Analysis of Biological Networks: Pattern Discovery and Module Detection

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Outline

1. Biological Networks
   - Definition, problems, practical implications
2. Prior Work
   - Non-orthogonal decomposition of binary matrices
   - Module detection through analysis of microarray data
3. Current Work
   - Mining biological networks for frequent molecular interaction patterns
   - Alignment of protein interaction networks
4. Ongoing and Future Work
   - Module detection based on phylogeny profiles
   - Constructing reference module maps

Biological Networks

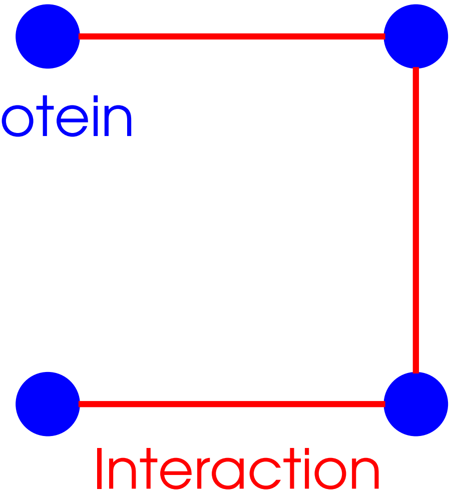
- Interactions between biomolecules that drive cellular processes
  - Genes, proteins, enzymes, chemical compounds
  - Mass & energy generation, information transfer
  - Coarser level than sequences in life's complexity pyramid

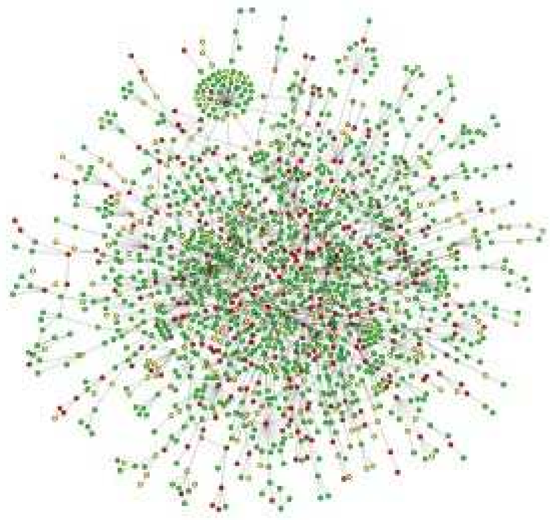
- Experimental/induced data in various forms
  - Protein interaction networks
  - Gene regulatory networks
  - Metabolic & signaling pathways

- What do we gain from analysis of cellular networks?
  - Modular analysis of cellular processes
  - Understanding evolutionary relationships at a higher level
  - Assigning functions to proteins through interaction information
  - Intelligent drug design: block protein, preserve pathway

Protein Interaction Networks

- Interacting proteins can be discovered experimentally
  - Two-hybrid
  - Mass spectrometry
  - Phage display



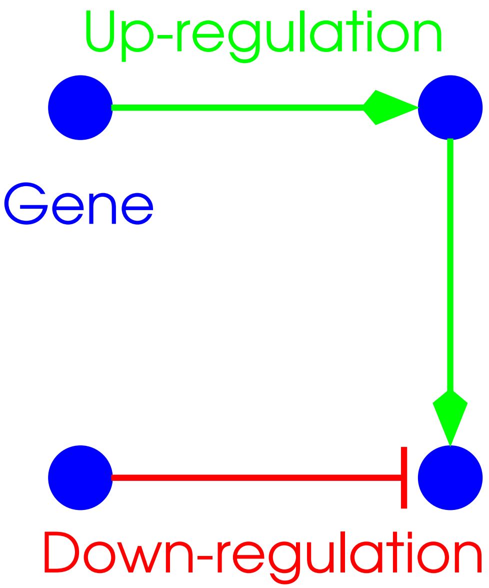


S. Cerevisae protein interaction network

Source: Jeong et al. Nature 411: 41-42, 2001.

Gene Regulatory Networks

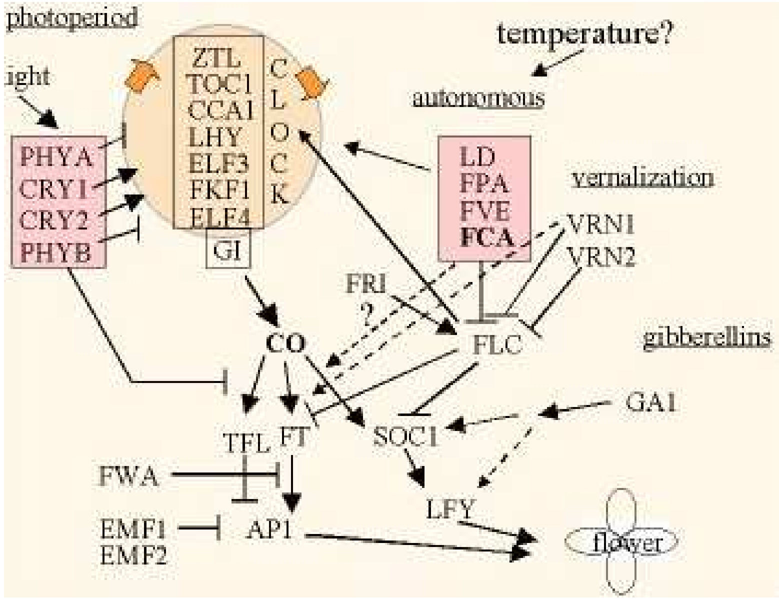
- Genes regulate each others' expression
  - A simple model: Boolean networks
  - Can be derived from gene expression data



Up-regulation

Gene

Down-regulation

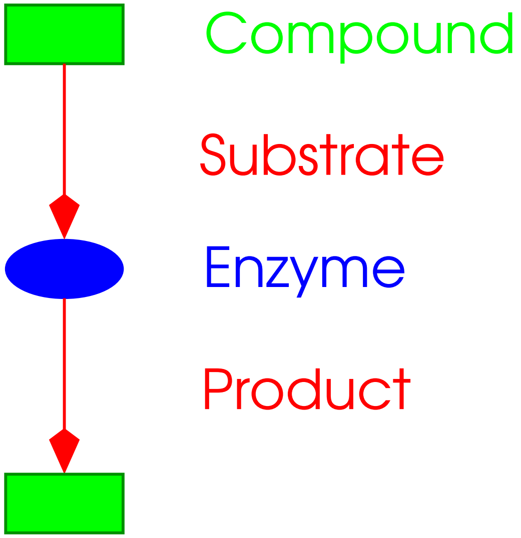


Genetic network that controls flowering time in *A. Thaliania*

Source: Blazquez et al. EMBO Reports 2: 1078-1082, 2001

Metabolic Pathways

- Chains of reactions that perform a particular metabolic function
  - Reactions are linked to each other through substrate-product relationships
  - Directed hypergraph/ graph models

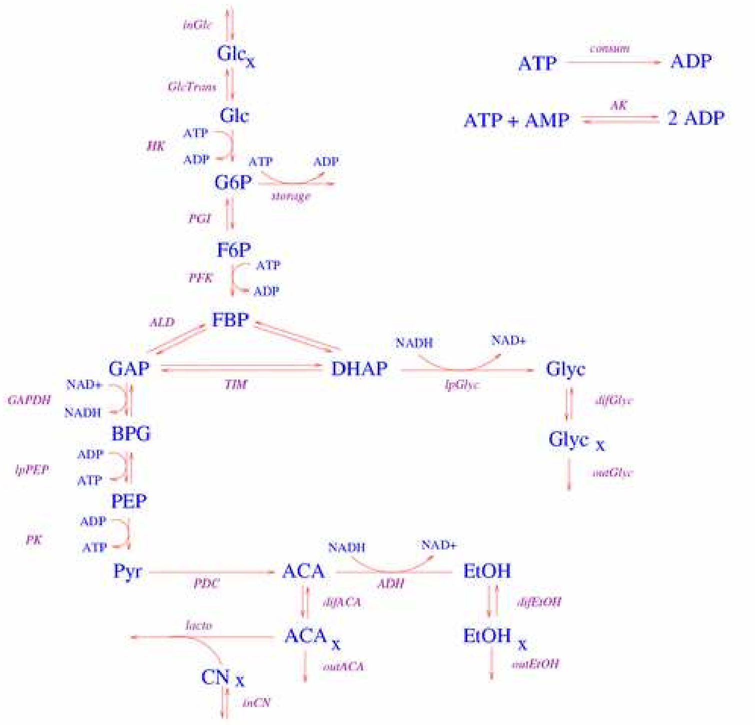


Compound

Substrate

Enzyme

Product



Glycolysis pathway in *S. Cerevisae*

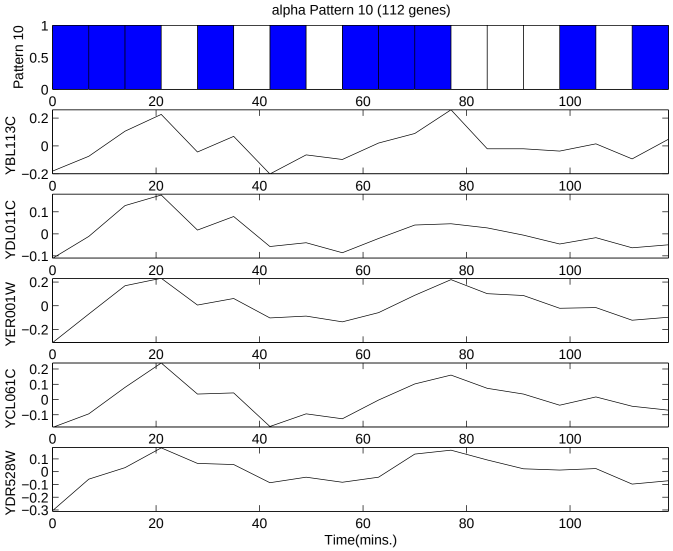
Source: Hynne et al. Biophysical Chemistry, 94, 121-163, 2001.

Prior Work

- Non-orthogonal decomposition of binary matrices
  - Find a compact set of vectors that represent the entire matrix
  - Recursive decomposition through rank-one approximations
  - Fast (linear-time) iterative heuristics for computing approximations

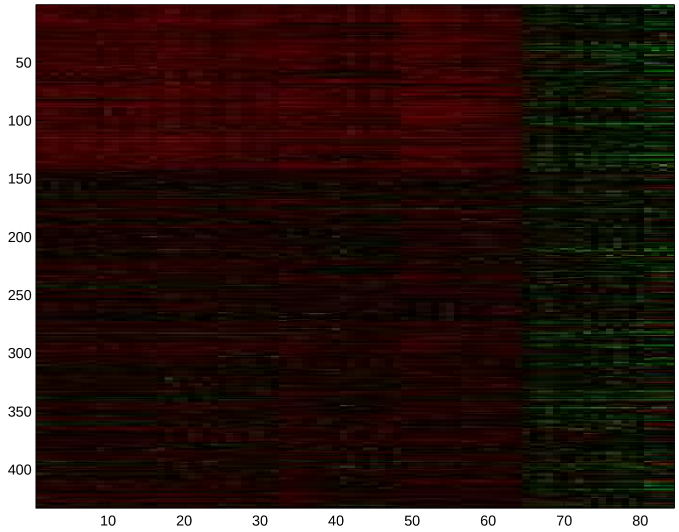
Analysis of gene expression data

Patterns of regulation



"Algorithms for bounded-error correlation of high dimensional data in microarray experiments" Koyutürk, Grama, Szpankowski: *CSB'03.*

Biclustering



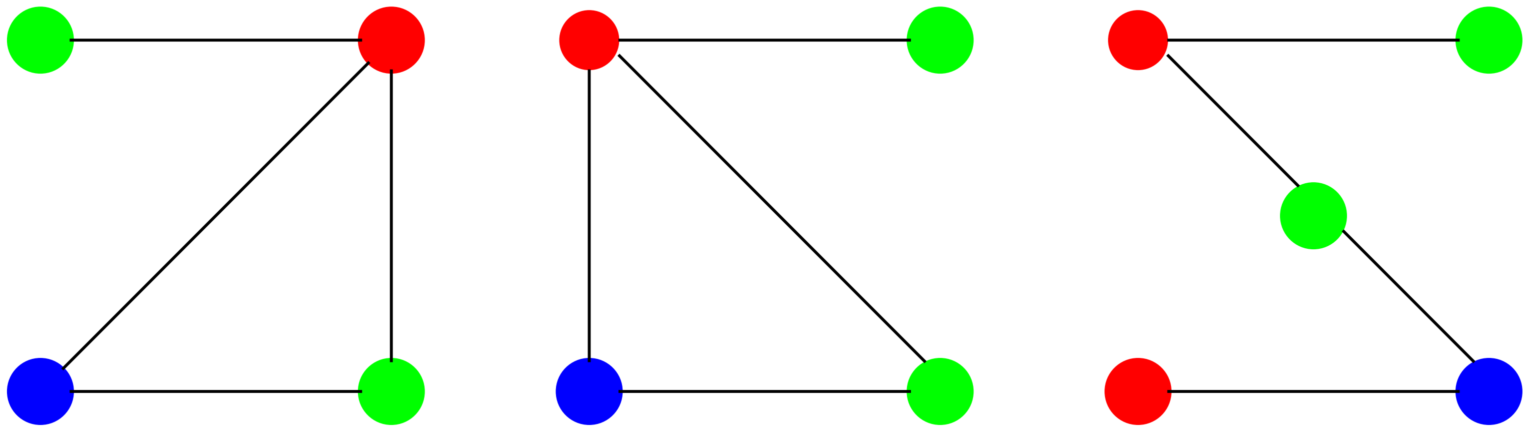
"Biclustering gene-feature matrices for statistically significant dense patterns" Koyutürk, Grama, Szpankowski: *CSB'04.*

Analysis of Biological Networks

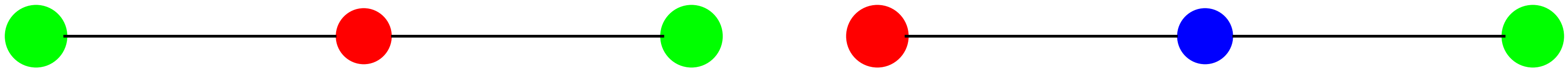
- Evolution thinks modular
  - Selective pressure on preserving interactions
  - Functional modules, protein complexes are highly conserved

- Computational methods for discovery and analysis modules and complexes
  - Graph clustering: Functionally related entities are densely connected
  - Graph mining: Common topological motifs, frequent interaction patterns reveal modularity
  - Graph alignment: Conservation/divergence of modules and pathways
  - Module maps: Canonical pathways across species
  - Phylogenetic analysis: Genes/proteins that belong to a common module are likely to have co-evolved

Graph Mining



Graph database



Subgraphs with frequency 3

Extending Frequent Itemset Mining to Graph Mining

- Given a set of transactions, find sets of items that are frequent in these transactions

- Extensively studied in data mining literature

- Algorithms exploit downward closure property
  - A set is frequent only if all of its subsets are frequent
  - Generate itemsets from small to large, pruning supersets of infrequent sets

- Can be generalized to mining graphs
  - transaction → graph
  - item → node, edge
  - itemset → subgraph

- However, the graph mining problem is consderably more difficult

Graph Mining Challenges

- Subgraph Isomorphism
  - For counting frequencies, it is necessary to check whether a given graph is a subgraph of another one
  - NP-complete

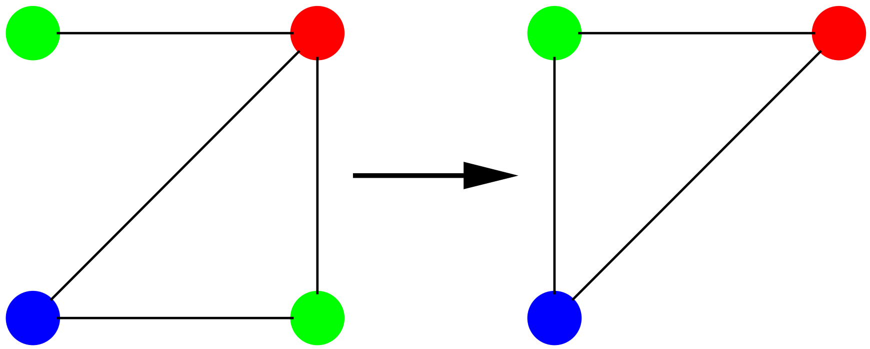
- Canonical labeling
  - To avoid redundancy while generating subgraphs, canonical labeling of graphs is necessary
  - Equivalent to subgraph isomorphism

- Connectivity
  - Patterns of interest are generally connected, so it is necessary to only generate connected subgraphs

- Existing algorithms mainly focus on minimizing redundancy and mining & extending simple substructures
  - AGM, FSG, gSpan, SPIM, CLOSEGRAPH

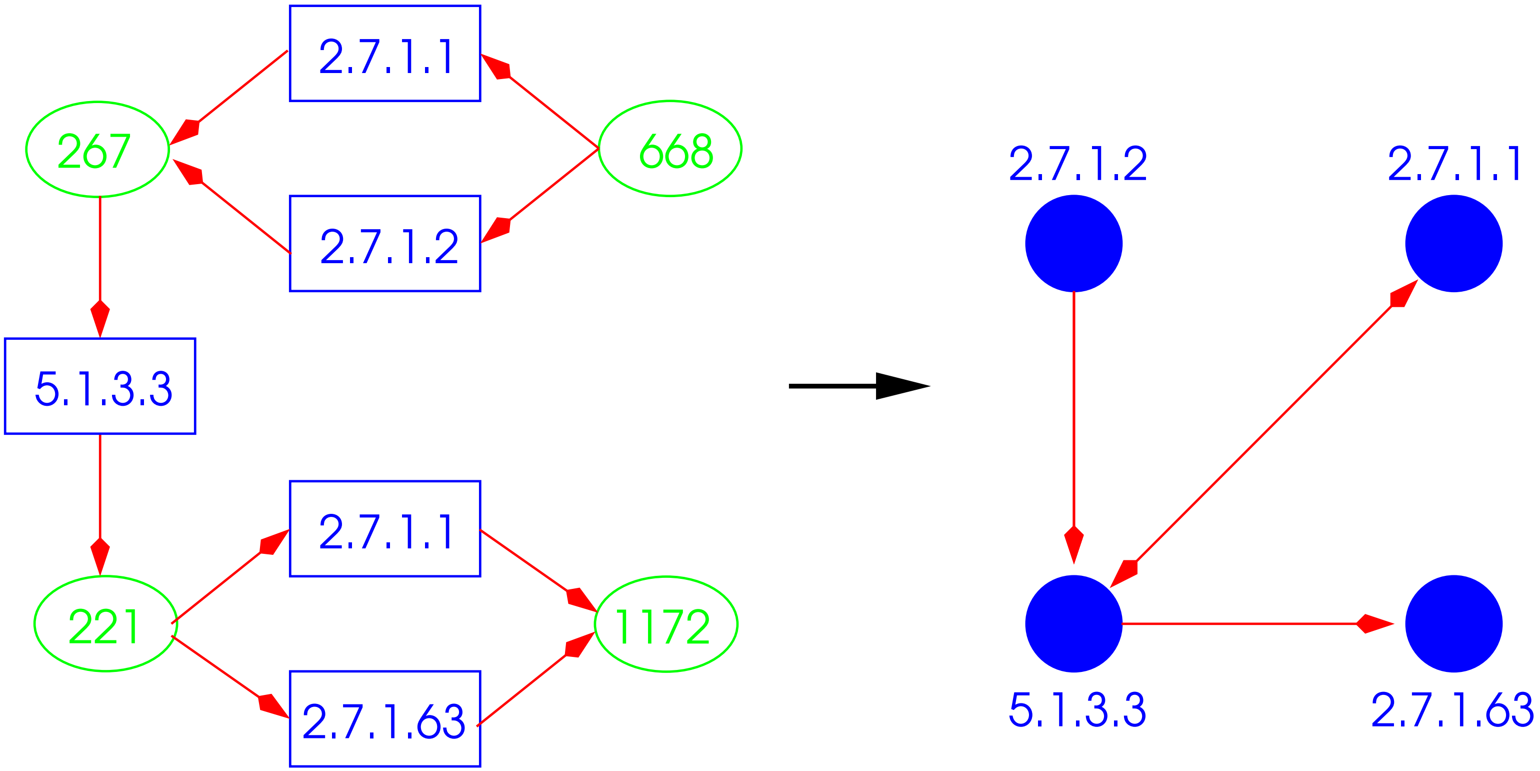
Uniquely-Labeled Graphs

- Contract nodes with identical label into a single node
- No subgraph isomorphism
  - Graphs are uniquely identified by their edge sets
- Frequent subgraphs are preserved ⇒ No information loss
  - Subgraphs that are frequent in general graphs are also frequent in their uniquely-labeled representation
- Discovered frequent subgraphs are still biologically interpretable!



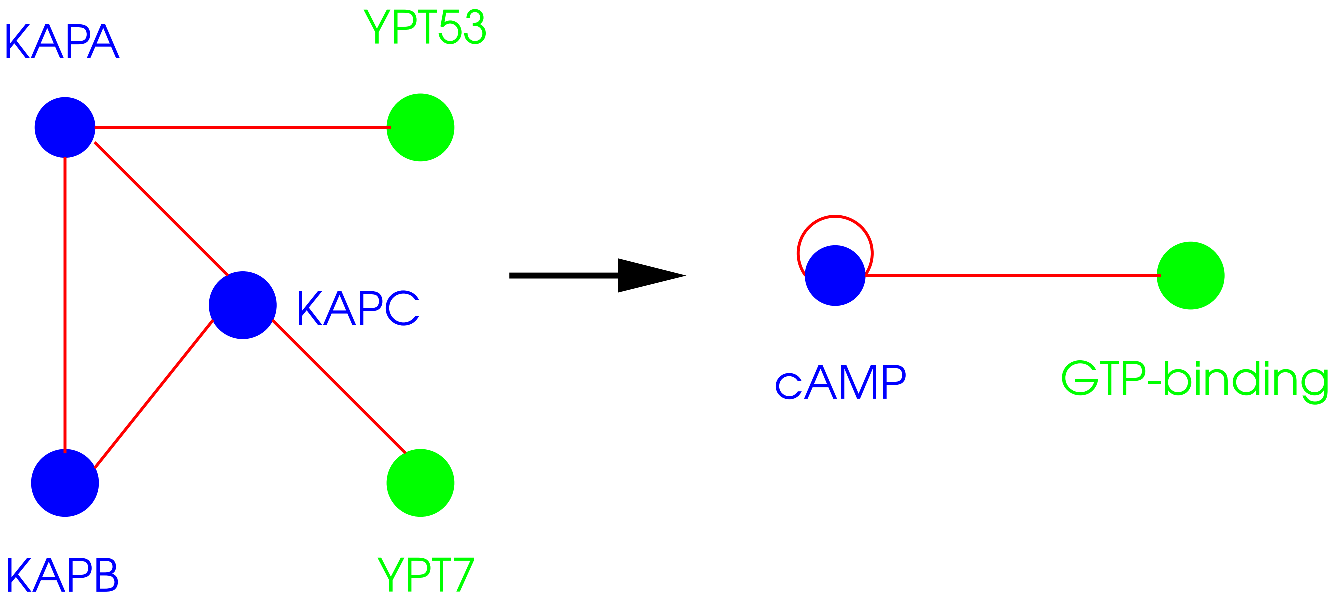
Node Contraction in Metabolic Pathways

- Uniquely-labeled directed graph model
  - Nodes represent enzymes
  - Global labeling by enzyme nomenclature (EC numbers)
  - A directed edge from one enzyme to the other implies that the second consumes a product of the first



Node Contraction in Protein Interaction Networks

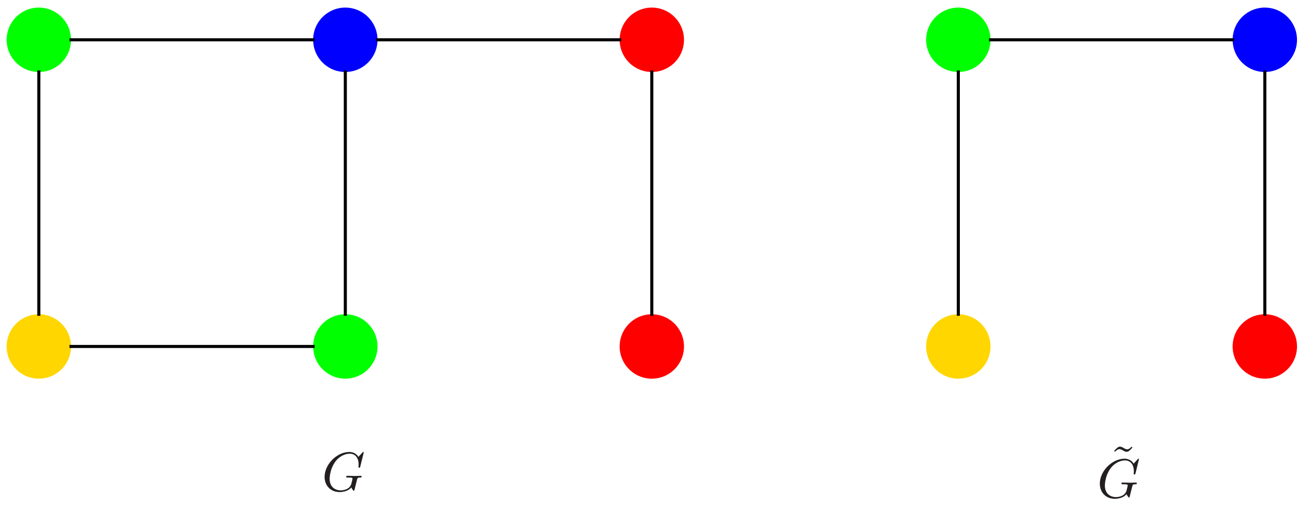
- Relating proteins in different organisms
  - Clustering: Orthologous proteins show sequence similarities
  - Phlyogenetic analysis: Allows multi-resolution analysis among distant species
  - Literature, ortholog databases
- Contraction
  - Interaction between proteins → interaction between protein families



Preservation of Subgraphs

Theorem: Let $\tilde{G}$ be the uniquely-labeled graph obtained by contracting the same-label nodes of graph G. Then, if S is a subgraph of G, $\tilde{S}$ is a subgraph of $\tilde{G}$.

Corollary: The uniquely-labeled representation of any frequent subgraph is frequent in the set of uniquely-labeled graphs.



Simplifying the Graph Mining Problem

Observation: A uniquely-labeled graph is uniquely determined by the set of its edges.

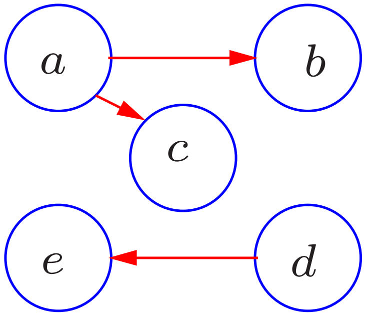
Maximal Frequent Subgraph Mining Problem

Given a set of labeled graphs $\{G_1, G_2, ..., G_m\}$, find all connected graphs S such that S is a subgraph of at least σm of the graphs (is frequent) and no supergraph of S is frequent (is maximal).

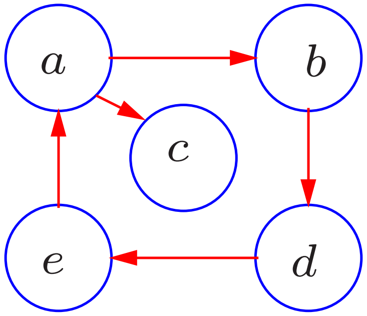
Maximal Frequent Edgeset Mining Problem

Given a set of edge transactions $\{E_1, E_2, ..., E_m\}$, find all connected edge sets F such that F is a subset of at least σm of the edge transactions (is frequent) and no superset of F is frequent (is maximal).

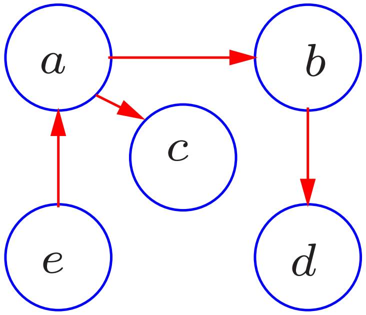
From Graphs to Edgesets



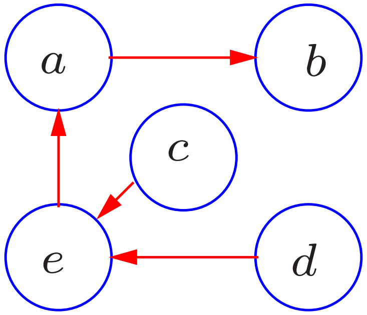
G_1



G_2



G_3



G_4

$\longrightarrow$

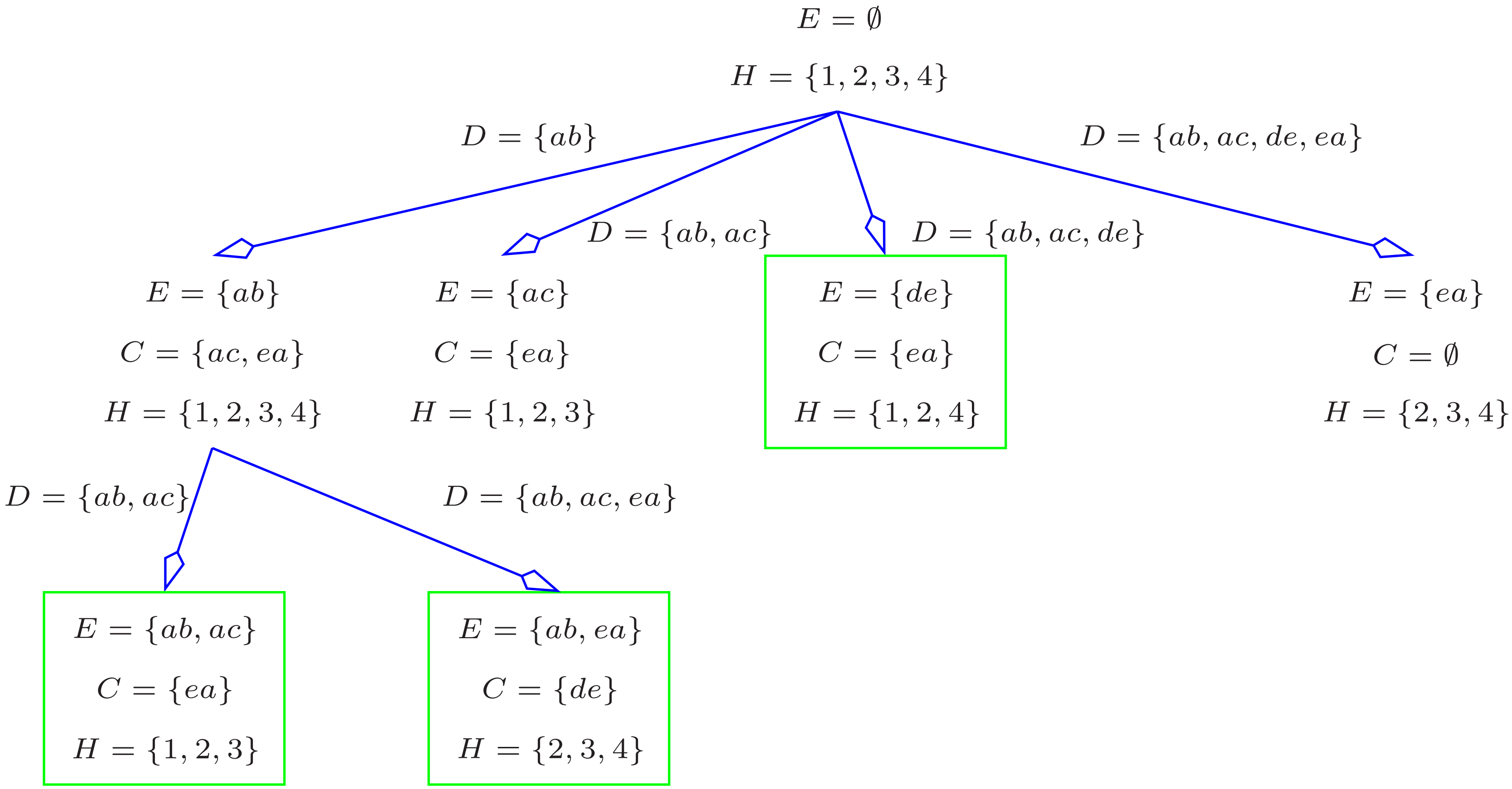
$F_1 = \{ab, ac, de\}$

$F_2 = \{ab, ac, bc, de, ea\}$

$F_3 = \{ab, ac, bc, ea\}$

$F_4 = \{ab, ce, de, ea\}$

MULE: Mining Uniquely Labeled Graphs



$E = \emptyset$

$H = \{1,2,3,4\}$

$D = \{ab\}$

$D = \{ab, ac\}$

$D = \{ab, ac, de\}$

$D = \{ab, ac, de, ea\}$

$E = \{ab\}$

$C = \{ac, ea\}$

$H = \{1,2,3,4\}$

$E = \{ac\}$

$C = \{ea\}$

$H = \{1,2,3\}$

$E = \{de\}$

$C = \{ea\}$

$H = \{1,2,4\}$

$E = \{ea\}$

$C = \emptyset$

$H = \{2,3,4\}$

$D = \{ab, ac\}$

$D = \{ab, ac, ea\}$

$E = \{ab, ac\}$

$C = \{ea\}$

$H = \{1,2,3\}$

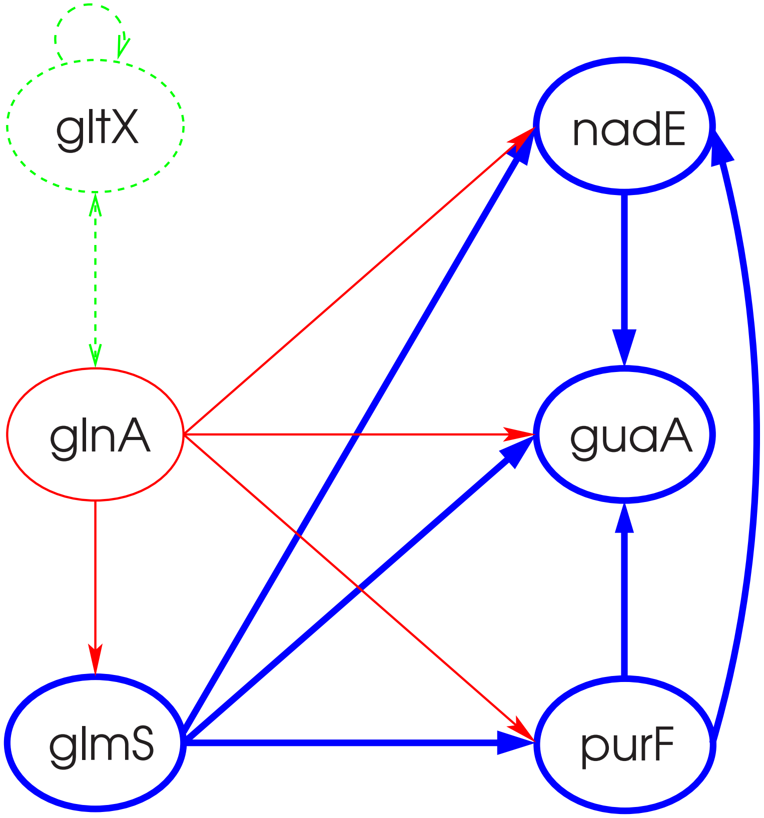
$E = \{ab, ea\}$

$C = \{de\}$

$H = \{2,3,4\}$

Frequent Sub-Pathways in KEGG

Glutamate metabolism (155 organisms)



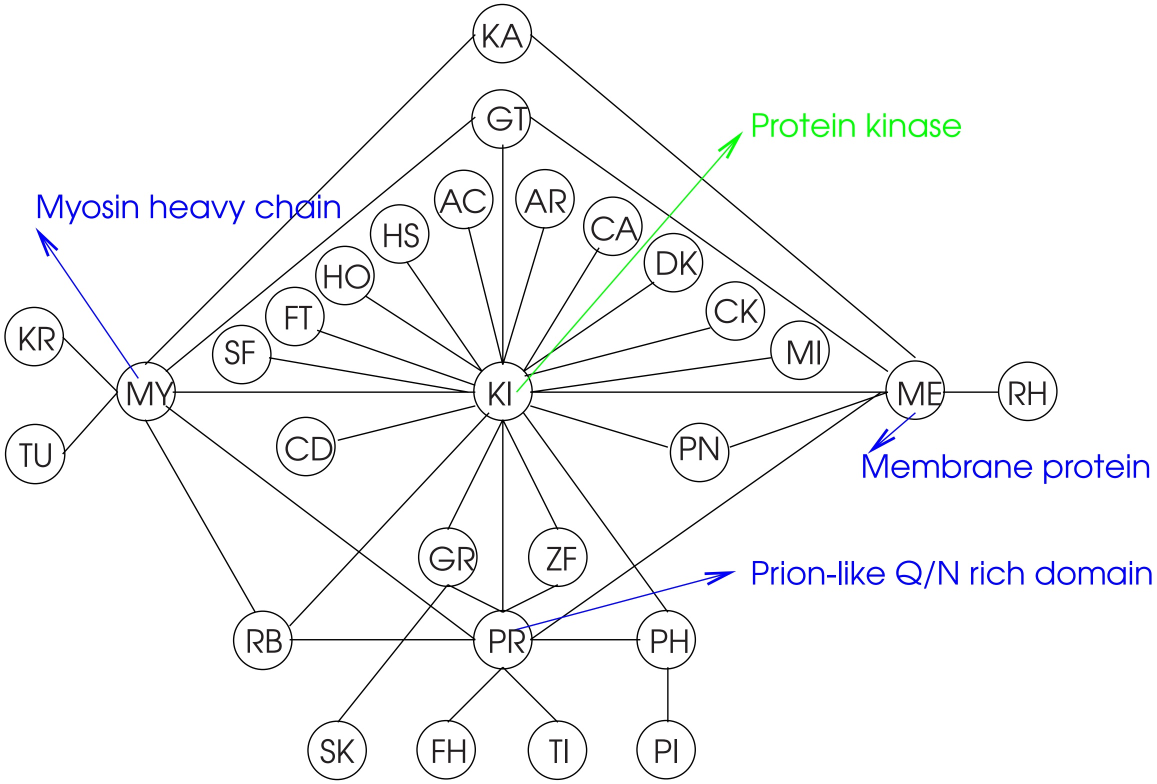
45 (29%) organisms

30 (19%) organisms

22 (14%) organisms

Frequent Interaction Patterns in DIP

- Protein interaction networks for 7 organisms
  - Ecoli, Hpylo, Scere, Celeg, Dmela, Mmusc, Hsapi
  - 44070 interactions between 16783 proteins
- Clustering with TribeMCL & node contraction
  - 30247 interactions between 6714 protein families



Runtime Characteristics

Comparison with isomorphism-based algorithms

		FSG			Mule		
Dataset	Minimum Support (%)	Runtime (secs.)	Largest pattern	Number of patterns	Runtime (secs.)	Largest pattern	Number of patterns
Glutamate	20	0.2	9	12	0.01	9	12
	16	0.7	10	14	0.01	10	14
	12	5.1	13	39	0.10	13	39
	10	22.7	16	34	0.29	15	34
	8	138.9	16	56	0.99	15	56
Alanine	24	0.1	8	11	0.01	8	11
	20	1.5	11	15	0.02	11	15
	16	4.0	12	21	0.06	12	21
	12	112.7	17	25	1.06	16	25
	10	215.1	17	34	1.72	16	34

Extraction of contracted patters

Glutamate metabolism, $\sigma = 8\%$				Alanine metabolism, $\sigma = 10\%$			
Size of contracted pattern	Extraction time (secs.)		Size of extracted pattern	Size of contracted pattern	Extraction time (secs.)		Size of extracted pattern
	FSG	gSpan			FSG	gSpan	
15	10.8	1.12	16	16	54.1	10.13	17
14	12.8	2.42	16	16	24.1	3.92	16
13	1.7	0.31	13	12	0.9	0.27	12
12	0.9	0.30	12	11	0.4	0.13	11
11	0.5	0.08	11	8	0.1	0.01	8
Total number of patterns: 56				Total number of patterns: 34			
Total runtime of FSG alone: 138.9 secs.				Total runtime of FSG alone :215.1 secs.			
Total runtime of Mule+FSG: 0.99+100.5 secs.				Total runtime of Mule+FSG: 1.72+160.6 secs.			
Total runtime of Mule+gSpan: 0.99+16.8 secs.				Total runtime of Mule+gSpan: 1.72+31.0 secs.			

Aligning Protein Interaction Networks

- Defining graph alignment is difficult in general
  - Biological meaning
  - Mathematical modeling

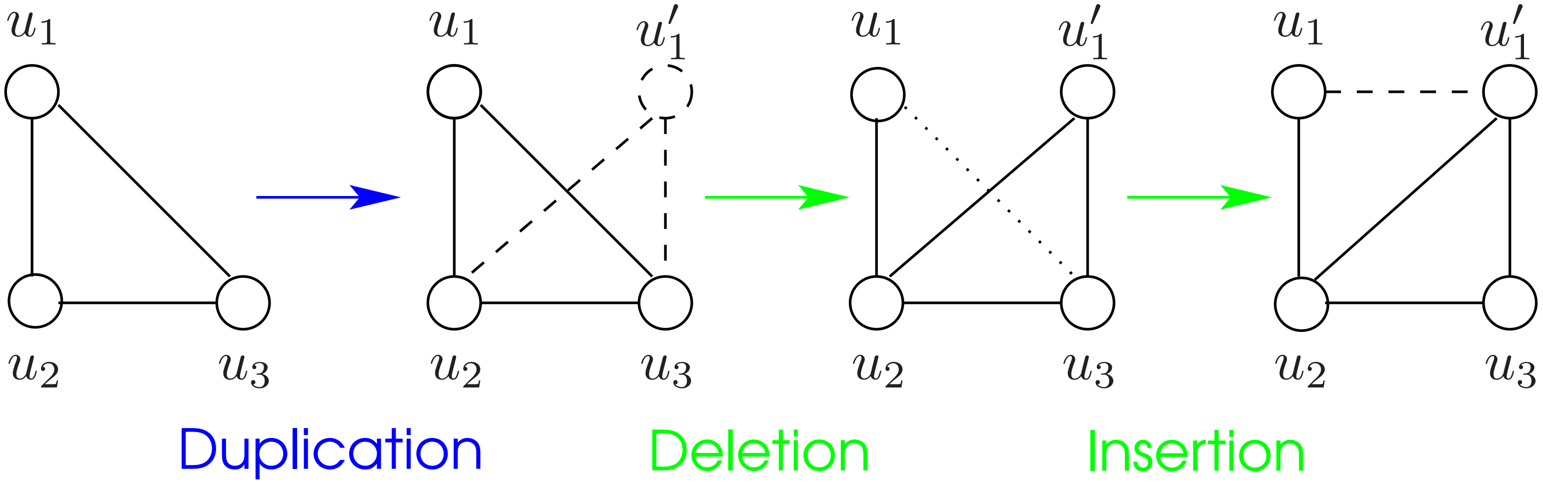
- Existing algorithms are based on simplified formulations
  - PathBLAST aligns pathways (linear chains) to render problem computationally tractable
  - Motif search algorithms look for small topological motifs, do not take into account conservation of proteins

- Our approach
  - Aligns subsets of proteins based on the observation that modules and complexes
  - Guided by models of evolution

Evolution of Protein Interaction Networks

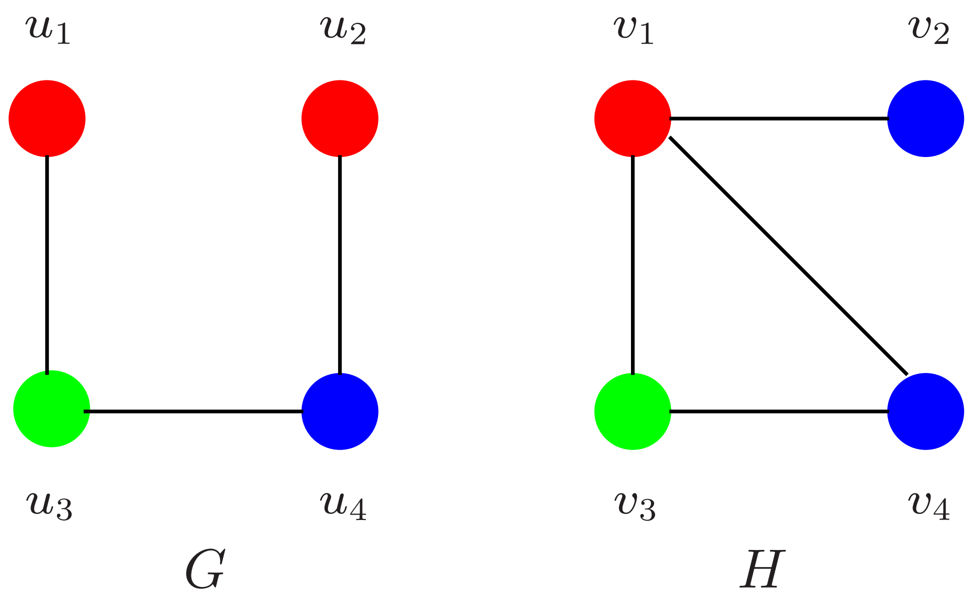
- Duplication/divergence models for the evolution of protein interaction networks
  - Interactions of duplicated proteins are also duplicated
  - Duplicated proteins rapidly lose interactions through mutations

- This provides us with a simplified basis for solving a very hard problem



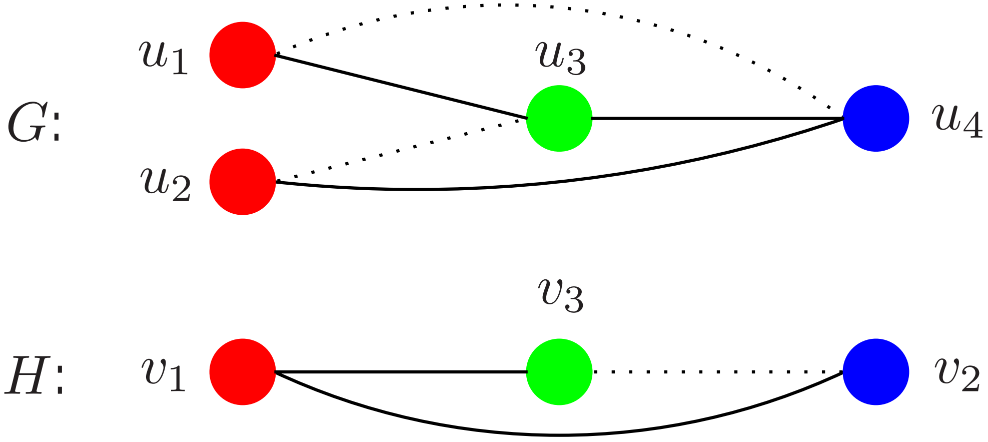
Aligning Protein Interaction Networks: Input

- PINs $G(U, E)$ and $H(V, F)$
- Sparse similarity function $S(u, v)$ for all $u, v \in U \cup V$
  - If $S(u, v) > 0$, u and v are homologous



Local Alignment Induced by Subsets of Proteins

- Alignment induced by protein subset pair $P = \{\tilde{U} \in U, \tilde{V} \in V\}$: $\mathcal{A}(\mathcal{P}) = \{\mathcal{M}, \mathcal{N}, \mathcal{D}\}$
  - A match $\in \mathcal{M}$ correspons to two pairs of homolog proteins from each protein subset such that both pairs interact in both PINs. A match is associated with score μ.
  - A mismatch $\in \mathcal{N}$ corresponds to two pairs of homolog proteins from each PIN such that only one pair is interacting. A mismatch is associated with penalty ν.
  - A duplication $\in D$ corresponds to a pair of homolog proteins that are in the same protein subset. A duplication is associated with penalty δ.



Alignment induced by protein subset pair $\{\{u_1, u_2, u_3, u_4\}, \{v_1, v_2, v_3\}\}$

Pairwise Local Alignment of PINs

- Alignment score:
  $\sigma(\mathcal{A}(P)) = \sum_{M \in \mathcal{M}} \mu(M) - \sum_{N \in \mathcal{N}} \nu(N) - \sum_{D \in \mathcal{D}} \delta(D)$
  - Matches are rewarded for conservation of interactions
  - Duplications are penalized for differentiation after split
  - Mismatches are penalized for divergence and experimental error

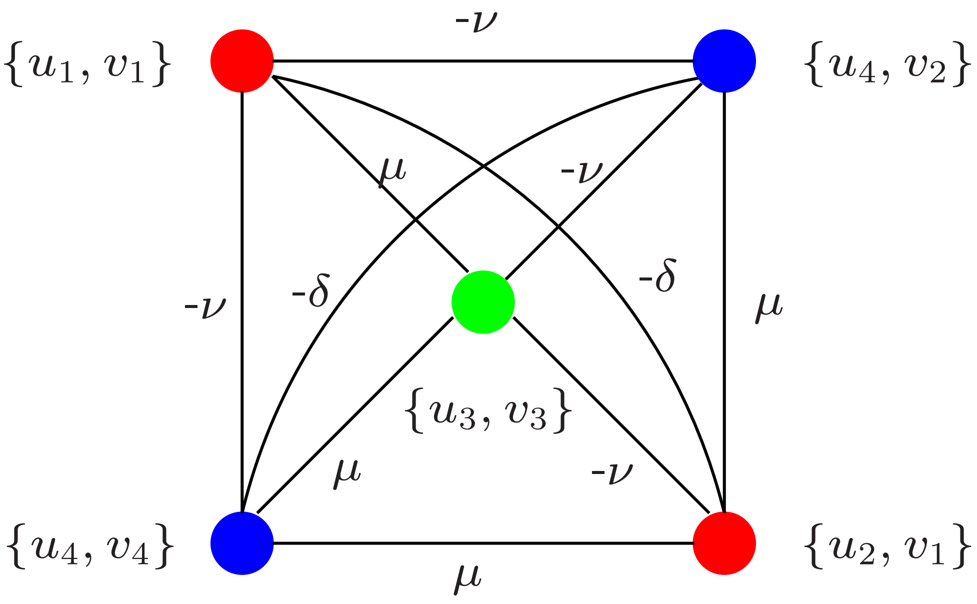
- All scores and penalties are functions of similarity between associated proteins

- Problem: Find all protein subset pairs with alignment score larger than a certain threshold.
  - High scoring protein subsets are likely to correspond to conserved modules or complexes

- A graph equivalent to BLAST

Weighted Alignment Graph $\mathbf{G(V,E)}$

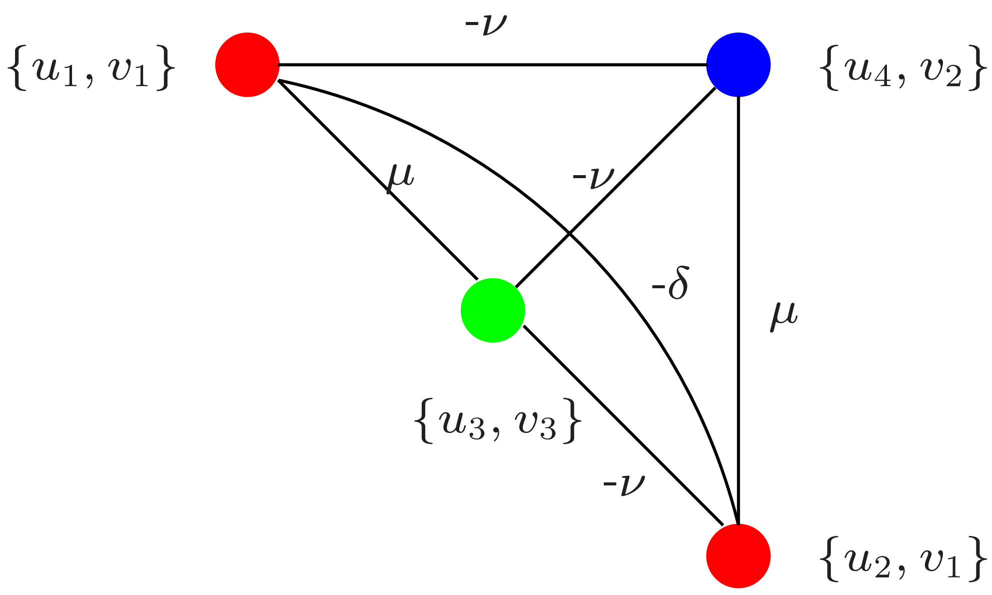
- $\mathbf{V}$ consists all pairs of homolog proteins $\mathbf{v} = \{u \in U, v \in V\}$
- An edge $\mathbf{vv'} = \{uv\}\{u'v'\}$ in $\mathbf{E}$ is a
  - match edge if $uu' \in E$ and $vv' \in V$, with weight $w(\mathbf{vv'}) = \mu(uv, u'v')$
  - mismatch edge if $uu' \in E$ and $vv' \notin V$ or vice versa, with weight $w(\mathbf{vv'}) = -\nu(uv, u'v')$
  - duplication edge if $S(u,u') > 0$ or $S(v,v') > 0$, with weight $w(\mathbf{vv'}) = -\delta(u,u')$ or $w(\mathbf{vv'}) = -\delta(v,v')$



Maximum Weight Induced Subgraph Problem

- Definition: (MAWISH)
  - Given graph $\mathbf{G}(\mathbf{V}, \mathbf{E})$ and a constant ϵ, find $\tilde{\mathbf{V}} \in \mathbf{V}$ such that $\sum_{\mathbf{v},\mathbf{u}\in\tilde{\mathbf{V}}} w(\mathbf{vu}) \geq \epsilon$.
  - NP-complete

- Theorem: (MAWISH $\equiv$ Pairwise alignment)
  - If $\tilde{\mathbf{V}}$ is a solution for the MAWISH problem on $\mathbf{G}(\mathbf{V}, \mathbf{E})$, then $P = \{\tilde{U}, \tilde{V}\}$ induces an alignment $\mathcal{A}(P)$ with $\sigma(\mathcal{A}) \geq \epsilon$, where $\tilde{\mathbf{V}} = \tilde{U} \times \tilde{V}$.



A Greedy Algorithm for MaWISh

- Greedy graph growing
  - Start with a heavily connected node, put it in $\tilde{\mathbf{V}}$
  - Choose $\mathbf{v}$ that is most heavily connected to $\tilde{\mathbf{V}}$ and put it in $\tilde{\mathbf{V}}$ until no $\mathbf{v}$ is positively connected to $\tilde{\mathbf{V}}$.
  - If total weight of the subgraph induced by $\tilde{\mathbf{V}}$ is greater than a threshold, return $\tilde{\mathbf{V}}$
  - Works in linear time.
- As modules and complexes are densely connected within the module and loosely connected to the rest of the network, this algorithm is expected to be effective.
- For all local alignments, remove discovered subgraph and run the greedy algorithm again.
- If the number of homologs for each protein is constant, construction of alignment graph and solution of the MaWISh takes $O(|E| + |F|)$ time.

Scoring Matches, Mismatches and Duplications

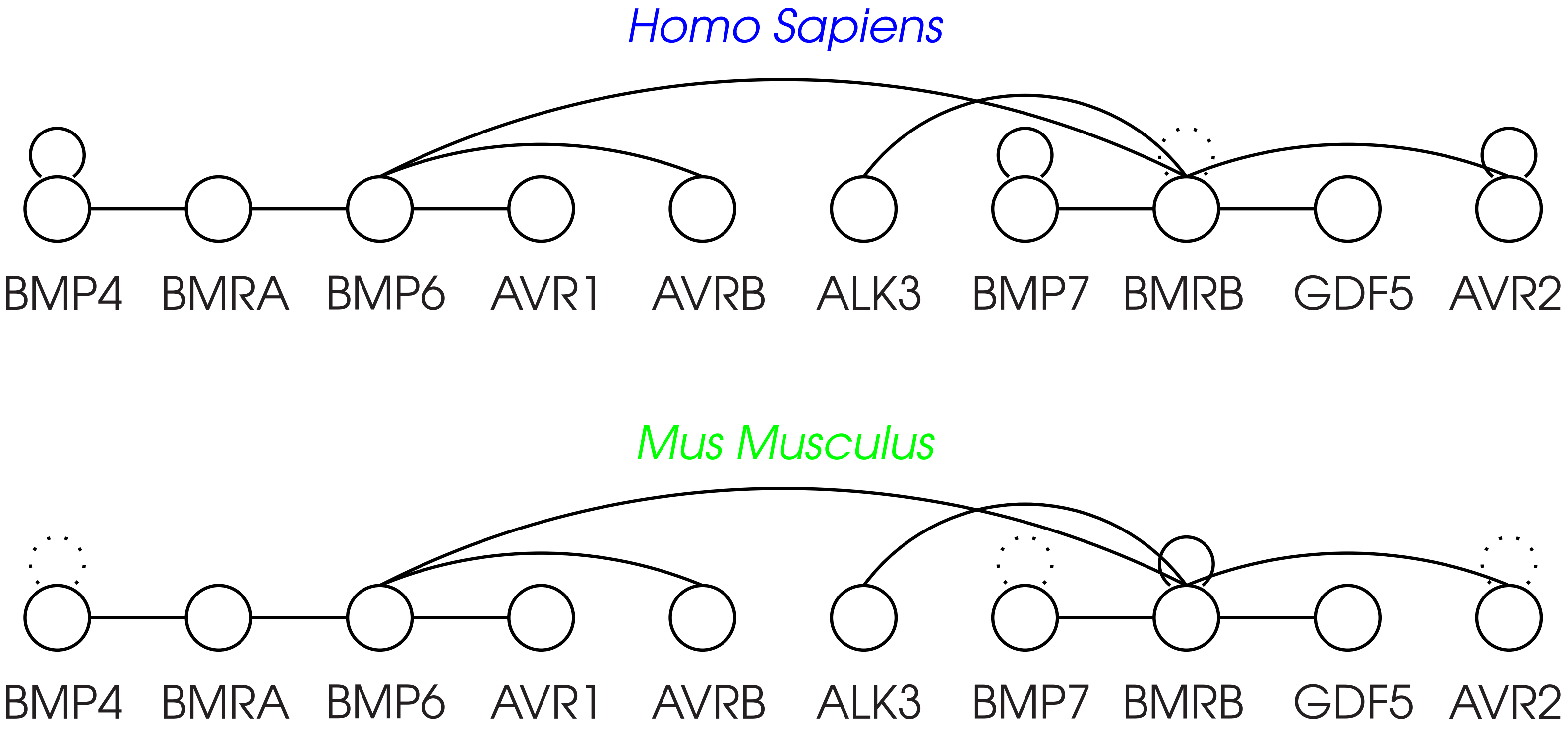
- Quantizing similarity between two proteins
  - Confidence in two proteins being orthologous (paralogous)
  - BLAST E-value: $S(u, v) = log_{10}\frac{p(u,v)}{p_{random}}$
  - Ortholog clustering: $S(u, v) = c(u)c(v)$

- Match score
  - $\mu(uu', vv') = \bar{\mu}\min\{S(u, v), S(u', v')\}$

- Mismatch penalty
  - $\nu(uu', vv') = \bar{\nu}\min\{S(u, v), S(u', v')$

- Duplication penalty
  - $\delta(u, u') = \bar{\delta}(d - S(u, u'))$

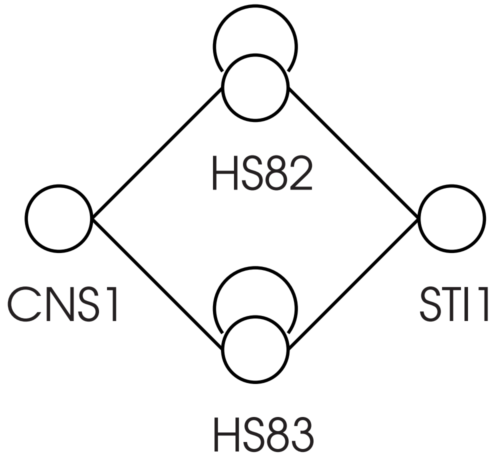
Alignment of Human and Mouse PINs



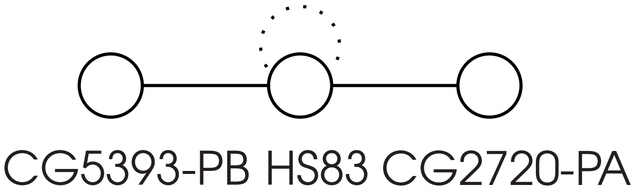
A conserved subnet that is part of
transforming growth factor beta receptor signaling pathway

Alignment of Yeast and Fly PINs

Saccharomyces Cerevisiae



Drosophila Melanogaster



A conserved subnet that is part of response to stress

Ongoing Work on PIN Alignment

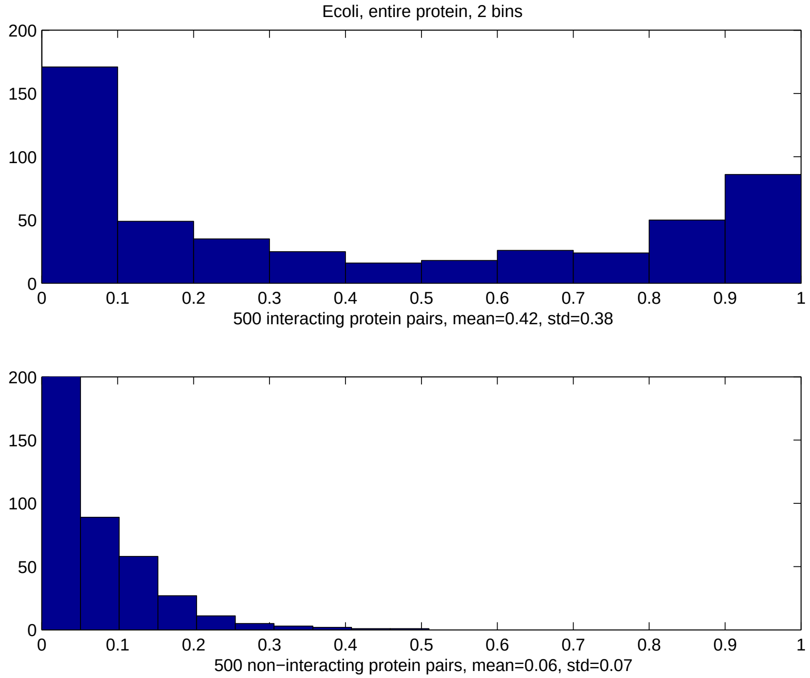
- Assessing statistical significance
  - Constructing a refence model based on models of evolution
- BLAST-like search queries for network alignment
  - Given a query graph, find all high-scoring local alignments in a database of PINs
- Multiple Graph Alignment (CLUSTAL, BLASTCLUST)
  - How to combine graph mining and pairwise alignment
- Web-based interface for PIN alignment queries

Constructing Module Maps

- Find functional modules in a comprehensive PIN (yeast) through graph clustering
- Find best matches to these modules in several species through pairwise alignment
- Construct canonical module maps using these alignments
- Analyze canonical pathways on these maps

Clustering Phylogeny Profiles for Module Detection

- Interacting proteins are likely to have co-evolved
- Phylogeny profiles have been successful in predicting interactions
- We can discover functional modules and complexes through clustering phylogeny profiles
- However, significant challenges remain



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