h G(h, h^t)$



NMF - Proof Sketch

- Lemma (not proved here):
- $K_{ab}(h^t) = \frac{\delta_{ab}(W^T W h^t)_a}{h_a^t}$
- $G(h, h^t) = F(h^t) + (h - h^t)^T \nabla F(h^t) + \frac{1}{2}(h - h^t)^T K(h^t)(h - h^t)$
- Is an auxiliary function for F(h)
- (Easy to see that G(h,h) = F(h))
- $h^{t+1} = argmin_h G(h, h^t)$ gives the update:
- $H_{ab} = H_{ab} \frac{(W^T X)_{ab}}{(W^T W H)_{ab}}$

NMF

- Now in genomic context
- $X = WH,\ x_{.j} = \Sigma_k w_{.k} h_{kj} = Wh_{.j}$
- X is M x N matrix, M patients and N features
- W is M x k matrix, k is the number of modules
- H is k x N matrix

- W's columns are basis vectors for the features (e.g genes), H matrix is the coefficients

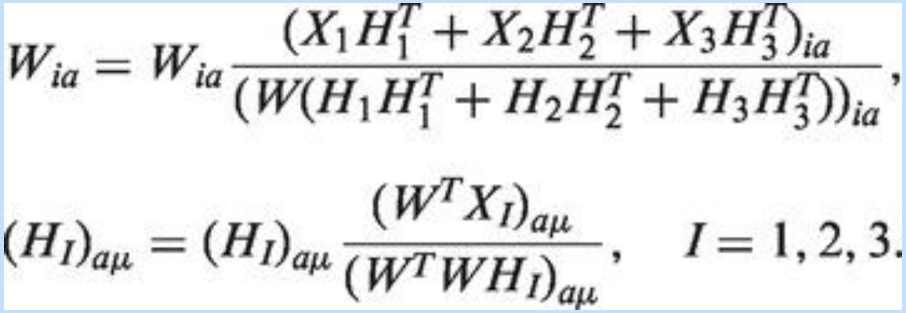
Joint NMF

- $X = WH$, $x_{.j} = \Sigma_k w_{.k} h_{kj} = Wh_{.j}$
- $X_l = WH_l$
- X_l is $\mathrm{M} \times N_l$ matrix, M patients and N_l features
- W is M x k matrix, k is the number of modules
- H_l is $k\ x\ N_l$ matrix

- Basis vectors (W) are identical in all omics, different coefficient matrices

Joint NMF

- $X_l = WH_l$
- Optimization problem is $\min \Sigma_l \, ||X_l - WH_l||_F^2, W \geq 0 \, , H_l \geq 0$
- Adapt the single matrix multiplicative update rule (ex):
- $H_{ab} = H_{ab} \frac{(W^T X)_{ab}}{(W^T W H)_{ab}}, W_{ab} = W_{ab} \frac{(X H^T)_{ab}}{(X H H^T)_{ab}}$



$$W_{ia} = W_{ia} \frac{(X_1 H_1^T + X_2 H_2^T + X_3 H_3^T)_{ia}}{(W(H_1 H_1^T + H_2 H_2^T + H_3 H_3^T))_{ia}},$$

$$(H_I)_{a\mu} = (H_I)_{a\mu} \frac{(W^T X_I)_{a\mu}}{(W^T W H_I)_{a\mu}}, \quad I = 1, 2, 3.$$

Joint NMF

- $X_l = WH_l$
- Can cluster W's rows or H's columns to get clustering of the samples or of the features
- Here, look for md-modules
- Allow each feature to belong to more than one md-module
- look at each H_l, and for each of its k rows, include features with z score (using feature's mean and std) exceeding some threshold

$$z_{ij} = \frac{x_{ij} - \mu_i}{\sigma_i}$$

Joint NMF

- look at each H_l, and for each of the k vectors, include features with z score (using feature's mean and std) exceeding some threshold

$$z_{ij} = \frac{x_{ij} - \mu_i}{\sigma_i}$$

- Similarly, look at Columns of W and associate a patient with an md-module if its z-score exceeds some threshold

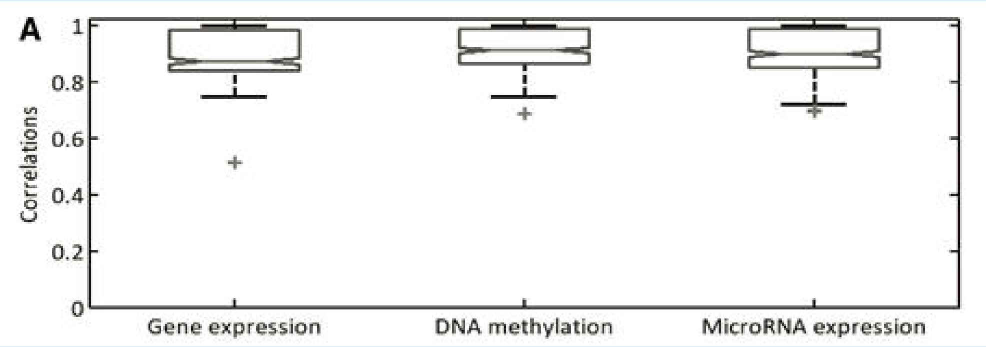
- The output is k md-modules, with features from each omic (and samples) associated with them

Joint NMF - Results

- Use ovarian cancer data from TCGA
- 385 samples
- 3 omics: gene expression, methylation and miRNA expression
- Negative values – double all features, one with positive and one with absolute value of negative
- K = 200 md-modules
- Cover ~3000 genes, ~2000 methylation sites, 270 miRNAs
- Average module sizes are ~240 genes, ~162 methylation sites and ~14 miRNAs (high overlap)

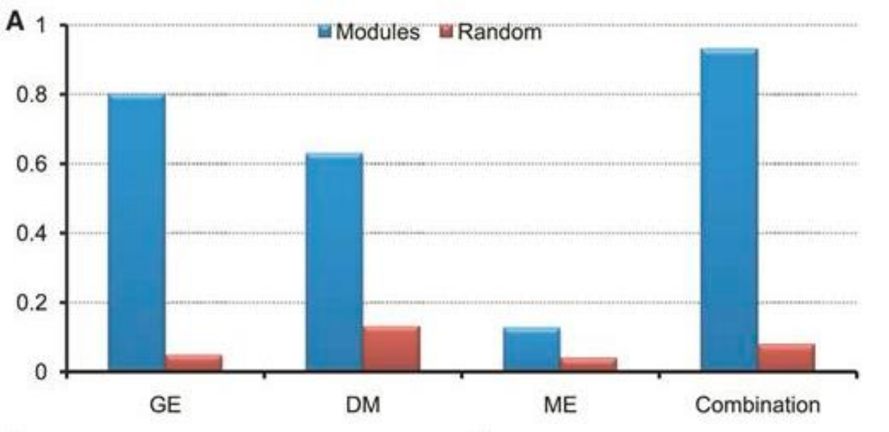
Joint NMF - Results

- Correlations of observed and reconstructed features
- The model doesn't lose "too much" information



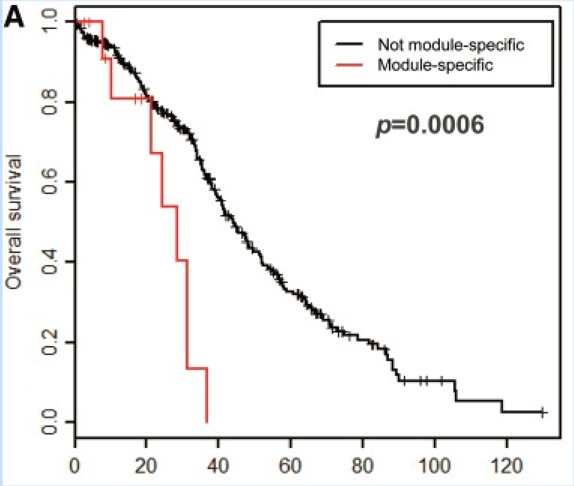
Joint NMF - Results

- For each module, look at:
  - Gene expression features in that module
  - Genes adjacent to methylation sites in the module
  - Genes regulated by miRNAs in the module
- Count md-modules with (FDR corrected) GO enrichment of at least one term
- Compare to random md-modules
- Combining all omics is more biologically meaningful



Joint NMF - Results

- 22 md-modules are enriched in genes with a known role in cancer (with p-value < 0.05). Note we would expect 10 by chance.
- 20 modules contain patients with significantly different age characteristics compared to patients not in the module
- (In plot: survival analysis for patients in module 166 compared to other patients. Didn't mention how many modules have different survival).



Joint NMF - Recap

- Low dimension + non negativity constraint
- $X_l = WH_l$
- Optimized using multiplicative update rules
- Look for md-modules: sets of features from all omics that largely determine the observed data
- Md-modules calculated from the factorization with z-score
- Run on TCGA ovarian cancer data
- Looking at all omics gives higher enrichment in md-modules compared to each omic alone

Outline

- Introduction
- Cluster of Clusters (COCA)
- iCluster
- Nonnegative Matrix Factorization (NMF)
- Similarity Network Fusion (SNF)
- Multiple Kernel Learning (MKL)

Similarity Network Fusion

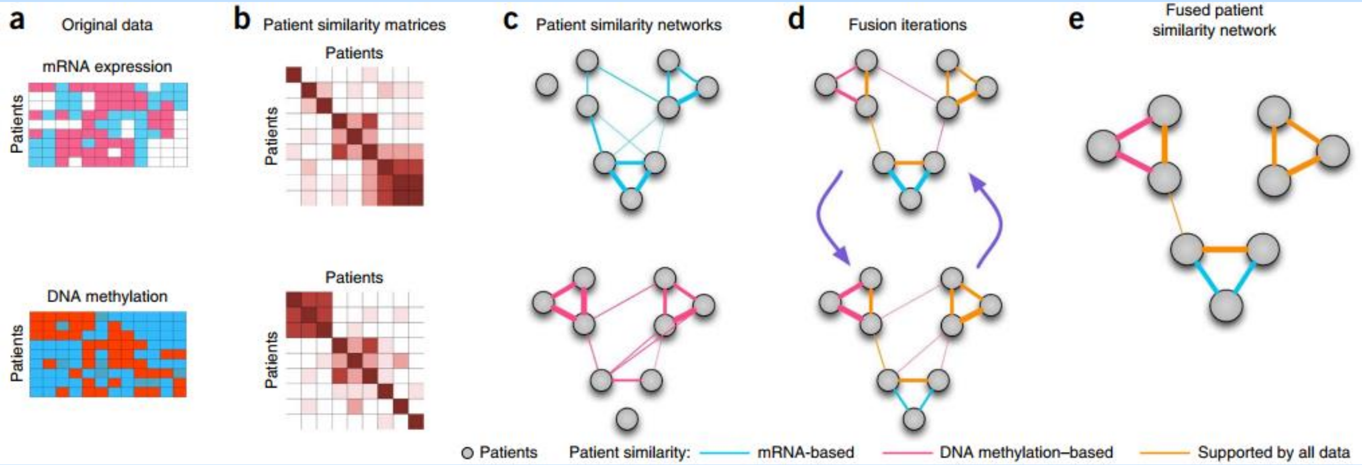
- Bo Wang, …, Anna Goldenberg (Nature Methods, 2014)
  - University of Toronto
- Number of features >> number of samples
- Dimension reduction methods' complexity depends on the number of features
- Formulations with non-convex / no closed form solution objective functions, so have to try many different initialization points

Similarity Network Fusion

- Idea: cluster based only on patients' similarity
- Aside from similarity computation, complexity is a function of the number of patients
- Less sensitivity to feature selection in practice
- More difficult to give interpretation to features as part of the model
- (Can still do analysis once we have the clusters, e.g. differentially expressed genes)

Similarity Network Fusion

- Construct similarity network for each omic
- Nodes are patients, edges' weights are similarity of patients in omic
- Iteratively update the weights of the networks, bringing the networks closer
- Obtain fused network



Similarity Network Fusion

- Initialization:

$$\mathbf{W}(i,j) = \exp\left(-\frac{\rho^2(x_i, x_j)}{\mu\varepsilon_{i,j}}\right)$$

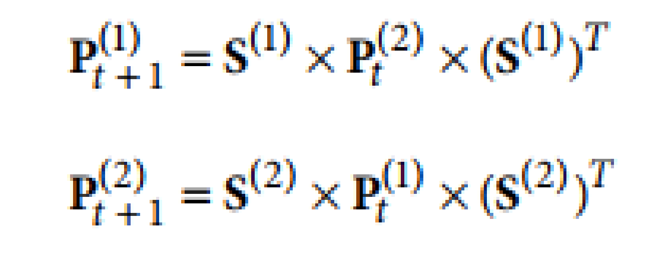
$$\mathbf{P}(i,j) = \begin{cases} \dfrac{\mathbf{W}(i,j)}{2\Sigma_{k \neq i}\mathbf{W}(i,k)}, j \neq i \\ 1/2, j = i \end{cases}$$

$$\mathbf{S}(i,j) = \begin{cases} \dfrac{\mathbf{W}(i,j)}{\Sigma_{k \in N_i}\mathbf{W}(i,k)}, & j \in N_i \\ 0 & \text{otherwise} \end{cases}$$

- $\epsilon_{i,j}$ measures the average distance of i and j to their neighbors, to correct for density
- N_i - k nearest neighbors of sample i, different k than the cluster number (~15-20 in practice)
- W – similarity, P – relative similarity, S – relative similarity within nearest neighbors
- P will be updated in each iteration

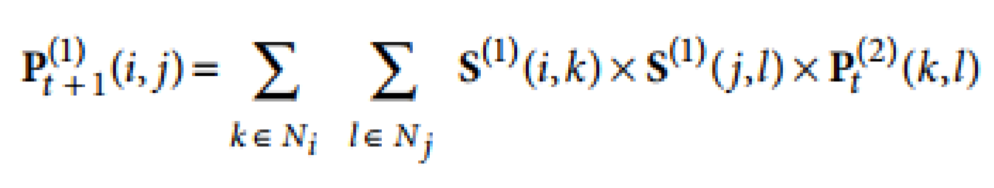
Similarity Network Fusion

- W – similarity, P – relative similarity, S – relative similarity within nearest neighbors
- (Assume for now we have two omics)
- P is updated in each iteration:



$$\mathbf{P}_{t+1}^{(1)} = \mathbf{S}^{(1)} \times \mathbf{P}_t^{(2)} \times (\mathbf{S}^{(1)})^T$$

$$\mathbf{P}_{t+1}^{(2)} = \mathbf{S}^{(2)} \times \mathbf{P}_t^{(1)} \times (\mathbf{S}^{(2)})^T$$



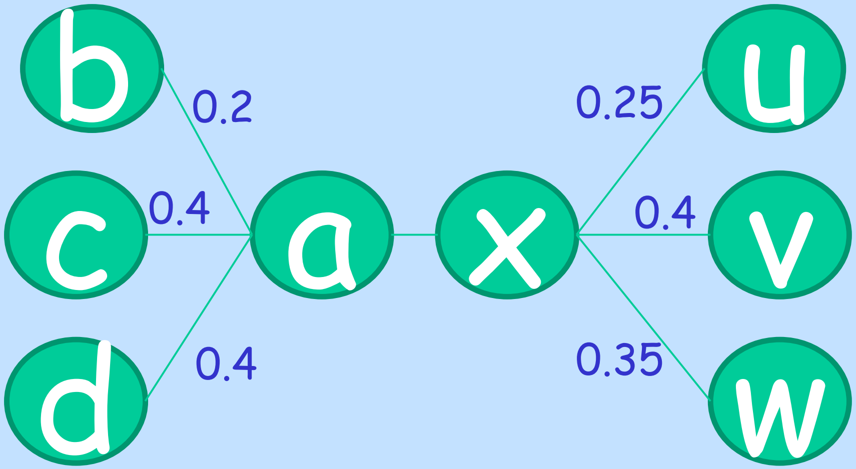
$$\mathbf{P}_{t+1}^{(1)}(i,j) = \sum_{k \in N_i} \sum_{l \in N_j} \mathbf{S}^{(1)}(i,k) \times \mathbf{S}^{(1)}(j,l) \times \mathbf{P}_t^{(2)}(k,l)$$

Similarity Network Fusion

$$\mathbf{P}^{(1)}_{t+1} = \mathbf{S}^{(1)} \times \mathbf{P}^{(2)}_{t} \times (\mathbf{S}^{(1)})^T$$

$$\mathbf{P}^{(2)}_{t+1} = \mathbf{S}^{(2)} \times \mathbf{P}^{(1)}_{t} \times (\mathbf{S}^{(2)})^T$$

$$\mathbf{P}^{(1)}_{t+1}(i,j) = \sum_{k \in N_i} \sum_{l \in N_j} \mathbf{S}^{(1)}(i,k) \times \mathbf{S}^{(1)}(j,l) \times \mathbf{P}^{(2)}_{t}(k,l)$$



i	j	$p_t^2(i,j)$
b	u	0.02
b	v	0.007
b	w	0.01
c	u	0.09
c	v	0.08
c	w	0.003
d	u	0.05
d	v	0.008
d	w	0.03

$$p_{t+1}^1 = 0.2 * 0.25 * 0.02 + 0.2 * 0.4 * 0.007 + \cdots$$

Similarity Network Fusion

$$\mathbf{P}^{(1)}_{t+1} = \mathbf{S}^{(1)} \times \mathbf{P}^{(2)}_{t} \times (\mathbf{S}^{(1)})^{T}$$

$$\mathbf{P}^{(2)}_{t+1} = \mathbf{S}^{(2)} \times \mathbf{P}^{(1)}_{t} \times (\mathbf{S}^{(2)})^{T}$$

$$\mathbf{P}^{(1)}_{t+1}(i,j) = \sum_{k \in N_i} \sum_{l \in N_j} \mathbf{S}^{(1)}(i,k) \times \mathbf{S}^{(1)}(j,l) \times \mathbf{P}^{(2)}_{t}(k,l)$$

- Intuition: weighted average of neighbor similarities
- Only neighbors – for robustness
- (P normalized and made symmetric at the end of every iteration)
- Converges!
- After a few iterations:

$$\mathbf{P}^{(c)} = \frac{\mathbf{P}^{(1)}_{t} + \mathbf{P}^{(2)}_{t}}{2} .$$

- For more than two omics:

$$\mathbf{P}^{(v)} = \mathbf{S}^{(v)} \times \left(\frac{\Sigma_{k \neq v} \mathbf{P}^{(k)}}{m-1} \right) \times (\mathbf{S}^{(v)})^{T}, v = 1,2,\cdots,m$$

- We now have one network

Spectral Clustering

- Cluster similarity matrix S

- Assume two clusters of ~ equal size
- If s_i, s_j belong to the same cluster, then S(i, j) >> 0
- Otherwise, S(i, j) << 0

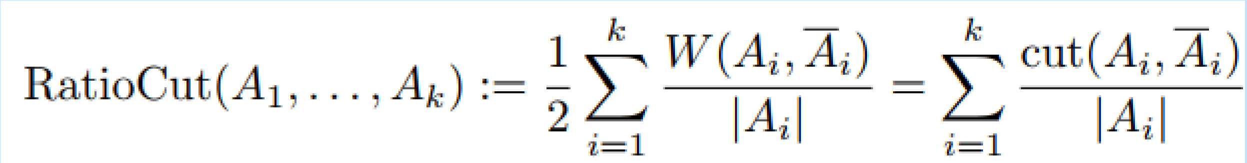
- $\Sigma_{i,j}\left(v_i - v_j\right)^2 S(i,j)$ is maximized when $v_i = \frac{1}{\sqrt{n}}$ for first cluster, $v_i = -\frac{1}{\sqrt{n}}$ for second cluster
- Instead of enforcing $v_i = \pm\frac{1}{\sqrt{n}}$, constraint $\left|\left|v\right|\right|_2 = 1, \left|\left|v\right|\right|_1 = 0$

Spectral Clustering

- Instead of enforcing $v_i = \pm\frac{1}{\sqrt{n}}$, constraint $||v||_2 = 1, ||v||_1 = 0$
- $min\Sigma_{i,j}\left(v_i - v_j\right)^2 S(i,j), s.t. v^T v = 1, ||v||_1 = 0$
- Define L = D-S, where D is the row sum diagonal matrix. L is the graph's Laplacian.
- $\mathrm{v^T L v} = \frac{1}{2}\Sigma_{i,j}\left(v_i - v_j\right)^2 S(i,j)$
- (Note the resemblance to PCA optimization problem –max $v^T X^T X v, v^T v = 1$)
- Solution is second smallest eigenvector of L
- (Second – because $||v||_1 = 0$, v orthogonal to $\vec{1}$)

Spectral Clustering

- $min\Sigma_{i,j}\left(v_i - v_j\right)^2 S(i,j), s.t. v^t v = 1, ||v||_1 = 0$
- Can now use v to cluster the samples, for example positive v values belong to one cluster and negative to the other
- Can be derived as an approximation to:



$$\mathrm{RatioCut}(A_1, \ldots, A_k) := \frac{1}{2}\sum_{i=1}^{k} \frac{W(A_i, \overline{A}_i)}{|A_i|} = \sum_{i=1}^{k} \frac{\mathrm{cut}(A_i, \overline{A}_i)}{|A_i|}$$

- For more than two clusters, find 2,…,k smallest eigenvectors of L
- The problem is solved by clustering V's rows (using k-means for example)

Similarity Network Fusion

- After a few iteration: $$\mathbf{P}^{(c)} = \frac{\mathbf{P}_t^{(1)} + \mathbf{P}_t^{(2)}}{2}.$$
- Cluster the network using spectral clustering (slightly different variation)
- Can use the network for other tasks
- Reminder: cox proportional hazards model
- Can use the network for regularization while learning the cox model's parameters such that similar patients will have similar prognoses

SNF - Results

- Use gene expression, methylation and micro RNA
- 5 different cancer types: Glioblastoma Multiforme (aggressive brain cancer), breast, kidney, lung and colon
- Each cancer type has 90-215 patients
- (Different heuristics to choose k, $\mu = 0.5$ empirically)
- Compare prognosis using log rank for > 2 groups

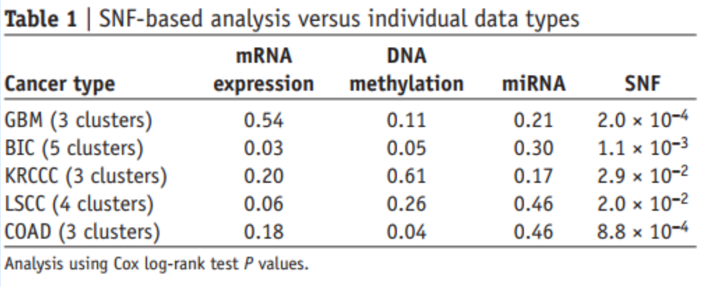
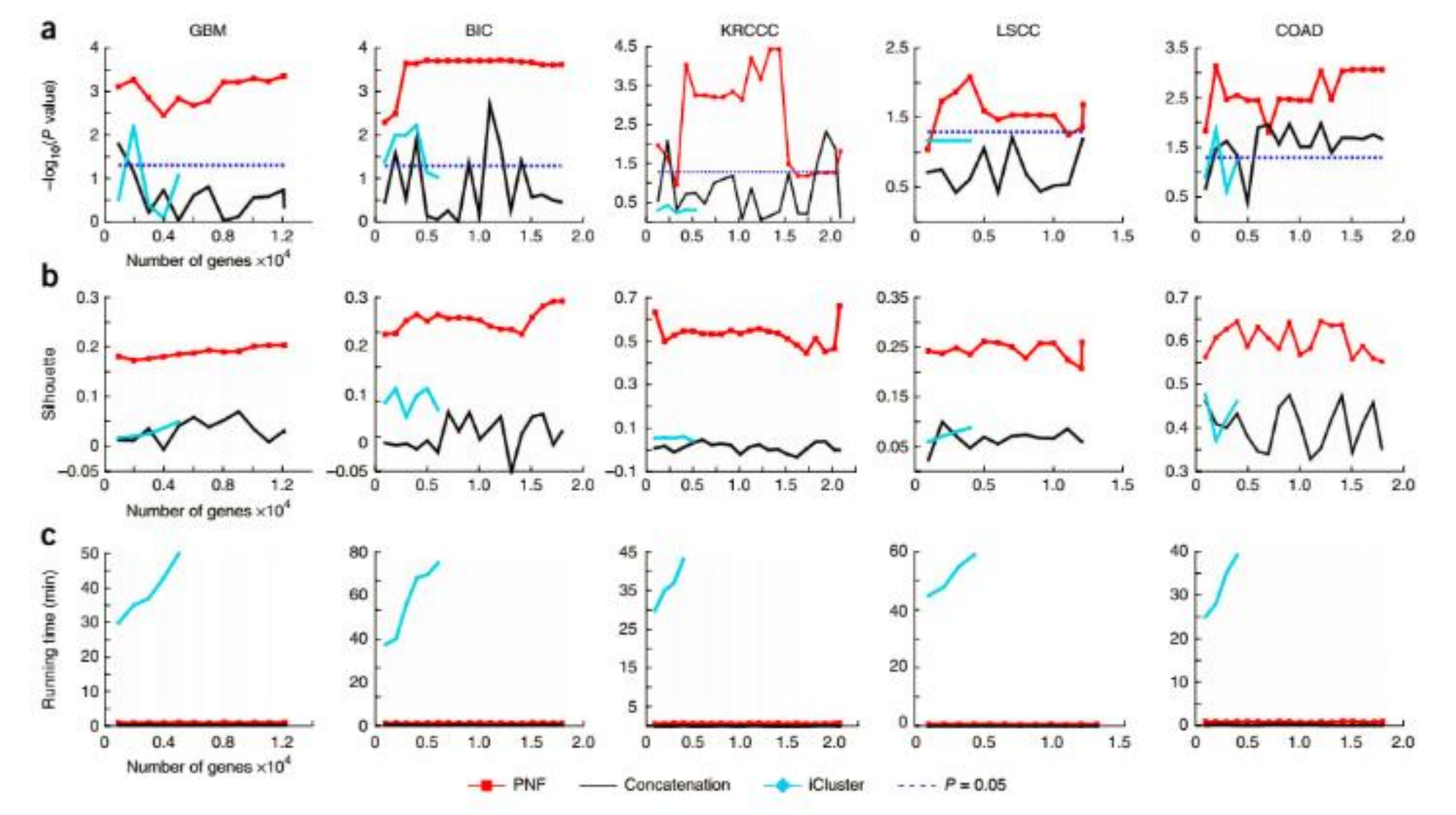


Table 1 | SNF-based analysis versus individual data types

Cancer type	mRNA expression	DNA methylation	miRNA	SNF
GBM (3 clusters)	0.54	0.11	0.21	2.0×10^{-4}
BIC (5 clusters)	0.03	0.05	0.30	1.1×10^{-3}
KRCCC (3 clusters)	0.20	0.61	0.17	2.9×10^{-2}
LSCC (4 clusters)	0.06	0.26	0.46	2.0×10^{-2}
COAD (3 clusters)	0.18	0.04	0.46	8.8×10^{-4}

Analysis using Cox log-rank test *P* values.

SNF - Results


a
b
c
GBM
BIC
KRCCC
LSCC
COAD
$-\log_{10}(P \text{ value})$
Silhouette
Running time (min)
Number of genes ×10^4
0
0.4
0.8
1.2
0.5
1.0
1.5
2.0
PNF
Concatenation
iCluster
$P = 0.05$

SNF - Recap

- Similarity based
- Patient similarity network per omic, followed by iteratively bringing the networks close to one another, until we have one network
- Cluster the network with spectral clustering
- Additional usages for the network
- Run on TCGA data from multiple tissues, and compare to iCluster, single omic and concatenation

Outline

- Introduction
- Cluster of Clusters (COCA)
- iCluster
- Nonnegative Matrix Factorization (NMF)
- Similarity Network Fusion (SNF)
- Multiple Kernel Learning (MKL)

Multiple Kernel Learning

- Speicher and Pfeifer (2015, bioinformatics)
  - Max Planck Institute for Informatics
- Similarity based method
- Multiple Kernel Learning – the general idea of using several kernels
- Have been used on single data type, mainly in supervised context but also in unsupervised
- Idea: use different kernels for different omics, together with multiple kernel dimension reduction algorithms

Graph Embedding

- x_i are input vectors, W is input similarity graph, D is diagonal constraint matrix

$$\underset{v}{\text{minimize}} \sum_{i,j=1}^{N} \left\| v^T x_i - v^T x_j \right\|^2 w_{ij}$$

$$\text{subject to} \quad \sum_{i=1}^{N} \left\| v^T x_i \right\|^2 d_{ii} = \text{const.}$$

- Look for v that projects x vectors to a line such that similarities are kept
- (D matrix is mainly in order to avoid the trivial solution)
- (Difference from spectral clustering?)

Graph Embedding

$$\underset{v}{\text{minimize}} \sum_{i,j=1}^{N} \left|\left| v^T x_i - v^T x_j \right|\right|^2 w_{ij}$$

$$\text{subject to} \quad \sum_{i=1}^{N} \left|\left| v^T x_i \right|\right|^2 d_{ii} = \text{const.}$$

- Can be shown that optimal v is necessarily in the span of the vectors:

$$v = \Sigma_{n=1}^{N} \alpha_n x_n$$

- (Kernel trick reminder: $K(x, y) = < \phi(x), \phi(y) >$)
- $v^t x_i - v^t x_j = \Sigma_{n=1}^{N} \alpha_n x_n^t x_i - \Sigma_{n=1}^{N} \alpha_n x_n^t x_j$ $= \Sigma_{n=1}^{N} \alpha_n K(n, i) - \Sigma_{n=1}^{N} \alpha_n K(n, j)$

Multiple Kernel Learning

- $v^t x_i - v^t x_j = \Sigma_{n=1}^N \alpha_n x_n^t x_i - \Sigma_{n=1}^N \alpha_n x_n^t x_j$
  $= \Sigma_{n=1}^N \alpha_n K(n,i) - \Sigma_{n=1}^N \alpha_n K(n,j)$
- We want different kernels for different omics

- $\Sigma_m \beta_m K_m,\ \beta_m \geq 0$ is also a kernel (ex)
- $K(n,i)$=$\Sigma_m \beta_m K_m(n,i)$
- $\Sigma_{n=1}^N \alpha_n \Sigma_m \beta_m K_m(n,i) = \alpha^t K^i \beta$

$$\mathscr{K}^i = \begin{pmatrix} K_1(1,i) & \cdots & K_M(1,i) \\ \vdots & \ddots & \vdots \\ K_1(N,i) & \cdots & K_M(N,i) \end{pmatrix} \in \mathbb{R}^{N \times M}$$

Multiple Kernel Learning

- From: $\underset{v}{\text{minimize}} \sum_{i,j=1}^{N} \left\| v^T x_i - v^T x_j \right\|^2 w_{ij} \quad \text{subject to} \quad \sum_{i=1}^{N} \left\| v^T x_i \right\|^2 d_{ii} = \text{const.}$

- To:

$$\underset{\boldsymbol{\alpha},\boldsymbol{\beta}}{\text{minimize}} \quad \sum_{i,j=1}^{N} \left\| \boldsymbol{\alpha}^T \mathscr{K}^i \boldsymbol{\beta} - \boldsymbol{\alpha}^T \mathscr{K}^j \boldsymbol{\beta} \right\|^2 w_{ij} \qquad \text{subject to} \quad \sum_{i,j=1}^{N} \left\| \boldsymbol{\alpha}^T \mathscr{K}^i \boldsymbol{\beta} \right\|^2 d_{ij} = \text{const.}$$

- With constraints:

$\beta_m \geq 0, m = 1, 2, \ldots, M.$ $\qquad \|\boldsymbol{\beta}\|_1 = 1$

$$\mathscr{K}^i = \begin{pmatrix} K_1(1,i) & \cdots & K_M(1,i) \\ \vdots & \ddots & \vdots \\ K_1(N,i) & \cdots & K_M(N,i) \end{pmatrix} \in \mathbb{R}^{N \times M}$$

Multiple Kernel Learning

$$\underset{\boldsymbol{\alpha},\boldsymbol{\beta}}{\text{minimize}} \quad \sum_{i,j=1}^{N} \left\| \boldsymbol{\alpha}^T \mathscr{K}^i \boldsymbol{\beta} - \boldsymbol{\alpha}^T \mathscr{K}^j \boldsymbol{\beta} \right\|^2 w_{ij} \qquad \text{subject to} \quad \sum_{i,j=1}^{N} \left\| \boldsymbol{\alpha}^T \mathscr{K}^i \boldsymbol{\beta} \right\|^2 d_{ij} = \text{const.}$$

$$\beta_m \geq 0,\ m = 1, 2, \ldots, M. \qquad \|\boldsymbol{\beta}\|_1 = 1$$

$$\mathscr{K}^i = \begin{pmatrix} K_1(1,i) & \cdots & K_M(1,i) \\ \vdots & \ddots & \vdots \\ K_1(N,i) & \cdots & K_M(N,i) \end{pmatrix} \in \mathbb{R}^{N \times M}$$

- W and D:

$$w_{ij} = \begin{cases} 1, & \text{if } i \in \mathscr{N}_k(j) \vee j \in \mathscr{N}_k(i) \\ 0, & \text{else} \end{cases} \qquad d_{ij} = \begin{cases} \sum_{n=1}^{N} w_{in}, & \text{if } i = j \\ 0, & \text{else.} \end{cases}$$

Multiple Kernel Learning

$$\underset{\boldsymbol{\alpha},\boldsymbol{\beta}}{\text{minimize}} \quad \sum_{i,j=1}^{N} \left\| \boldsymbol{\alpha}^T \mathscr{K}^i \boldsymbol{\beta} - \boldsymbol{\alpha}^T \mathscr{K}^j \boldsymbol{\beta} \right\|^2 w_{ij}$$

$$\text{subject to} \quad \sum_{i,j=1}^{N} \left\| \boldsymbol{\alpha}^T \mathscr{K}^i \boldsymbol{\beta} \right\|^2 d_{ij} = \text{const.}$$

- α projects points to a single dimension
- Use matrix A instead to project to a different dimension
- Dimension not necessarily equal to the number of clusters

Multiple Kernel Learning

$$\underset{\boldsymbol{\alpha},\boldsymbol{\beta}}{\text{minimize}} \quad \sum_{i,j=1}^{N} \left\| \boldsymbol{\alpha}^T \mathscr{K}^i \boldsymbol{\beta} - \boldsymbol{\alpha}^T \mathscr{K}^j \boldsymbol{\beta} \right\|^2 w_{ij}$$

$$\text{subject to} \quad \sum_{i,j=1}^{N} \left\| \boldsymbol{\alpha}^T \mathscr{K}^i \boldsymbol{\beta} \right\|^2 d_{ij} = \text{const.}$$

- Optimize A and β iteratively in an alternating manner
- β is optimized using semidefinite programming

$$\begin{array}{lll} \displaystyle\min_{x^1,\ldots,x^n \in \mathbb{R}^n} & \displaystyle\sum_{i,j\in[n]} c_{i,j}(x^i \cdot x^j) & \\ \text{subject to} & \displaystyle\sum_{i,j\in[n]} a_{i,j,k}(x^i \cdot x^j) \leq b_k & \forall k. \end{array}$$

- A is optimized by solving a generalized eigenvalue problem
- Cluster the data projection $\mathrm{A}^{\mathrm{t}}\mathrm{K}^{\mathrm{i}}\beta$ using k-means

MKL - Results

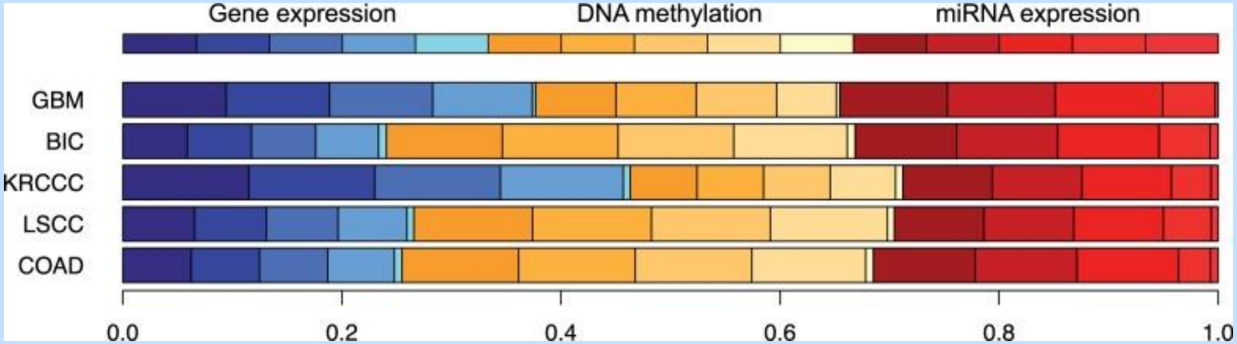
- Run on GBM, breast, lung, kidney and colon cancer (SNF dataset, ~90-215 patients per subtype)
- Use either 1 or 5 kernels per dataset:

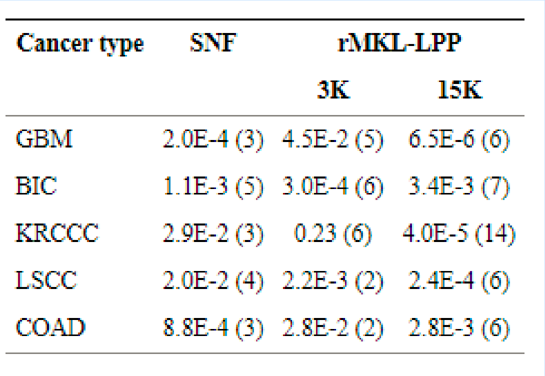
$$K(\mathbf{x},\mathbf{x}') = \exp(-\gamma\|\mathbf{x}-\mathbf{x}'\|^2)$$

- $\gamma = \frac{1}{2d^2}, \gamma_n = c_n\gamma, c_n \in \{10^{-6}, 10^{-3}, 1, 10^3, 10^6\}$
- Fix the dimension to 5, and choose k using silhouette score
- β values measure the effect of each kernel

MKL - Results

- β values measure the effect of each kernel
- Survival analysis comparing to SNF

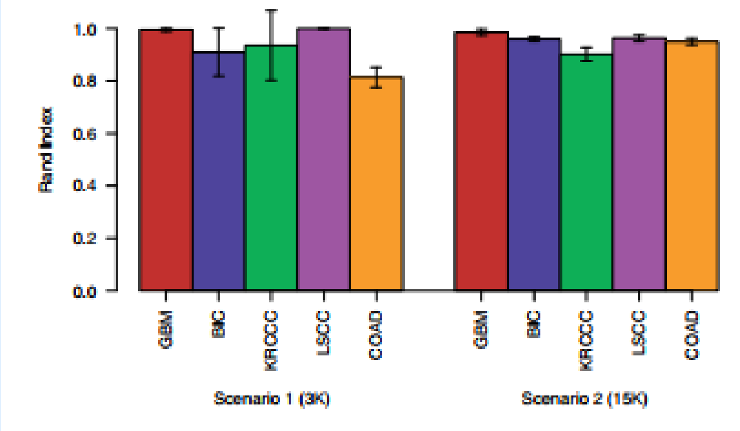


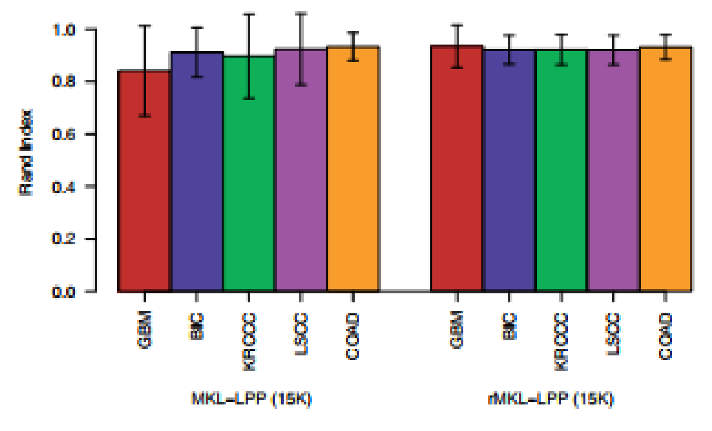


Cancer type	SNF	rMKL-LPP	
		3K	15K
GBM	2.0E-4 (3)	4.5E-2 (5)	6.5E-6 (6)
BIC	1.1E-3 (5)	3.0E-4 (6)	3.4E-3 (7)
KRCCC	2.9E-2 (3)	0.23 (6)	4.0E-5 (14)
LSCC	2.0E-2 (4)	2.2E-3 (2)	2.4E-4 (6)
COAD	8.8E-4 (3)	2.8E-2 (2)	2.8E-3 (6)

MKL - Results

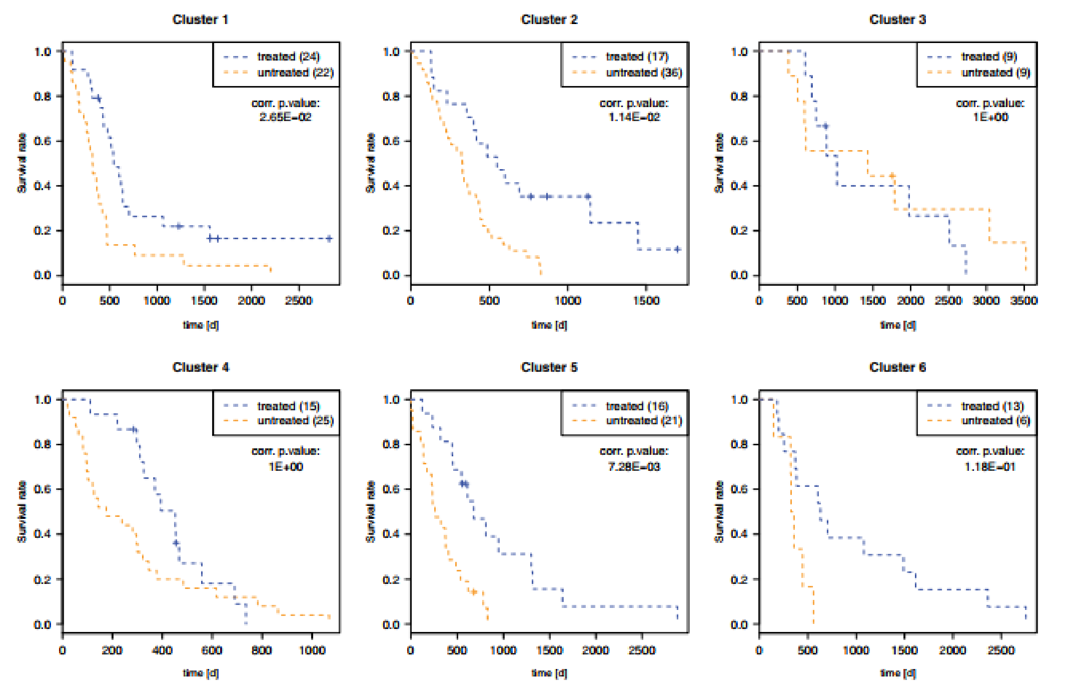
- Method's robustness
- Leave-one-out clustering: each patient is left out, the algorithm is run, and then the patient is added by projecting it and adding to the cluster with nearest mean
- Left – 3K vs 15k. Right – β sums to 1 constraint





MKL - Results

- Survival analysis does not consider the treatment given
- Response to Temozolomide (chemotherapy drug used for brain cancers) within different clusters



MKL - Recap

- Similarity based
- Graph embedding: dimension reduction such that neighbors in the original dimension remain close in the low dimension
- Use the kernel trick + different kernel(s) for each omic
- Compare prognosis to SNF and show the effect of multiple kernels on robustness

Summary

- Omic
- Multi omics data

- In this talk – methods that apply to numerical omics
- COCA – late integration

- Shared subspace models:
  - iCluster – probabilistic linear model
  - NMF – factorization with non negativity constraints

Summary

- Shared subspace models:
  - iCluster – probabilistic linear model
  - NMF – factorization with non negativity constraints
- Similarity based models:
  - Similarity network fusion – creating a unified similarity network
  - Multiple kernel learning – using different kernels for each omic
- Complexity and non-numerical omics vs. analysis of the feature within the model
- (Do different omics share the same underlying clustering?)

FIN