Bioinformatics 3

V 5 – Weak Indicators and Communities

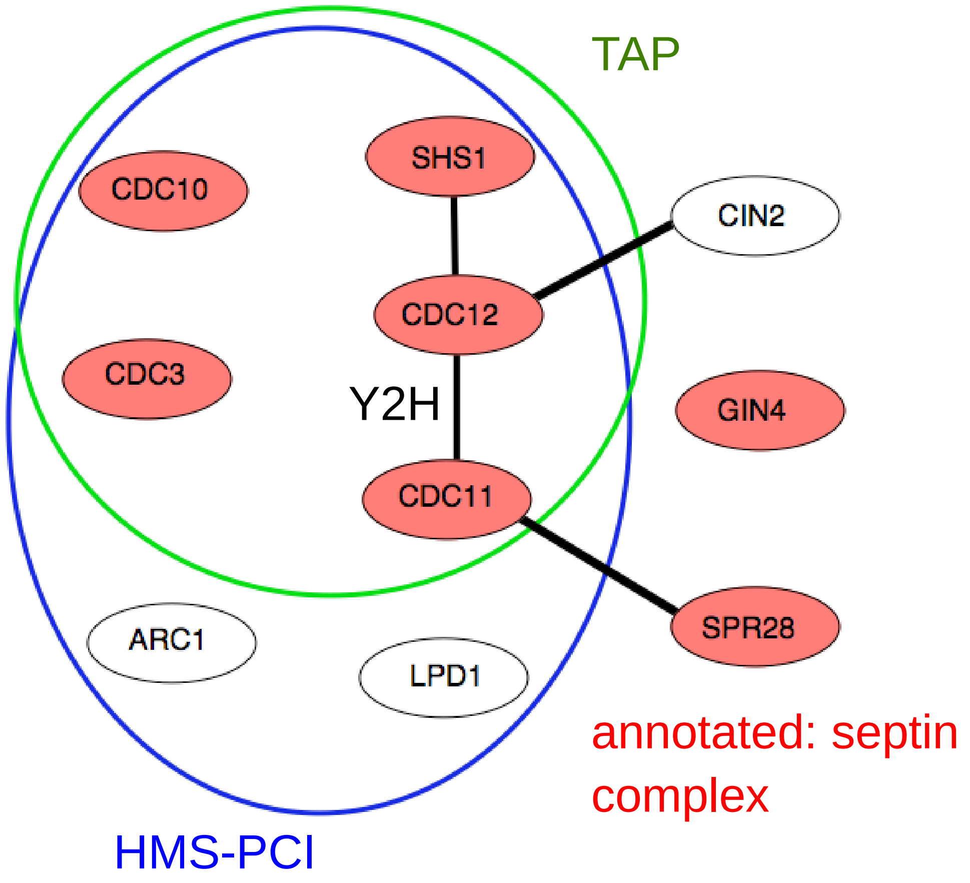
Fri, April 27, 2018

Noisy Data — Clear Statements?

For **yeast**: ~ 6000 proteins → ~18 million potential interactions
rough estimates: ≤ 100000 interactions occur

→ 1 true positive for 200 potential candidates = **0.5%**
→ **decisive** experiment must have **accuracy** << 0.5% false positives

Different experiments detect different interactions
For yeast: 80000 interactions known,
only 2400 found in > 1 experiment



Y2H: → many false positives
(up to 50% errors)

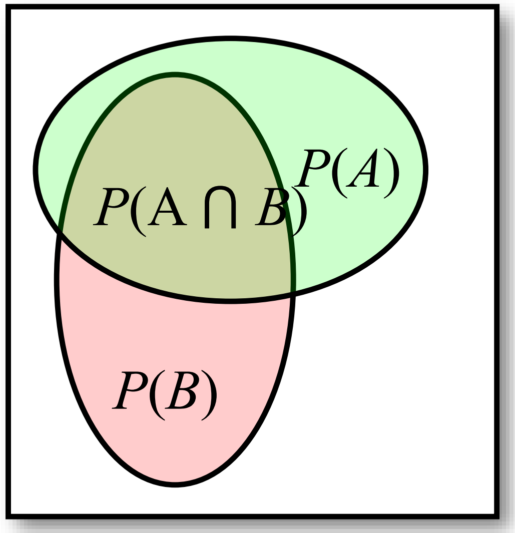
Co-expression: → gives indications at best

Combine weak indicators = ???

see: von Mering (2002)

Conditional Probabilities

Joint probability for "*A* and *B*":



$$P(A \cap B) = P(A|B)\,P(B) = P(B|A)\,P(A)$$

Solve for conditional probability for "*A* when *B* is true"
→ Bayes' Theorem:

$$P(A|B) = \frac{P(B|A)\,P(A)}{P(B)} = \frac{P(B|A)}{P(B)}\,P(A)$$

P(*A*) = prior probability (marginal prob.) for "*A*" → no prior knowledge about *A*

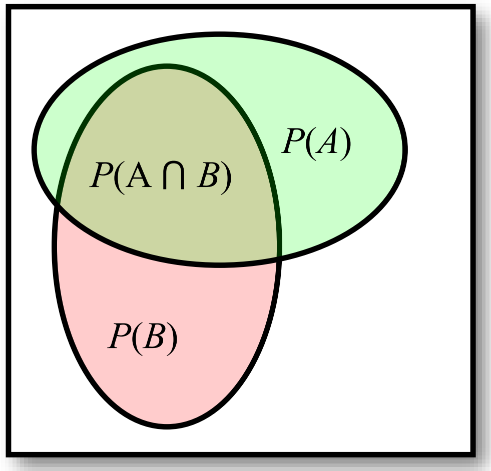
P(*B*) = prior probability for "*B*" → normalizing constant

P(*B* | *A*) = conditional probability for "*B* given *A*"

P(*A* | *B*) = posterior probability for "*A* given *B*"

→ Use information about *B* to improve knowledge about *A*

What are the Odds?



Express Bayes theorem

$$P(A|B) = \frac{P(B|A)\,P(A)}{P(B)} = \frac{P(B|A)}{P(B)}\,P(A)$$

in terms of odds:

- Also consider case "*A* does not apply": $P(\bar{A}|B) = \frac{P(B|\bar{A})}{P(B)}\,P(\bar{A})$

- odds for *A* when we know about *B*

(we will interpret *B* as information or features):

$$O(A|B) = \frac{P(A|B)}{P(\bar{A}|B)} = \frac{P(B|A)}{P(B|\bar{A})}\,\frac{P(A)}{P(\bar{A})} = \Lambda(A|B)\,O(A)$$

posterior odds for *A* likelihood ratio prior odds for *A*

Λ(*A* | *B*) → by how much does our knowledge about *A* improve?

2 types of Bayesian Networks

(1) **Naive Bayesian network**
→ independent odds

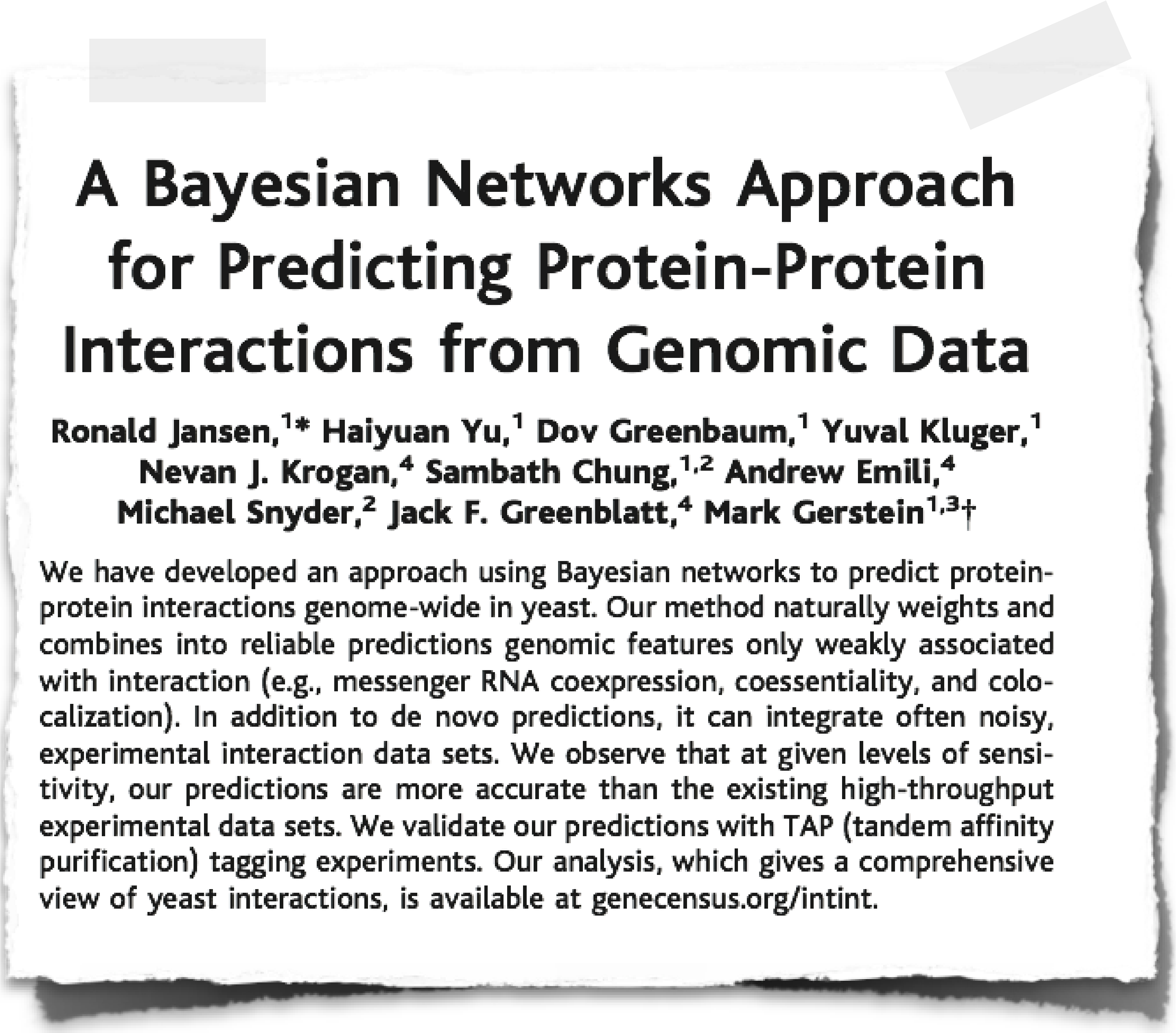
$$O(A|B,C) = \Lambda(A|B)\,\Lambda(A|C)\,O(A)$$

(2) **Fully connected Bayesian network**
→ table of joint odds

	B	!B
C	0.3	0.16
!C	0.4	0.14

$$\Leftrightarrow \Lambda(A|B,C)$$

Bayesian Analysis of Complexes



A Bayesian Networks Approach for Predicting Protein-Protein Interactions from Genomic Data

Ronald Jansen,[1*] Haiyuan Yu,[1] Dov Greenbaum,[1] Yuval Kluger,[1] Nevan J. Krogan,[4] Sambath Chung,[1,2] Andrew Emili,[4] Michael Snyder,[2] Jack F. Greenblatt,[4] Mark Gerstein[1,3]†

We have developed an approach using Bayesian networks to predict protein-protein interactions genome-wide in yeast. Our method naturally weights and combines into reliable predictions genomic features only weakly associated with interaction (e.g., messenger RNA coexpression, coessentiality, and colocalization). In addition to de novo predictions, it can integrate often noisy, experimental interaction data sets. We observe that at given levels of sensitivity, our predictions are more accurate than the existing high-throughput experimental data sets. We validate our predictions with TAP (tandem affinity purification) tagging experiments. Our analysis, which gives a comprehensive view of yeast interactions, is available at genecensus.org/intint.

Science 302 (2003) 449

Improving the Odds

Is a given protein pair **AB a complex** (from all that we know)?

$$O_{post}(\text{Complex}|f_1, f_2, \ldots) = \Lambda(\text{Complex}|f_1, f_2, \ldots)\, O_{prior}(\text{Complex})$$

likelihood ratio:
improvement of the odds when
we know about features *f*1, *f*2, …

Idea: determine from known complexes
and use for prediction of new complexes

prior odds for a
random pair AB to be
a complex

estimate (somehow)

Features used by Jansen et al (2003):

- 4 experimental data sets of complexes
- mRNA co-expression profiles
- biological functions annotated to the proteins (GO, MIPS)
- essentiality for the cell

Gold Standard Sets

To determine $\Lambda(\text{Complex}|f_1, f_2, \ldots) = \dfrac{P(f_1, f_2, \ldots|\text{Complex})}{P(f_1, f_2, \ldots|\text{no Complex})}$

→ use two data sets with **known** features f_1, f_2, … for **training**

Requirements for training data:

i) independent of the data serving as evidence

ii) large enough for good statistics

iii) free of systematic bias

Gold Standard Positive Set (GP):
8250 complexes from the hand-curated MIPS catalog of protein complexes
(MIPS stands for Munich Information Center for Protein Sequences)

Gold Standard Negative Set (GN):
2708746 (non-)complexes formed by proteins from different cellular compartments (assuming that such protein pairs likely do not interact)

Prior Odds

$$O_{prior}(\text{Complex}) = \frac{P(\text{Complex})}{P(\text{no Complex})} = \frac{P(\text{Complex})}{1 - P(\text{Complex})}$$

Jansen et al:

- estimated ≥ 30000 existing complexes in yeast
- 18 Mio. possible complexes → *P*(Complex) ≈ 1/600

→ O_{prior} = 1/600

→ The odds are 600 : 1 against picking a real complex at random

→ expect 50% good hits (TP ≥ FP) when Λ ≈ 600 and higher

Note: O_{prior} is mostly an educated guess

Essentiality

Test whether both proteins are essential (E) for the cell or not (N)
→ for protein complexes, EE or NN should occur more often

pos/neg: # of gold standard positives/ negatives with essentiality information

$$L(\mathrm{Ess}) = \frac{P(\mathrm{Ess}\,|\,\mathrm{pos})}{P(\mathrm{Ess}\,|\,\mathrm{neg})}$$

Essentiality	pos	neg	P(Ess|pos)	P(Ess|neg)	L(Ess)
EE	1114	81924	5,18E-01	1,43E-01	3,6
NE	624	285487	2,90E-01	4,98E-01	0,6
NN	412	206313	1,92E-01	3,60E-01	0,5
sum	2150	573724	1,00	1,00	

possible values of the feature

overlap of gold standard sets with feature values

In the „pos" case, the essentiality was only known for 2150 out of 8250 complexes of the gold-standard.

probabilities for each feature value

likelihood ratios

$$\frac{1114}{2150} = 0,518$$

$$\frac{0.19}{0.36} = 0,5$$

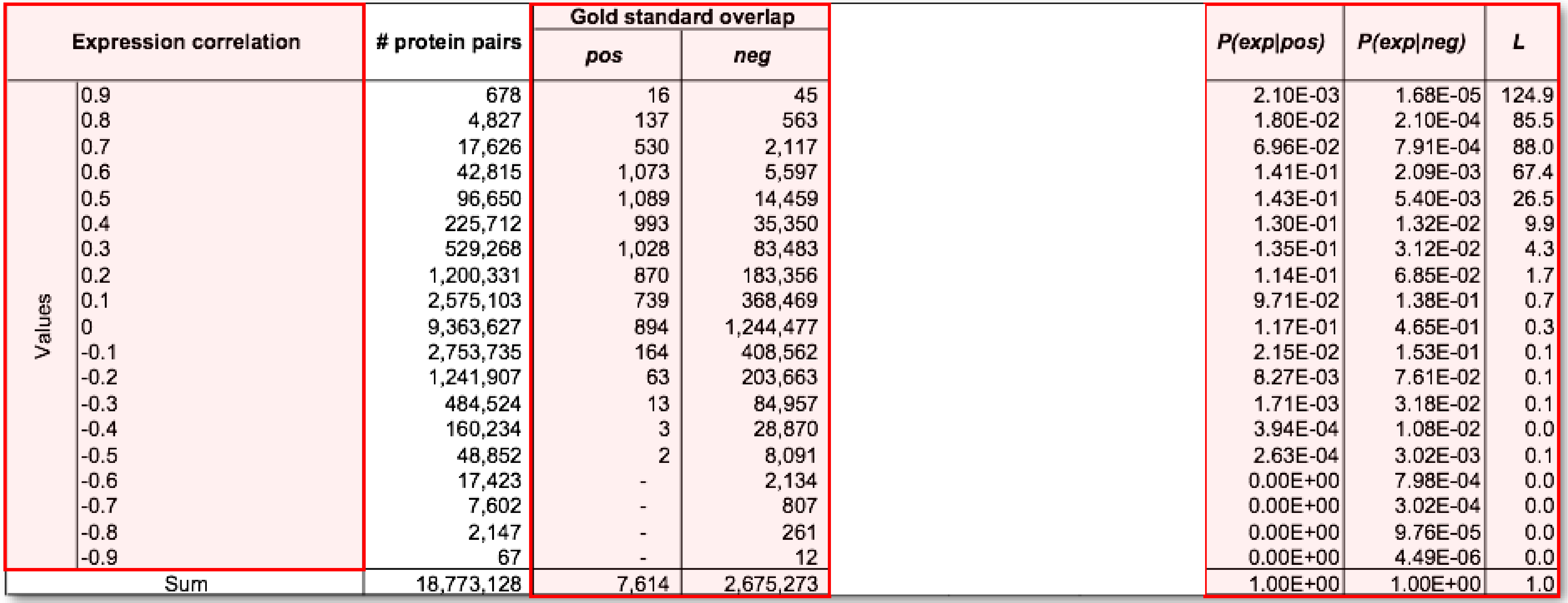
-> Essentiality is a weak feature!

mRNA Co-Expression

Publicly available expression data from

- the Rosetta compendium
- the yeast cell cycle

Correlation between the data sets
→ use principal component



	Expression correlation	# protein pairs	Gold standard overlap			P(exp|pos)	P(exp|neg)	L
			pos	*neg*				
Values	0.9	678	16	45		2.10E-03	1.68E-05	124.9
	0.8	4,827	137	563		1.80E-02	2.10E-04	85.5
	0.7	17,626	530	2,117		6.96E-02	7.91E-04	88.0
	0.6	42,815	1,073	5,597		1.41E-01	2.09E-03	67.4
	0.5	96,650	1,089	14,459		1.43E-01	5.40E-03	26.5
	0.4	225,712	993	35,350		1.30E-01	1.32E-02	9.9
	0.3	529,268	1,028	83,483		1.35E-01	3.12E-02	4.3
	0.2	1,200,331	870	183,356		1.14E-01	6.85E-02	1.7
	0.1	2,575,103	739	368,469		9.71E-02	1.38E-01	0.7
	0	9,363,627	894	1,244,477		1.17E-01	4.65E-01	0.3
	-0.1	2,753,735	164	408,562		2.15E-02	1.53E-01	0.1
	-0.2	1,241,907	63	203,663		8.27E-03	7.61E-02	0.1
	-0.3	484,524	13	84,957		1.71E-03	3.18E-02	0.1
	-0.4	160,234	3	28,870		3.94E-04	1.08E-02	0.0
	-0.5	48,852	2	8,091		2.63E-04	3.02E-03	0.1
	-0.6	17,423	-	2,134		0.00E+00	7.98E-04	0.0
	-0.7	7,602	-	807		0.00E+00	3.02E-04	0.0
	-0.8	2,147	-	261		0.00E+00	9.76E-05	0.0
	-0.9	67	-	12		0.00E+00	4.49E-06	0.0
Sum		18,773,128	7,614	2,675,273		1.00E+00	1.00E+00	1.0

-> Co-expression is a much better feature than essentiality!

Jansen et al, Science 302 (2003) 449

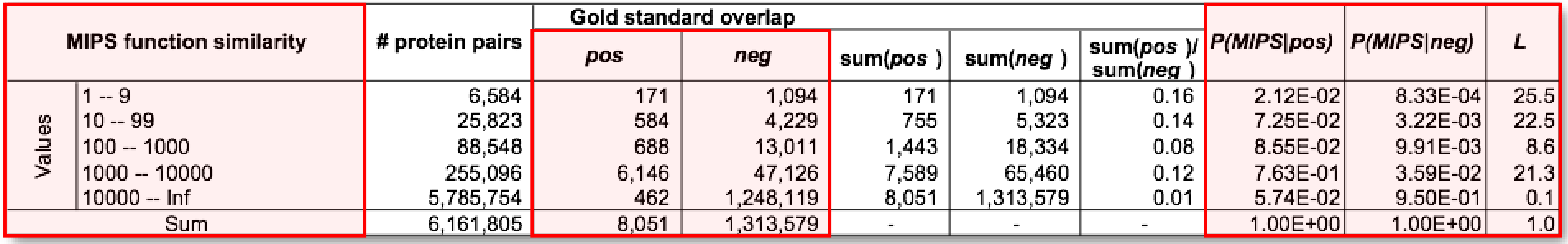
Biological Function

Use MIPS function catalog and Gene Ontology function annotations

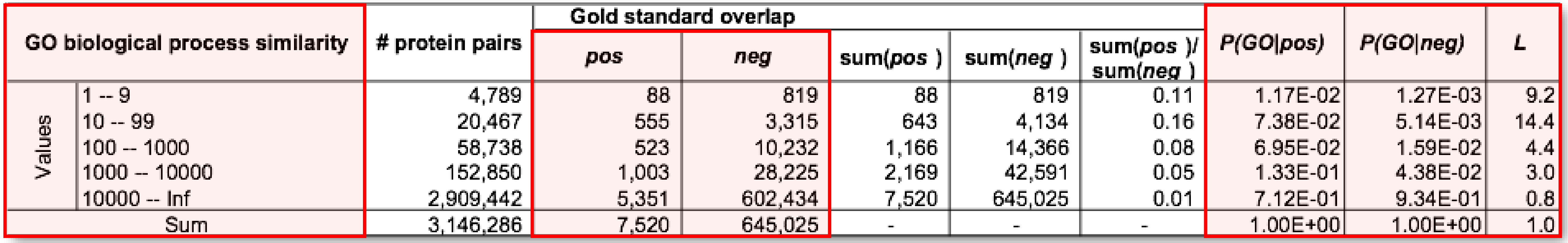
- determine functional class shared by the two proteins; small values (1-9)

Indicate highest MIPS function or GO BP similarity

- count how many of the 18 Mio potential pairs share this classification



MIPS function similarity		# protein pairs	Gold standard overlap					*P(MIPS|pos)*	*P(MIPS|neg)*	*L*
			pos	*neg*	sum(*pos*)	sum(*neg*)	sum(*pos*)/ sum(*neg*)			
Values	1 -- 9	6,584	171	1,094	171	1,094	0.16	2.12E-02	8.33E-04	25.5
	10 -- 99	25,823	584	4,229	755	5,323	0.14	7.25E-02	3.22E-03	22.5
	100 -- 1000	88,548	688	13,011	1,443	18,334	0.08	8.55E-02	9.91E-03	8.6
	1000 -- 10000	255,096	6,146	47,126	7,589	65,460	0.12	7.63E-01	3.59E-02	21.3
	10000 -- Inf	5,785,754	462	1,248,119	8,051	1,313,579	0.01	5.74E-02	9.50E-01	0.1
Sum		6,161,805	8,051	1,313,579	-	-	-	1.00E+00	1.00E+00	1.0



GO biological process similarity		# protein pairs	Gold standard overlap					*P(GO|pos)*	*P(GO|neg)*	*L*
			pos	*neg*	sum(*pos*)	sum(*neg*)	sum(*pos*)/ sum(*neg*)			
Values	1 -- 9	4,789	88	819	88	819	0.11	1.17E-02	1.27E-03	9.2
	10 -- 99	20,467	555	3,315	643	4,134	0.16	7.38E-02	5.14E-03	14.4
	100 -- 1000	58,738	523	10,232	1,166	14,366	0.08	6.95E-02	1.59E-02	4.4
	1000 -- 10000	152,850	1,003	28,225	2,169	42,591	0.05	1.33E-01	4.38E-02	3.0
	10000 -- Inf	2,909,442	5,351	602,434	7,520	645,025	0.01	7.12E-01	9.34E-01	0.8
Sum		3,146,286	7,520	645,025	-	-	-	1.00E+00	1.00E+00	1.0

-> Co-Functionality is a semi-weak feature!

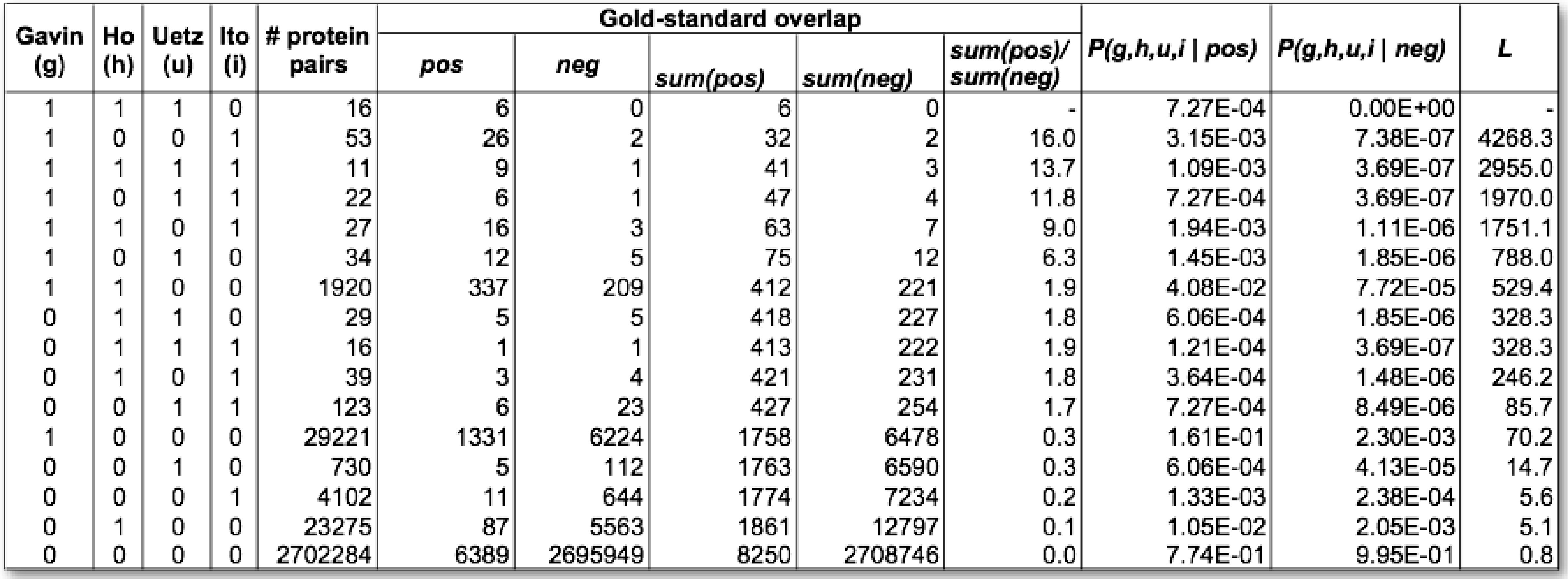
Jansen et al, Science 302 (2003) 449

Experimental Data Sets

In vivo pull-down:	Gavin et al, *Nature* **415** (2002) 141	31304 pairs
	Ho et al, *Nature* **415** (2002) 180	25333 pairs
HT-Y2H:	Uetz et al, *Nature* **403** (2000) 623	981 pairs
	Ito et al, *PNAS* **98** (2001) 4569	4393 pairs

4 experiments on overlapping PP pairs

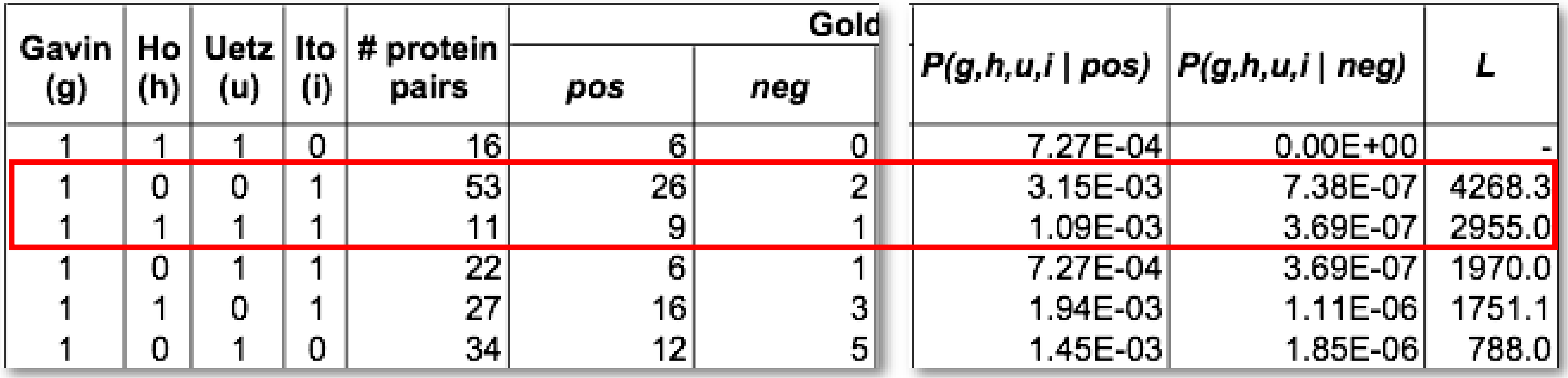
→ $2^4 = 16$ categories — table represents fully connected Bayes network



Gavin (g)	Ho (h)	Uetz (u)	Ito (i)	# protein pairs	Gold-standard overlap					*P(g,h,u,i | pos)*	*P(g,h,u,i | neg)*	*L*
					pos	*neg*	*sum(pos)*	*sum(neg)*	*sum(pos)/ sum(neg)*			
1	1	1	0	16	6	0	6	0	-	7.27E-04	0.00E+00	-
1	0	0	1	53	26	2	32	2	16.0	3.15E-03	7.38E-07	4268.3
1	1	1	1	11	9	1	41	3	13.7	1.09E-03	3.69E-07	2955.0
1	0	1	1	22	6	1	47	4	11.8	7.27E-04	3.69E-07	1970.0
1	1	0	1	27	16	3	63	7	9.0	1.94E-03	1.11E-06	1751.1
1	0	1	0	34	12	5	75	12	6.3	1.45E-03	1.85E-06	788.0
1	1	0	0	1920	337	209	412	221	1.9	4.08E-02	7.72E-05	529.4
0	1	1	0	29	5	5	418	227	1.8	6.06E-04	1.85E-06	328.3
0	1	1	1	16	1	1	413	222	1.9	1.21E-04	3.69E-07	328.3
0	1	0	1	39	3	4	421	231	1.8	3.64E-04	1.48E-06	246.2
0	0	1	1	123	6	23	427	254	1.7	7.27E-04	8.49E-06	85.7
1	0	0	0	29221	1331	6224	1758	6478	0.3	1.61E-01	2.30E-03	70.2
0	0	1	0	730	5	112	1763	6590	0.3	6.06E-04	4.13E-05	14.7
0	0	0	1	4102	11	644	1774	7234	0.2	1.33E-03	2.38E-04	5.6
0	1	0	0	23275	87	5563	1861	12797	0.1	1.05E-02	2.05E-03	5.1
0	0	0	0	2702284	6389	2695949	8250	2708746	0.0	7.74E-01	9.95E-01	0.8

Jansen et al, Science 302 (2003) 449

Statistical Uncertainties



Gavin (g)	Ho (h)	Uetz (u)	Ito (i)	# protein pairs	Gold		P(g,h,u,i | pos)	P(g,h,u,i | neg)	L
					pos	neg			
1	1	1	0	16	6	0	7.27E-04	0.00E+00	-
1	0	0	1	53	26	2	3.15E-03	7.38E-07	4268.3
1	1	1	1	11	9	1	1.09E-03	3.69E-07	2955.0
1	0	1	1	22	6	1	7.27E-04	3.69E-07	1970.0
1	1	0	1	27	16	3	1.94E-03	1.11E-06	1751.1
1	0	1	0	34	12	5	1.45E-03	1.85E-06	788.0

1) $L(1111) < L(1001)$

statistical uncertainty: $\Delta N = \sqrt{N+1}$

Overlap with all experiments is smaller → larger uncertainty

2) $L(1110)$ = NaN?

Use conservative lower bound → assume 1 overlap with GN

→ $L(1110) \geq 1970$

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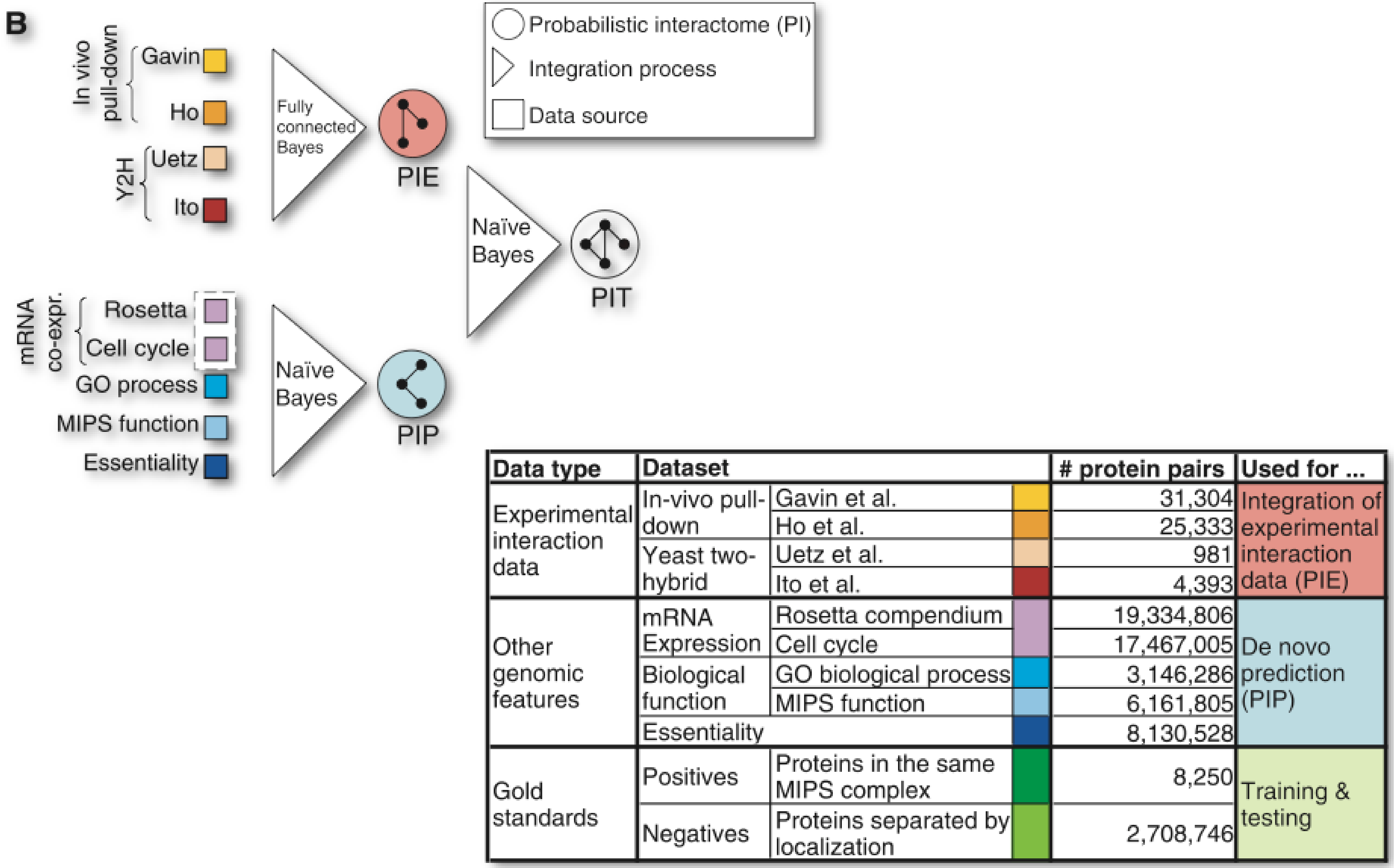
Overview



Data type	Dataset			# protein pairs	Used for ...
Experimental interaction data	In-vivo pull-down	Gavin et al.	🟨	31,304	Integration of experimental interaction data (PIE)
		Ho et al.	🟧	25,333	
	Yeast two-hybrid	Uetz et al.	🟨	981	
		Ito et al.	🟥	4,393	
Other genomic features	mRNA Expression	Rosetta compendium	🟪	19,334,806	De novo prediction (PIP)
		Cell cycle	🟪	17,467,005	
	Biological function	GO biological process	🟦	3,146,286	
		MIPS function	🟦	6,161,805	
	Essentiality		🟦	8,130,528	
Gold standards	Positives	Proteins in the same MIPS complex	🟩	8,250	Training & testing
	Negatives	Proteins separated by localization	🟩	2,708,746	

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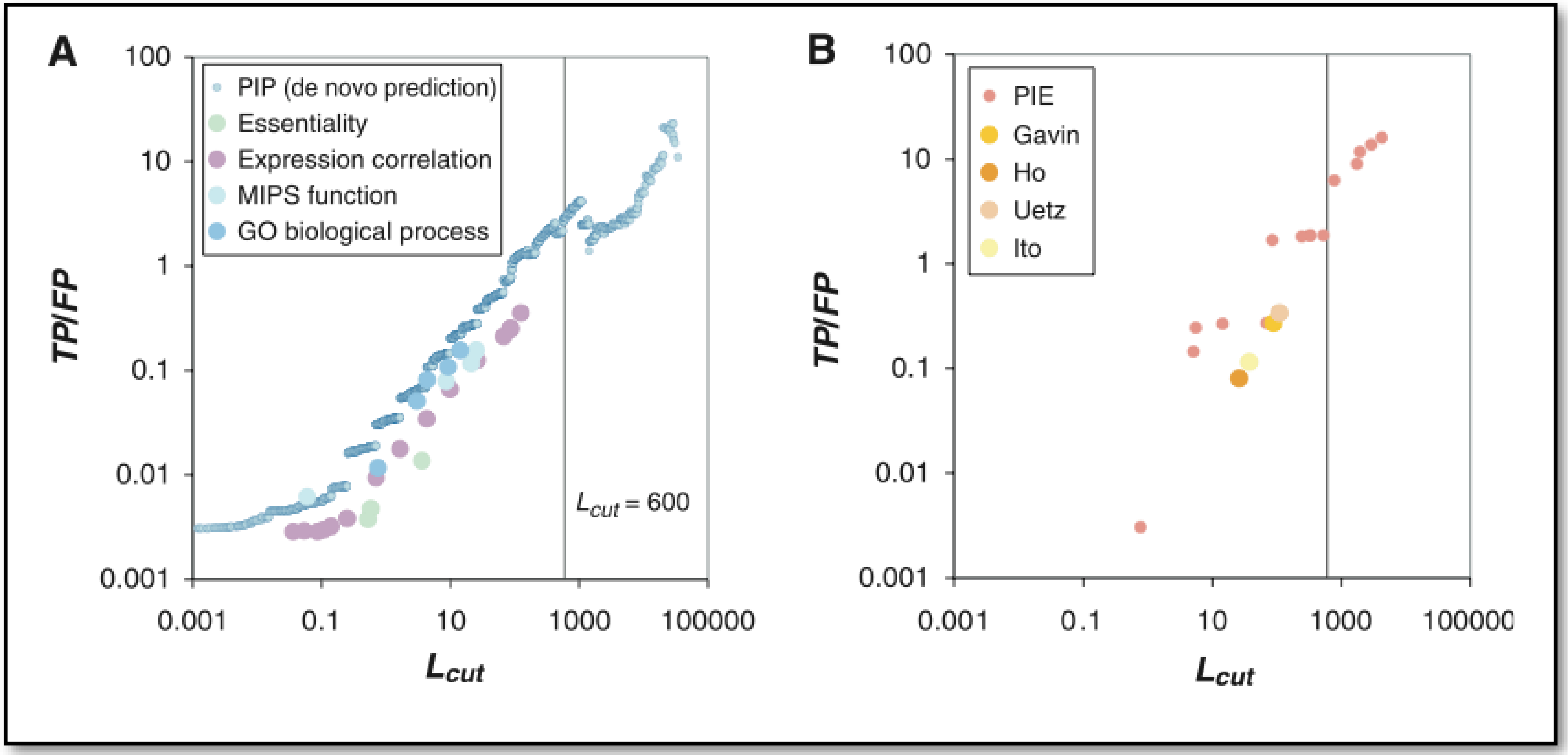
Performance of complex prediction

Predictions

Experimental data



None of the individual evidences alone was enough to get a likelihood ratio > 600,

neither predicted nor experimental evidences

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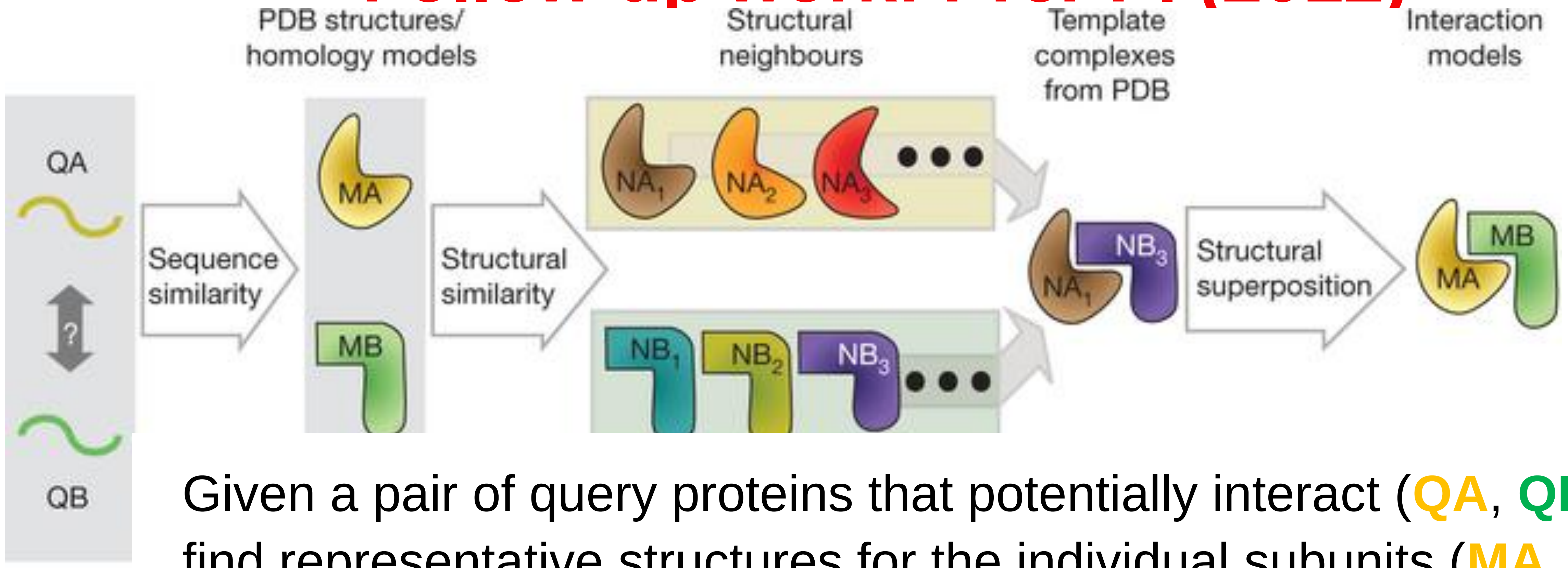
Follow-up work: PrePPI (2012)



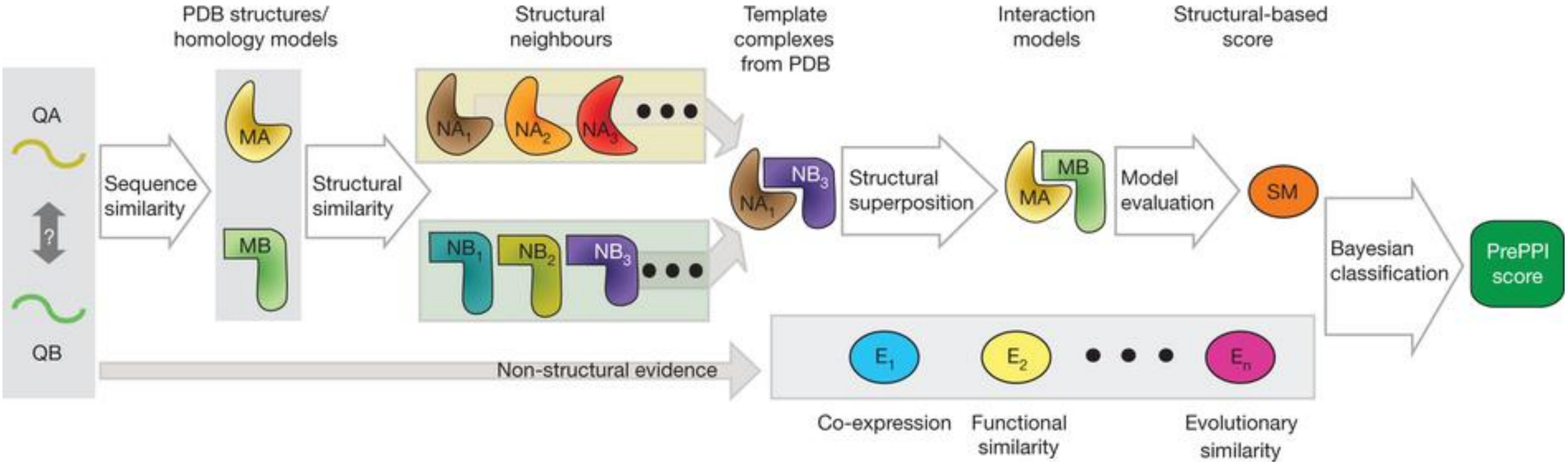
Given a pair of query proteins that potentially interact (QA, QB), try to find representative structures for the individual subunits (MA, MB) in the PDB, where available, or from homology model databases.

For each subunit, find both close and remote **structural neighbors**.
A ‘**template**’ for the interaction exists whenever a PDB structure contains a pair of inter-acting chains (e.g. NA_1–NB_3) that are structural neighbors of MA and MB, respectively.

A **model** is constructed by **superposing** the individual subunits, MA and MB, on their corresponding structural neighbors, NA_1 and NB_3.

Zhang et al, Nature (2012) 490, 556–560

Follow-up work: PrePPI (2012)



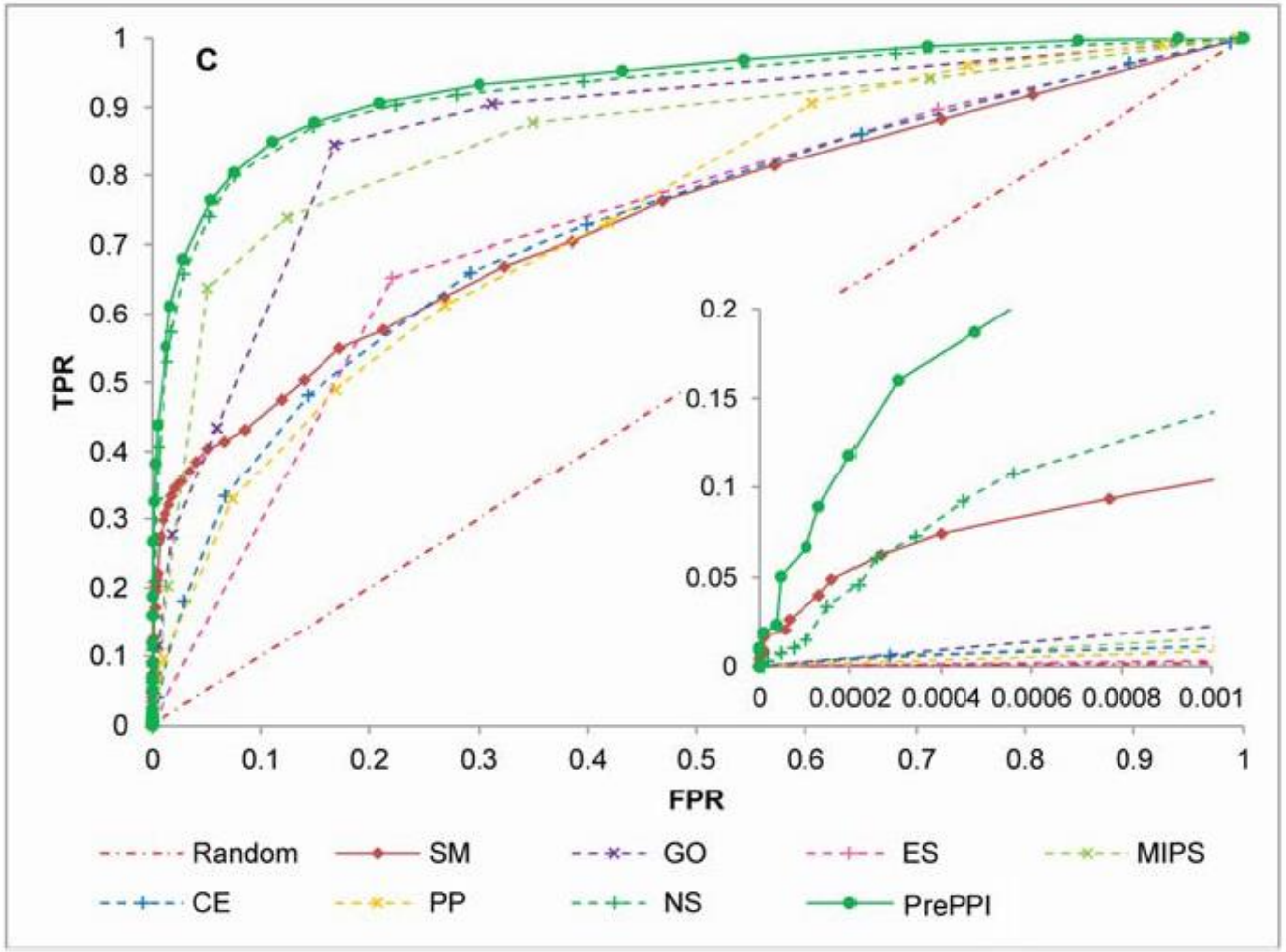
We assign 5 empirical-structure-based scores to each interaction model and then calculate a likelihood for each model to represent a true interaction by combining these scores using a Bayesian network trained on a high-confidence data set of positive interactors and a reference set of non-interactors.

We finally combine the structure-derived score (SM) with non-structural evidence associated with the query proteins (for example, co-expression, functional similarity) using a **naive Bayesian classifier**.

Zhang et al, Nature (2012) 490, 556–560

Results of PrePPI

Receiver-operator characteristics (ROC) for predicted yeast complexes.



Examined features:

- structural modeling (SM),
- GO similarity,
- protein essentiality (ES) relationship,
- MIPS similarity,
- co-expression (CE),
- phylogenetic profile (PP) similarity.

Also listed are 2 combinations:

- NS for the integration of all non-structure clues, i.e. GO, ES, MIPS, CE, and PP, and
- PrePPI for all structural and non-structure clues).

This gave 30.000 high-confidence PP interactions for yeast and 300.000 for human.

Jansen et al, Science 302 (2003) 449

Summary: Bayesian Analysis

Combination of weak features yields powerful predictions

- boosts odds via Bayes' theorem
- Gold standard sets for training the likelihood ratios

Bayes vs. other **machine learning** techniques:
(voting, unions, SVM, neuronal networks, decision trees, …)

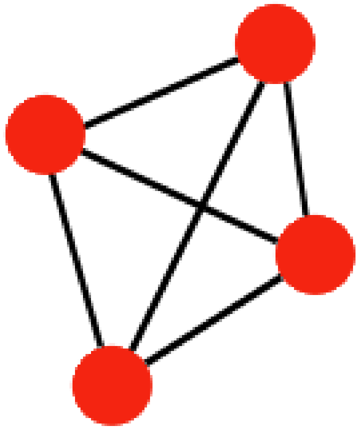
→ **arbitrary types** of data can be combined
→ weight data according to their **reliability**
→ include conditional relations between evidences
→ easily accommodates missing data (e.g., zero overlap with GN)
→ **transparent** procedure
→ predictions easy to **interpret**

Connected Regions

Observation: there are **more interactions inside** a complex than to the outside

→ how can one identify highly connected regions in a network?

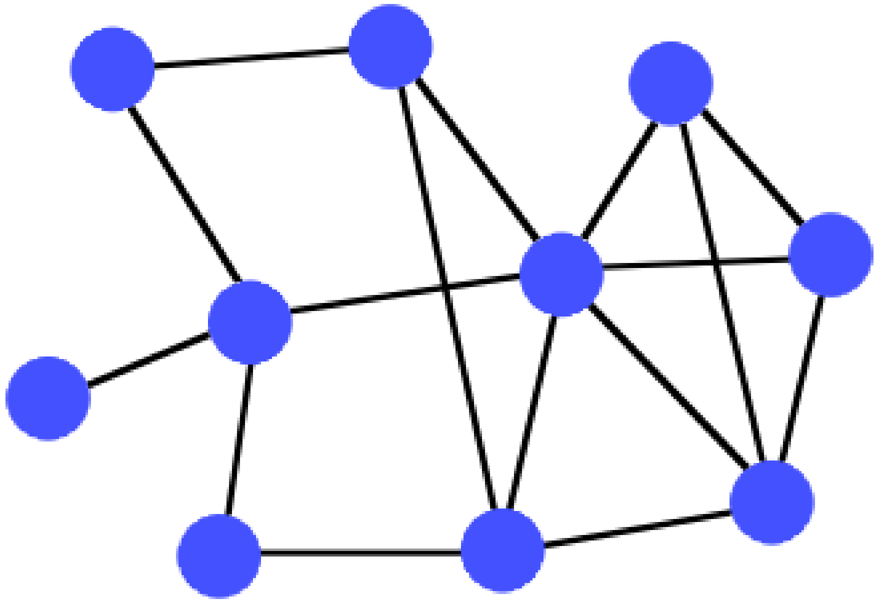
1) Fully connected region: **Clique**



clique := $G' = (V', E' = V'^{(2)})$

Problems with cliques:

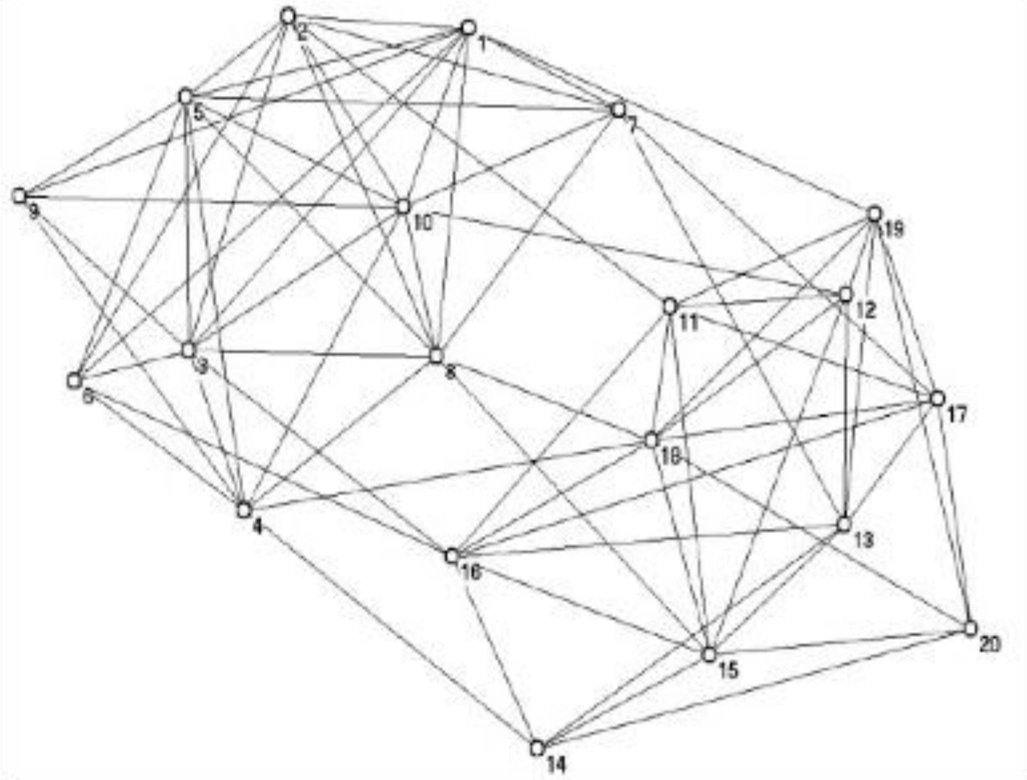
- finding cliques is **NP-hard** (but can be done in $O(N^2)$ for sparsely connected biological networks)
- **biological** protein complexes are **not** always **fully** connected



Communities

Community := subset of vertices, for which the **internal** connectivity is **denser** than to the outside

Aim: map network onto tree that reflects the community structure



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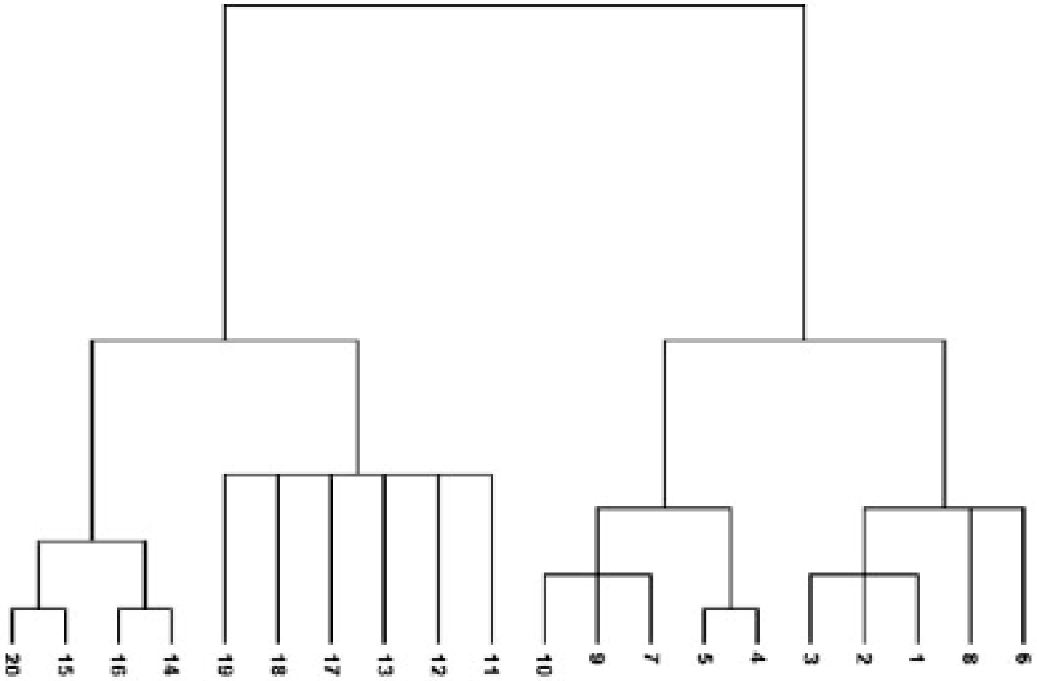


Radicchi et al, *PNAS* **101** (2004) 2658:

Define communities by agglomerative clustering

1) Assign a weight W_{ij} to each pair of vertices *i, j* that measures how "closely related" these two vertices are.

2) Iteratively add edges between pairs of nodes with decreasing W_{ij}

Measures for W_{ij}:

1) Number of **vertex-independent paths** between vertices *i* and *j*
(vertex-independent paths between *i* and *j*: no shared vertex except *i* and *j*)

Menger (1927): the number of vertex-independent paths equals the number of vertices that have to be removed to cut all paths between *i* and *j*
→ measure for network robustness

2) Number of **edge-independent paths** between *i* and *j*

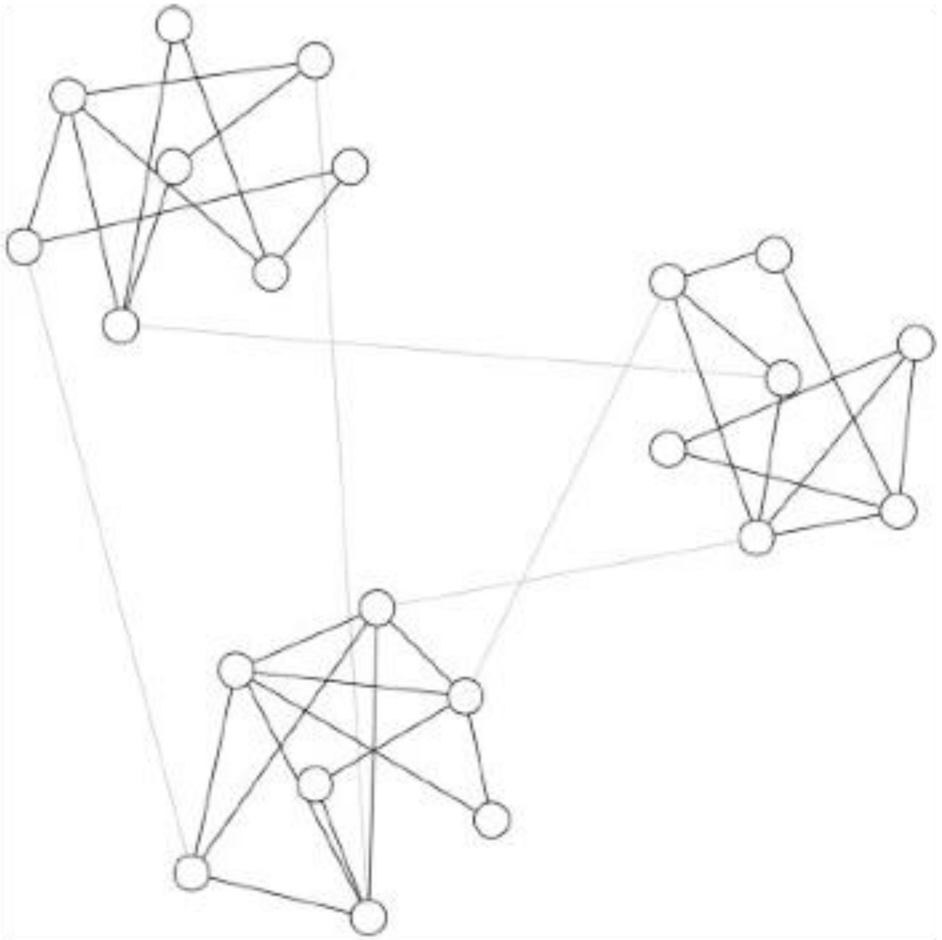
3) **Total number of paths** *L* between *i* and *j*
but $L = 0$ or ∞ → weight paths with their length α^L with $\alpha < 1$

Problem: vertices with a single link are separated from the communities

Vertex Betweenness

Freeman (1927): count on how many shortest paths a vertex is visited

For a graph *G = (V, E)* with *|V| = n*



Betweenness for vertex v:

$$C_B(\nu) = \frac{\sum_{s \neq \nu \neq t \in V} \sigma_{st}(\nu)}{(n-1)(n-2)}$$

σ_{st} (*v*) : shortest path including *v.*

There are *n* - 1 other vertices besides *v*.

They have shortest paths to *n* - 2 vertices.

-> Computing shortest paths takes O(n^2) operations

Alternative: **edge betweenness**

→ to how many shortest paths does this edge belong

Girvan-Newman Algorithm

Girvan, Newman, *PNAS* **99** (2002) 7821:

For a graph $G = (V, E)$ with $|V| = n$, $|E| = m$

1) Calculate **betweenness** for all *m* edges
2) **Remove** edge with highest betweenness
3) **Recalculate** betweenness for all affected nodes
4) **Repeat** from 2) until no more edge is left (at most *m* iterations)
5) Build up **tree** from *V* by reinserting vertices in reverse order

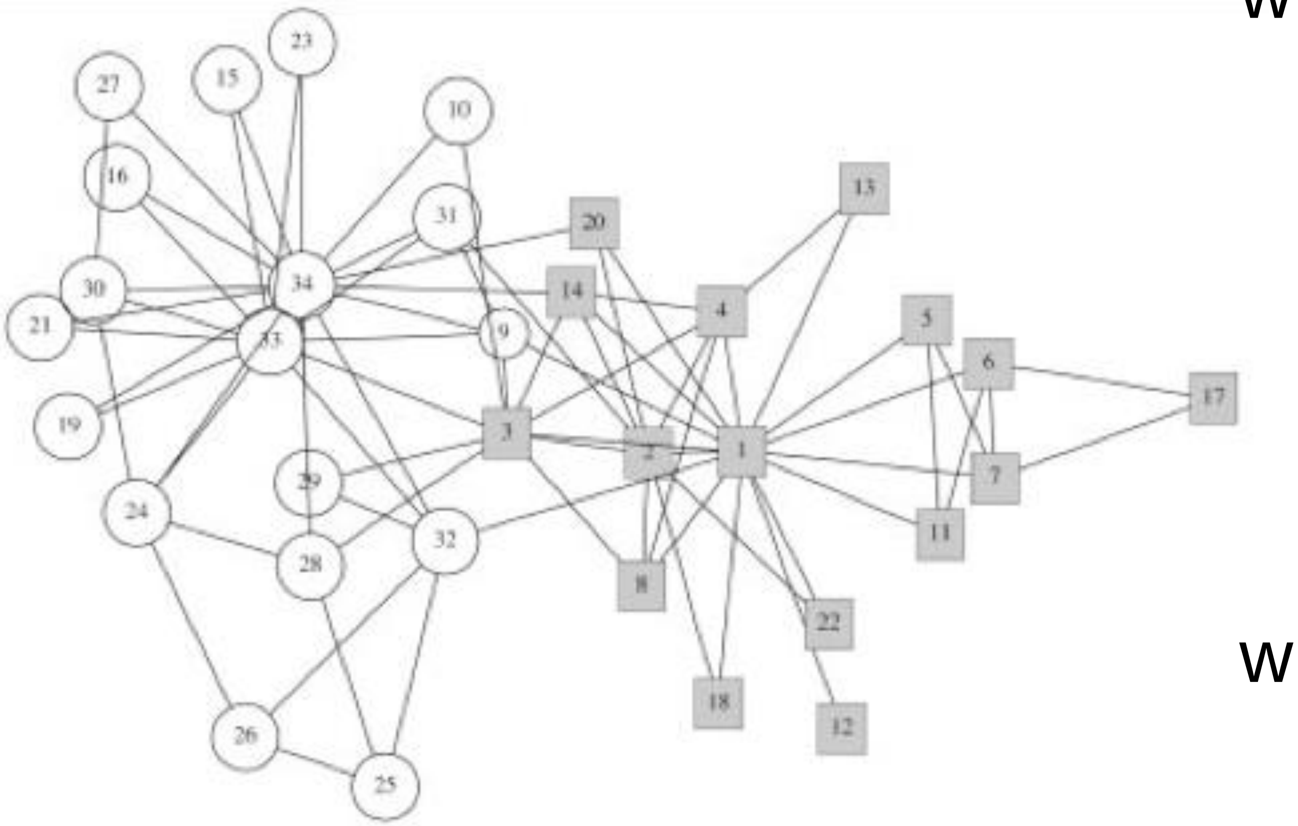
Works well, but **slow**: $O(mn^2) \approx O(n^3)$ for scale-free networks $(|E| = 2\ |V|)$

Reason for complexity: compute shortest paths (n^2) for *m* edges

→ recalculating a **global** property is expensive for larger networks

Zachary's Karate Club

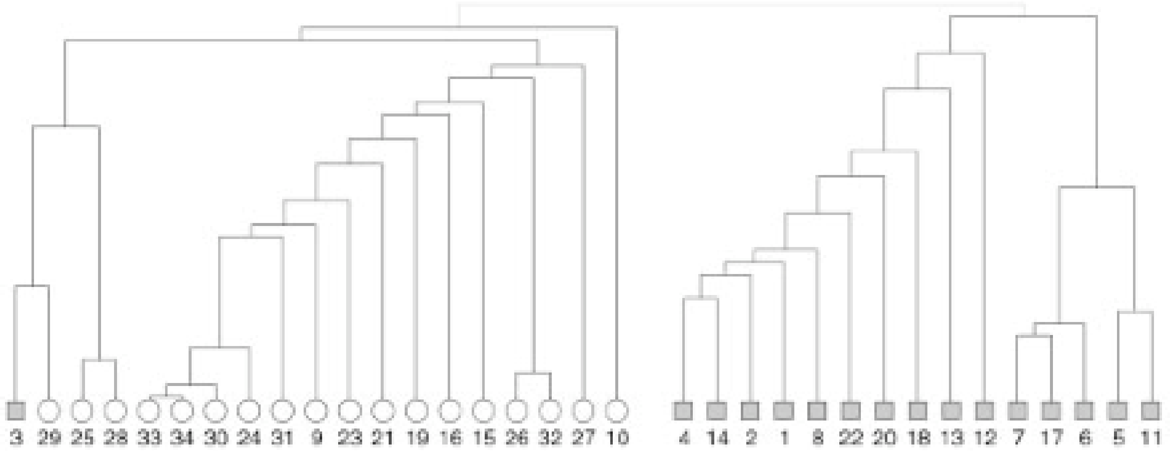
- observed friendship relations of 34 members over two years
- correlate fractions at break-up with calculated communities



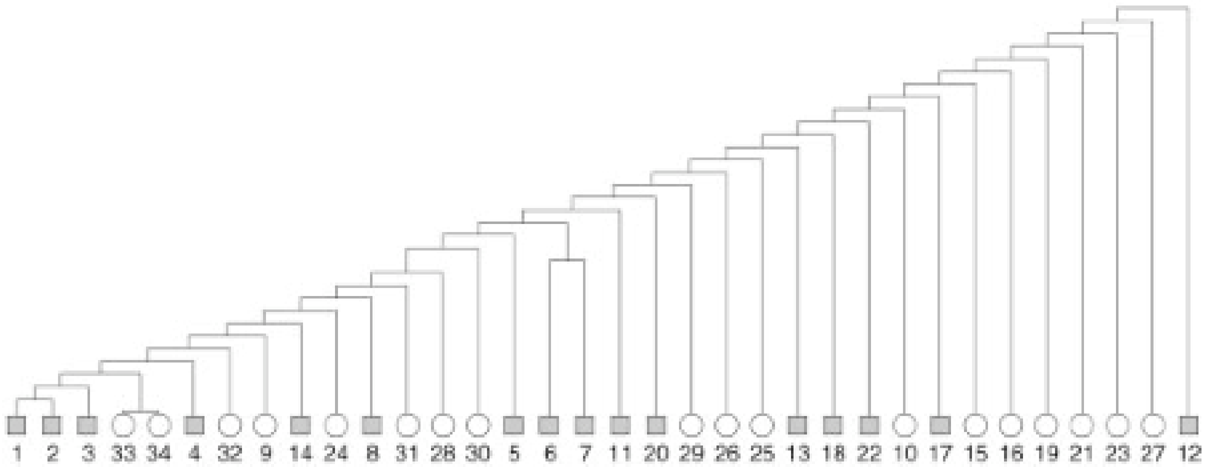
administrator's fraction

instructor's fraction

with edge betweenness:

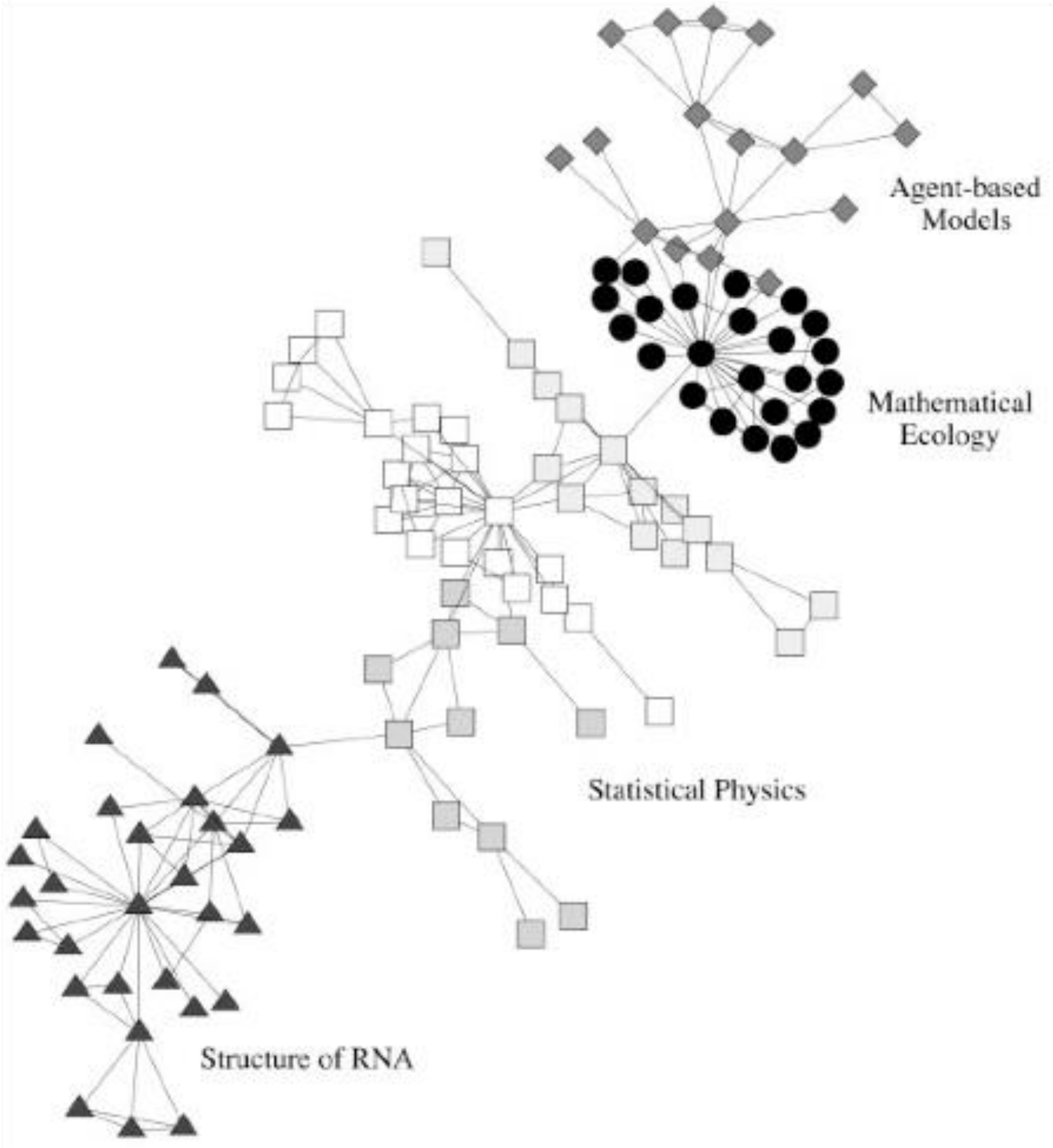


with number of edge-independent paths:



Girvan, Newman, *PNAS* **99** (2002) 7821

Collaboration Network



Vertices: scientists at the Santa Fe Institute.

Edge: two authors have co-authored a joint paper.

Show is the largest component of the Santa Fe Institute collaboration network.

The primary divisions detected by the GN algorithm are indicated by different vertex shapes.

Girvan, Newman, *PNAS* **99** (2002) 7821

Determining Communities Faster

Radicchi et al, *PNAS* **101** (2004) 2658:

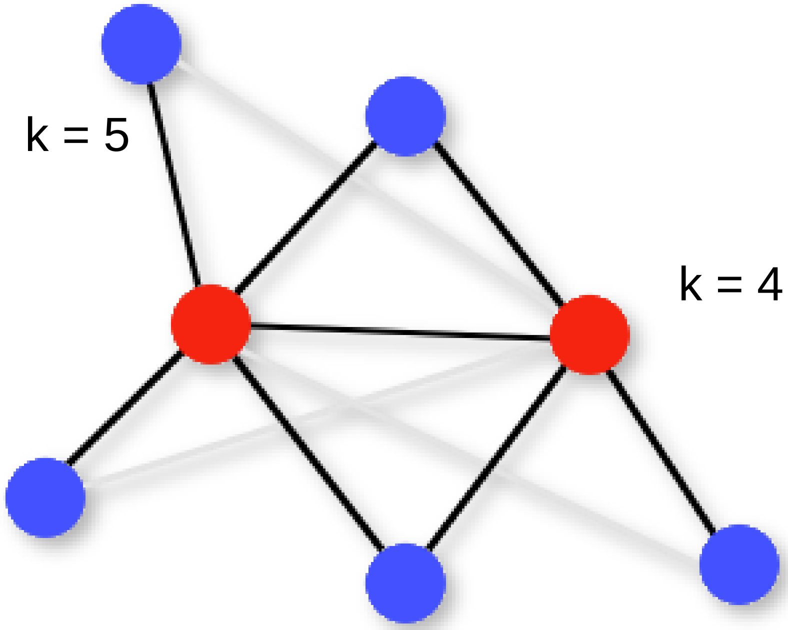
Determine edge weights via **edge-clustering coefficient**
→ local measure
→ much faster, esp. for large networks

Modified edge-clustering coefficient:
→ fraction of potential triangles
with edge between *i* and *j*

$$C_{i,j}^{(3)} = \frac{z_{i,j}^{(3)} + 1}{\min[(k_i - 1), (k_j - 1)]}$$

Here, $z_{i,j}^{(3)}$ is the number of triangles,
k_i and k_j are the degrees of nodes *i* and *j*.

Note: "+ 1" to remove degeneracy for $z_{i,j}^{(3)} = 0$



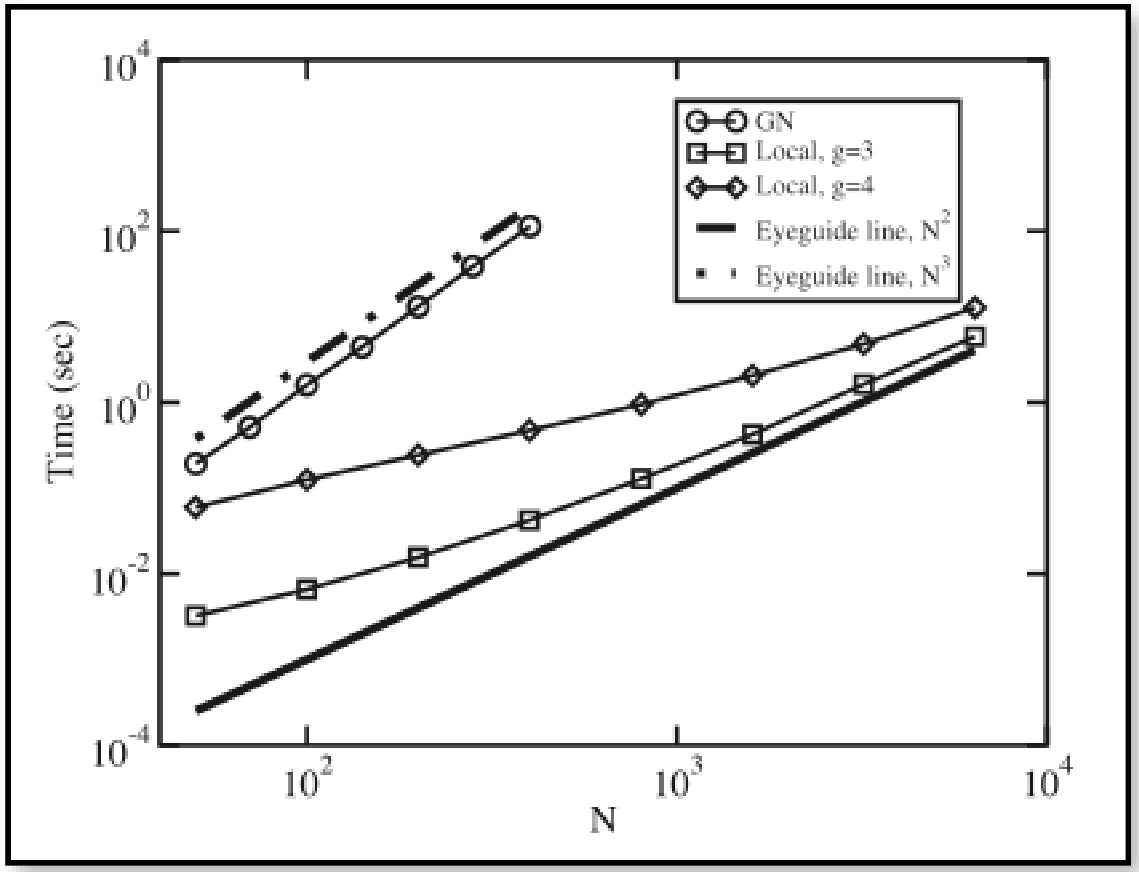
$C^{(3)} = (2+1) / 3 = 1$

Algorithm works exactly like GN-algorithm except that at each iteration, the edge is removed with smallest $C_{i,j}^{(3)}$

Performance

Instead of triangles: **cycles** of higher order *g*
→ continuous transition to a global measure

$$C_{i,j}^{(g)} = \frac{z_{i,j}^{(g)} + 1}{s_{i,j}^{(g)}}$$

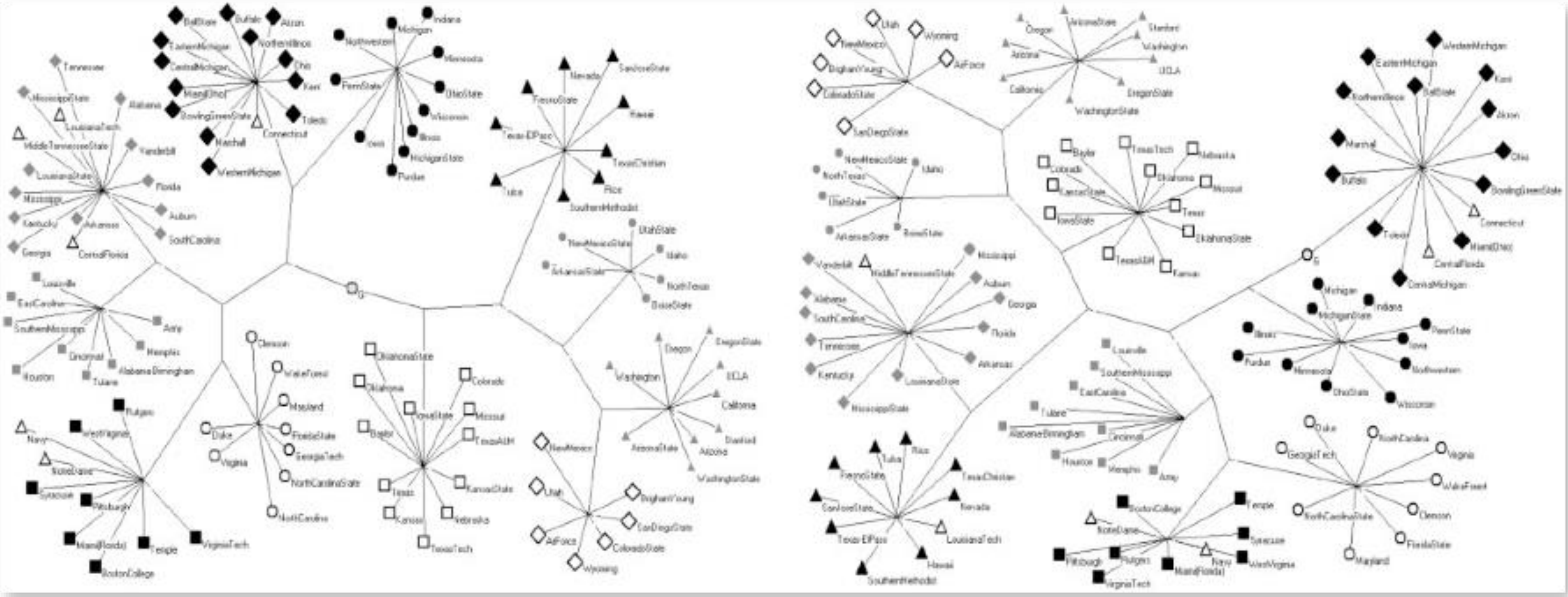


Radicchi *et al*-algorithm: O(N^2) for large networks

Radicchi et al, *PNAS* **101** (2004) 2658:

Comparison of algorithms

Data set: football teams from US colleges; different symbols = different conferences, teams played ca. 7 intraconference games and 4 inter-conference games in 2000 season.



Girven-Newman algorithm

Radicchi with *g* = 4

→ very similar communities

Strong Communities

"Community := subgraph with more interactions inside than to the outside"

A subgraph *V* is a **community** in a…

…**strong** sense when:

$$k_i^{in}(V) > k_i^{out}(V) \quad \forall i \in V$$

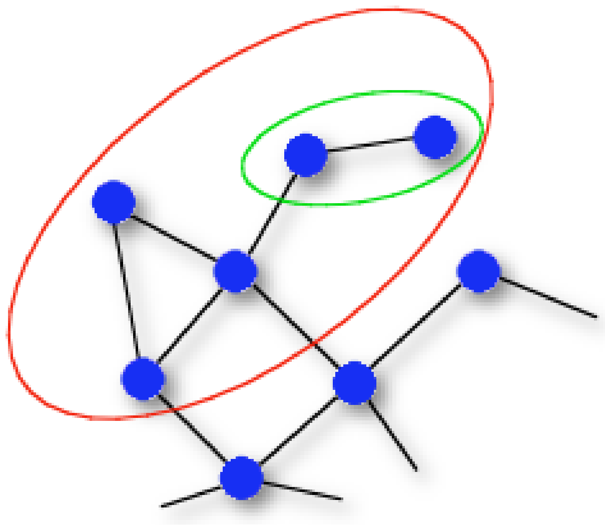
→ Check every node individually

…**weak** sense when:

$$\sum_{i \in V} k_i^{in}(V) > \sum_{i \in V} k_i^{out}(V)$$

→ allow for borderline nodes

Radicchi et al, *PNAS* **101** (2004) 2658



- Σ k_{in} = 2, Σ k_{out} = 1
  {k_{in}, k_{out}} = {**1,1**}, {1,0}
  → community in a weak sense

- Σ k_{in} = 10, Σ k_{out} = 2
  {k_{in}, k_{out}} = {2,1}, {2, 0}, {3, 1}, {2,0}, {1,0}
  → community in a strong and weak sense