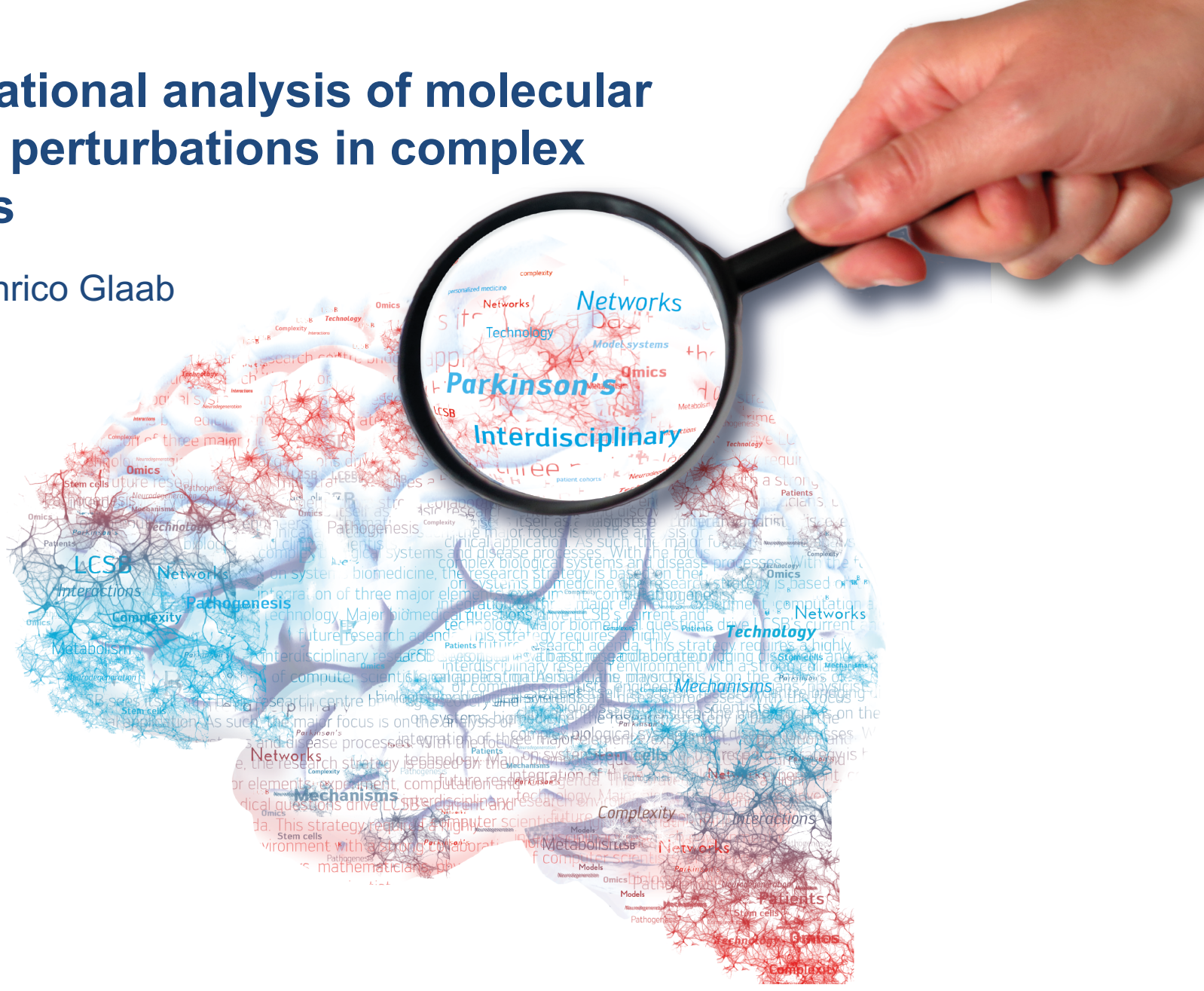
Computational analysis of molecular network perturbations in complex diseases

Speaker: Enrico Glaab

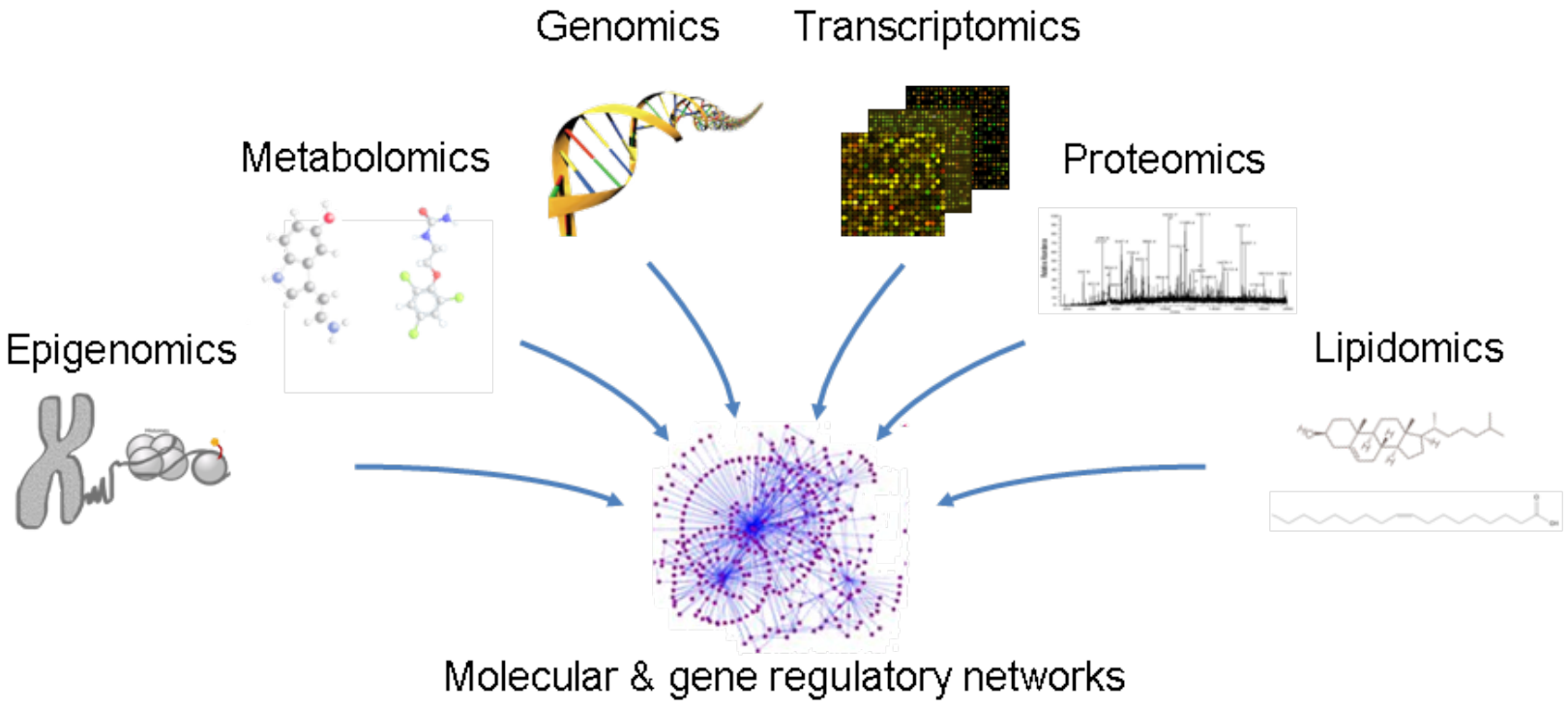


Luxembourg Centre for Systems Biomedicine (LCSB)

Luxembourg Centre
for Systems Biomedicine

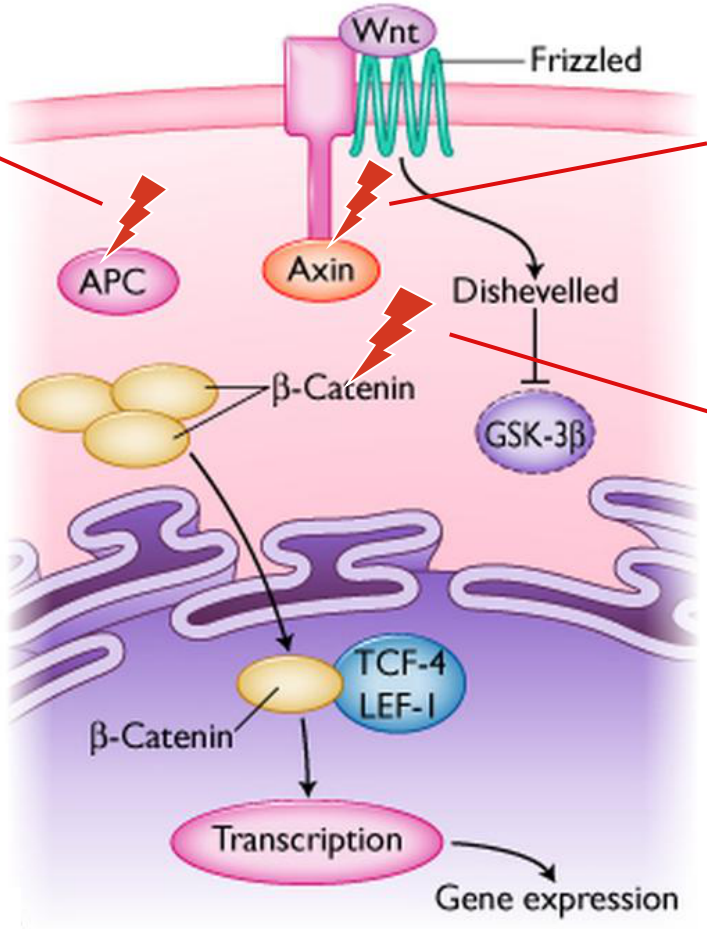
Group focus

Main goal: Interpret molecular changes in complex disorders by integrating diverse data types using network analyses



Motivation: Diseases as network perturbations

APC mutations
→ Colorectal cancer



AXIN1/2 mutations
→ Colorectal cancer

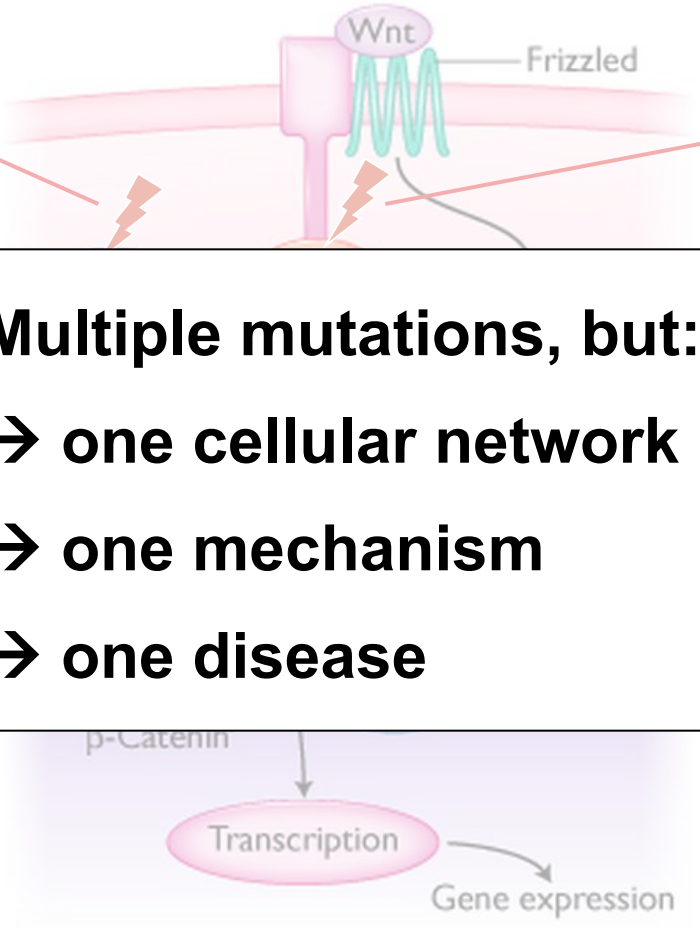
ß-Catenin mutations
→ Colorectal cancer

Wnt/β-catenin signaling pathway
(= affected by disease-related mutations)

Motivation: Diseases as network perturbations

APC mutations
→ Colorectal cancer

AXIN1/2 mutations
→ Colorectal cancer



Multiple mutations, but:

→ one cellular network

→ one mechanism

→ one disease

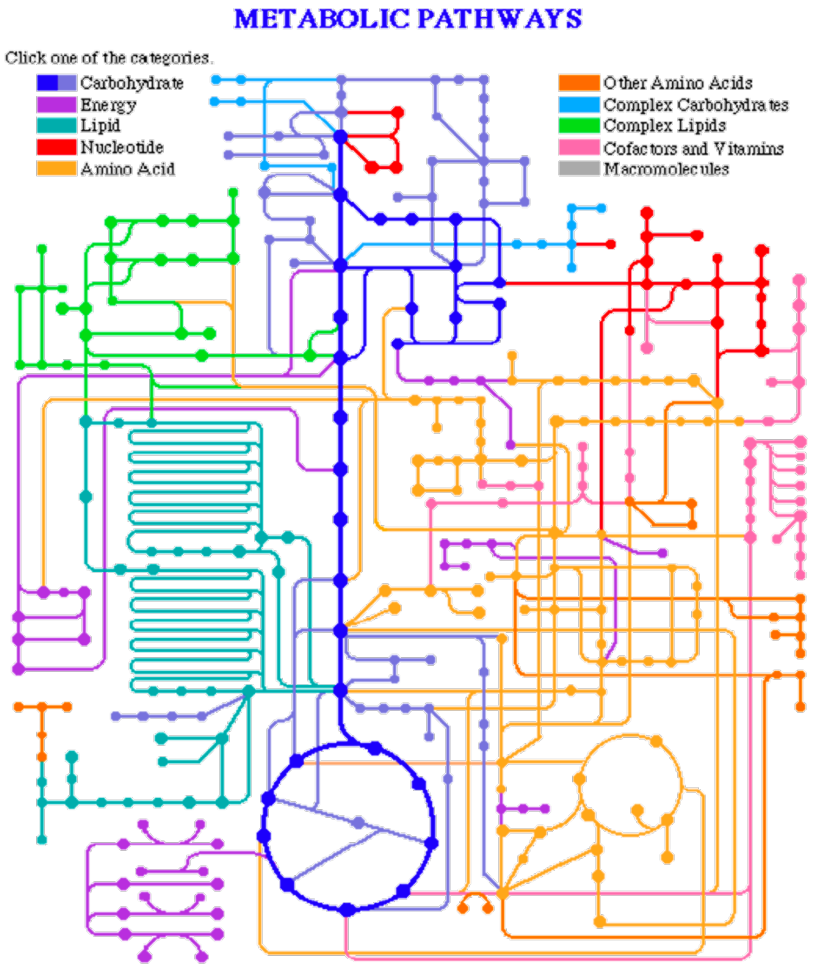
-Catenin mutations
→ Colorectal cancer

Wnt/β-catenin signaling pathway
(= affected by disease-related mutations)

Classical scientific approach: Reductionist Method

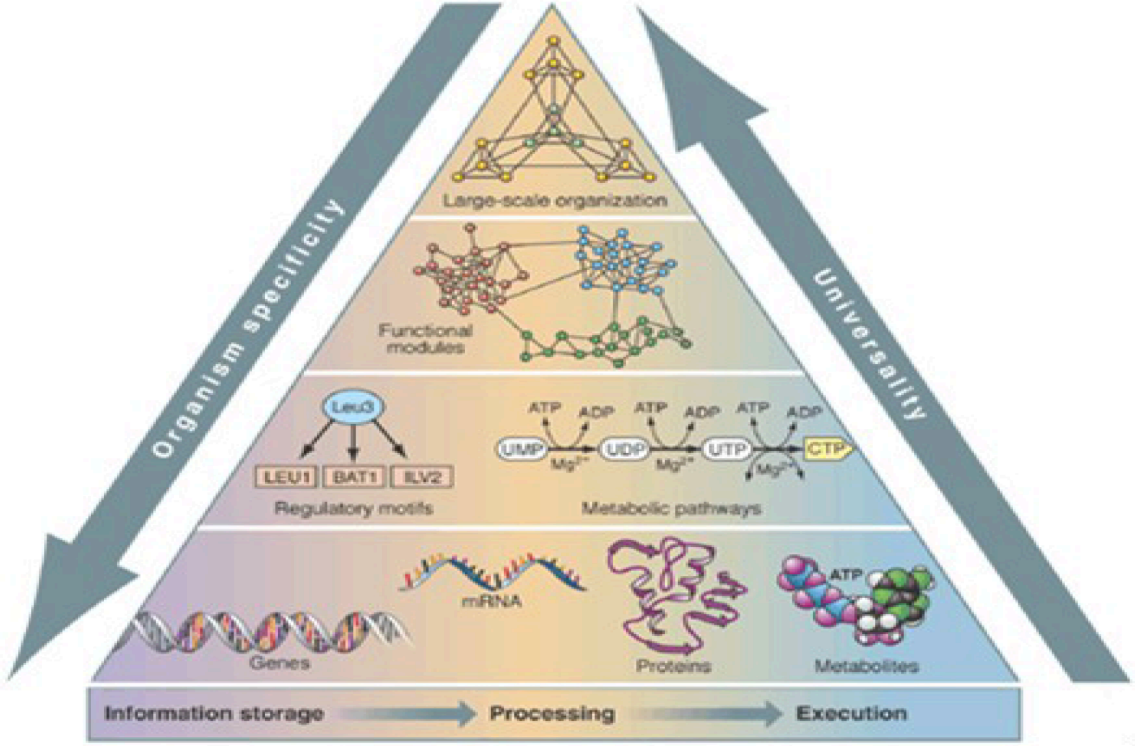
Reductionist Method:

- Hypotheses are specific and of narrow scope (local hypotheses)
- Understanding of an overall biological system (ecosystem, organism, cell) is supposed to be achieved by combining local insights
- however, the combinatorial nature of many biological systems challenges this method



Computational Systems Biology & Network Medicine

Systems biology: The study of an organism, viewed as an integrated and interacting network of genes, proteins and biochemical reactions.



Source: Oltvai & Barabasi, 2002

Computational Systems Biology (2)

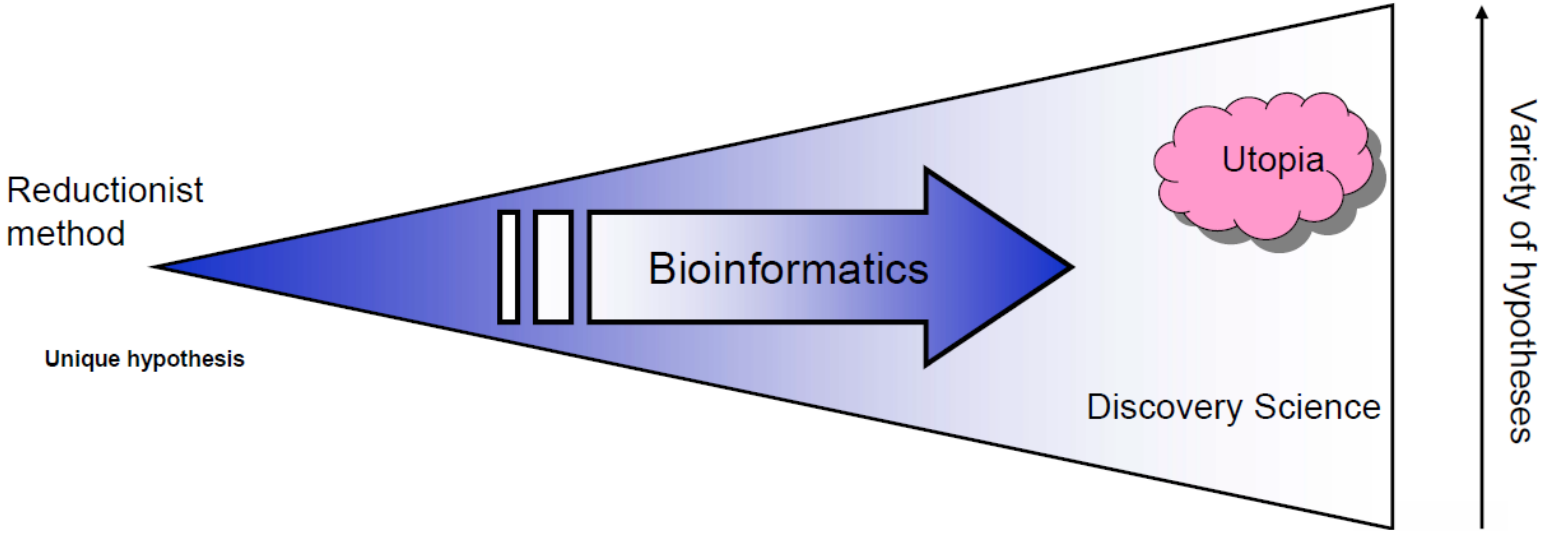
Two main driving forces:

- New high-throughput experimental profiling approaches enable organism-wide data collection:
  - Genomics → whole-genome sequencing
  - Transcriptomics → DNA chips / RNAseq
  - Interactomics → High-throughput Y2H screens

- Modern computers enable system-wide bioinformatics analyses for generating new valid or plausible hypotheses, e.g.:
  - which (combinations of) gene variations cause a disease?
  - which drugs inhibit the activity of target proteins most effectively?

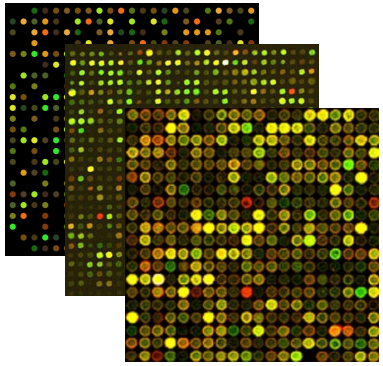
Computational Systems Biology (3)

- Bioinformatics is expected to drive progress in Systems Biology
- Some expectations may be too optimistic, but:
  - Bioinformatics can provide useful hypotheses for subsequent targeted experimental testing
  - Bioinformatics can help to select the most promising hypotheses from a larger set of plausible hypotheses

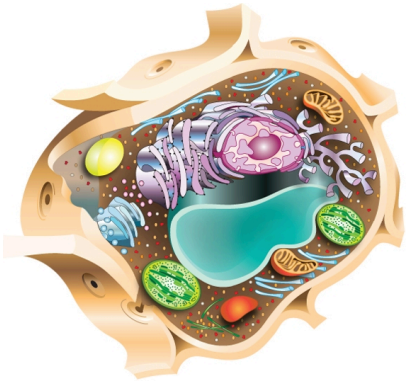


Challenge: From big data to biological function

Big data → **Statistics** → **Function**



Gene 1	4.3
Gene 2	–2.4
Gene 3	–3.1
Gene 4	1.8
...	...



Bottleneck:

Which changes are causal / secondary?

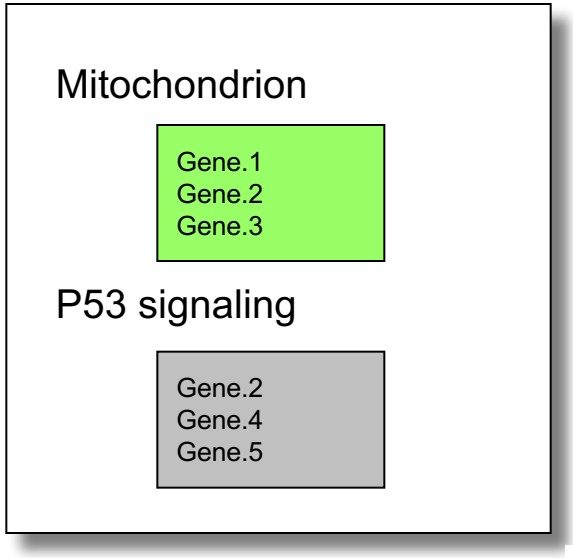
Which changes are correlative / predictive?

Which confounders modulate the system?

Which changes are disease-relevant / actionable?

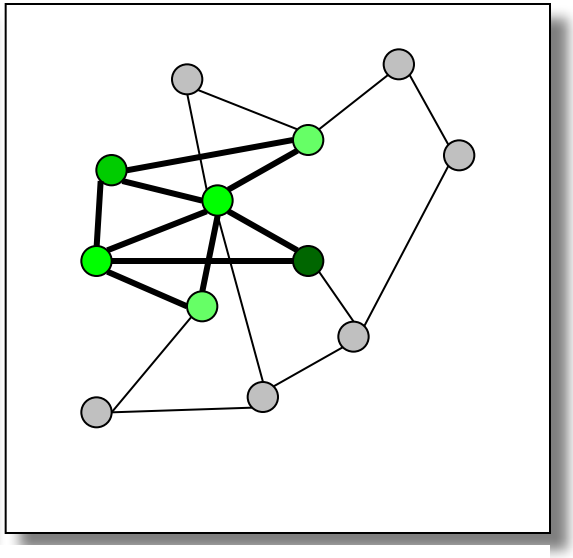
Representing and modeling cellular processes

GENE SETS



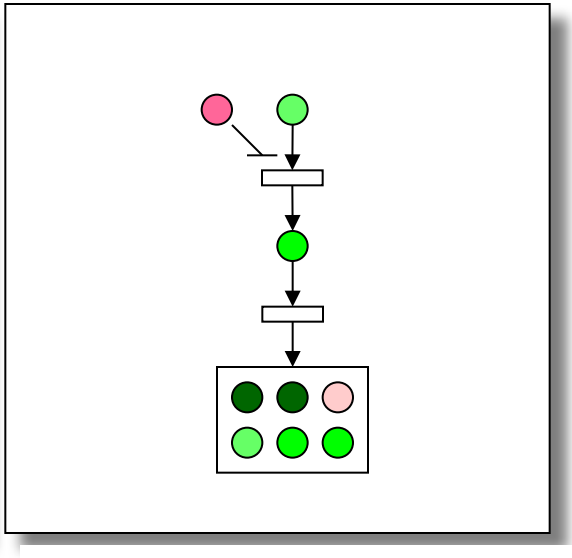
→ pure statistical scoring of enriched expression changes

NETWORKS



→ scoring of topological + expression criteria

PATHWAYS



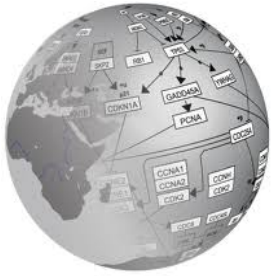
→ scoring of topology + expression changes + consistency criteria

Gene set / pathway resources

- Many public databases on functional gene sets and pathways available
- Both generic, multi-organism pathway collections covered and specialized collections (e.g. disease pathways: PD-Map, AlzMap)
- A total of over 10,000 public pathways available for the human species

BioCarta

KEGG Kyoto Encyclopedia of Genes and Genomes



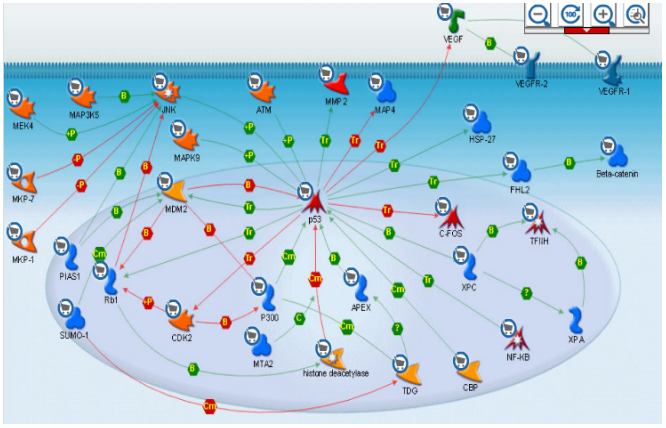
WikiPathways

GO the Gene Ontology

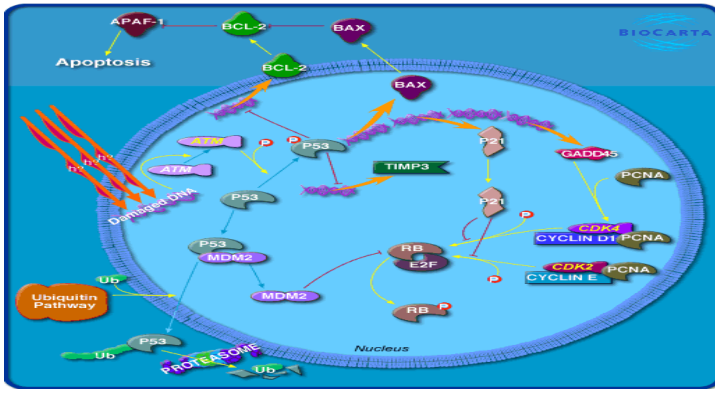
Reactome

Limitations of pathway databases

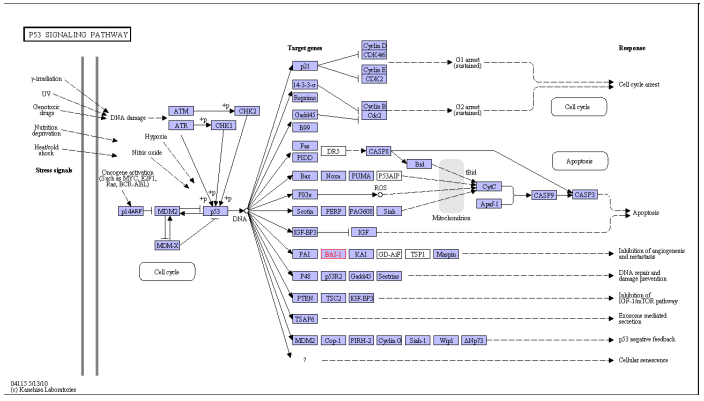
- manual curation → subjective decisions on pathway members & boundaries
- false-positive and false-negatives among molecular interactions
- database inconsistencies, e.g. “p53 signaling“:



Invitrogen iPath (p53 signaling)



BioCarta (p53 signaling)



KEGG (p53 signaling)

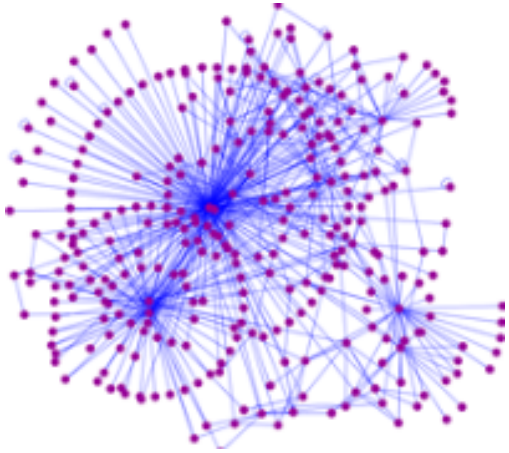
Improving pathway definitions using networks

- **Questions**: Can we make pathway definitions more objective? Can we improve existing pathways according to quantitative criteria?

- **Strategy**: Use genome-scale networks to redefine pathways:

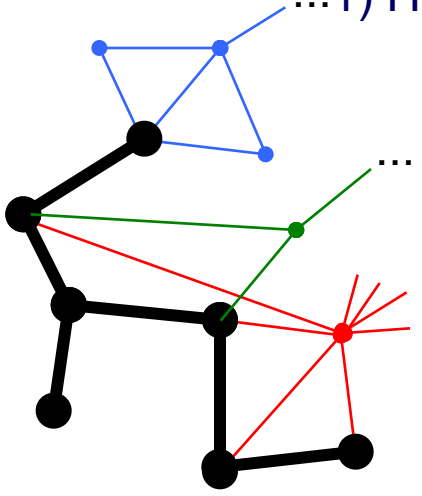
  - protein-protein interactions
  - genetic interactions
  - gene co-expression relations

  → large-scale, higher coverage, less biased
  → can also reveal communication between pathways (“cross-talk“)



Network-based pathway extension

Idea: Extend pathways by adding genes according to graph-theoretic criteria:



...1) High density of connections (“triangle-links“)

...2) High specificity of connections (more in than out)

...3) High coverage of connected pathway members

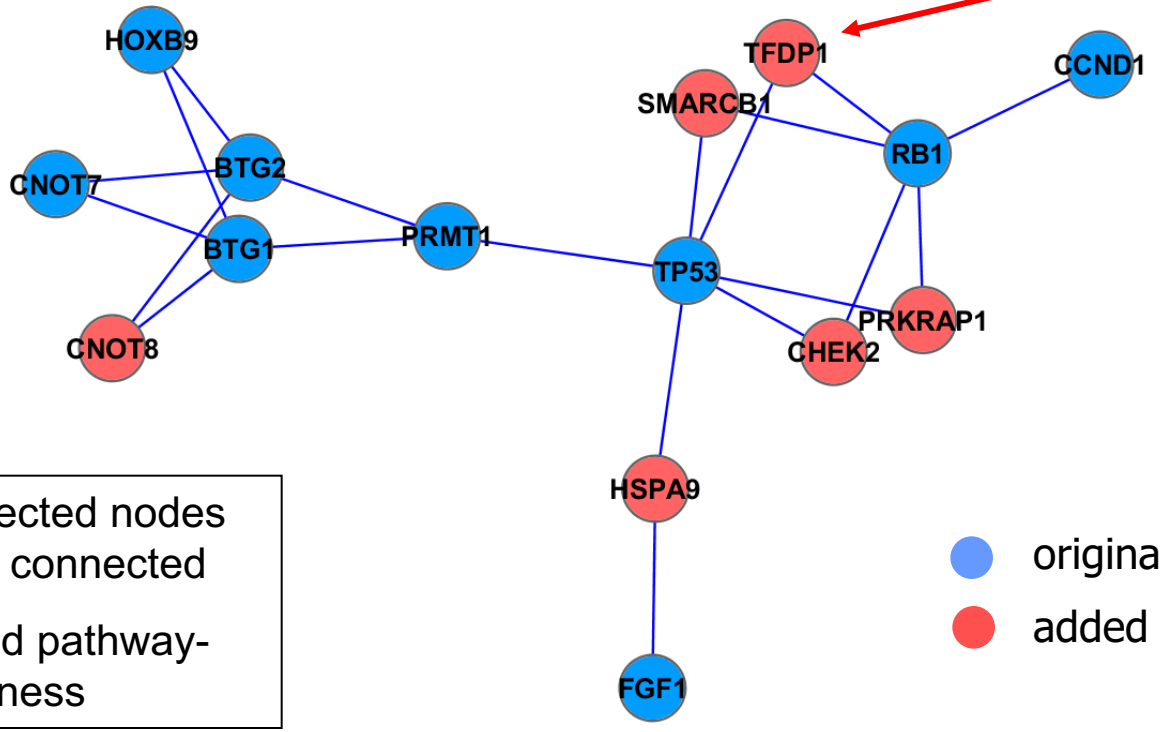
black = pathway members

red **blue** **green** = new candidate pathway members

Automated pathway extension: Example

Known cancer pathway: "*BTG family proteins and cell cycle regulation*" (BioCarta)

Mutations linked to colorectal cancer



→ Disconnected nodes become connected

→ increased pathway-compactness

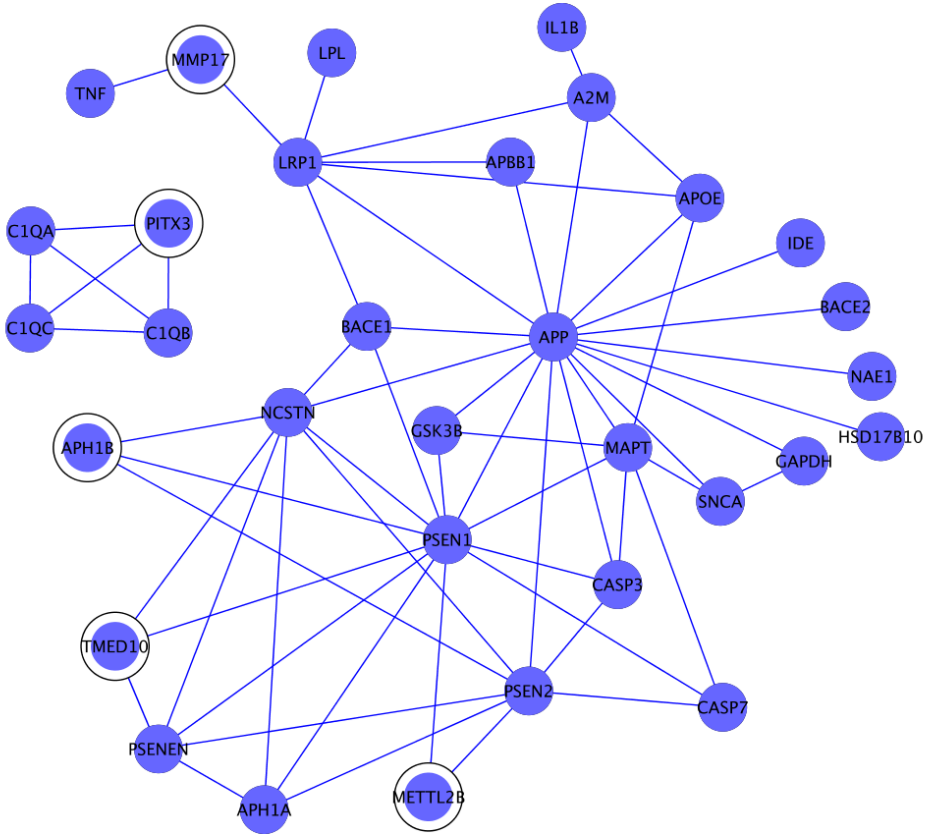
original pathway

added genes

Biological applications (1): Alzheimer‘s disease

- More than 20 proteins annotated in our molecular network
- 5 proteins added by the extension process (circled)
- 3 known to be associated with the disease
- 2 novel candidates: METTL2B, TMED10*

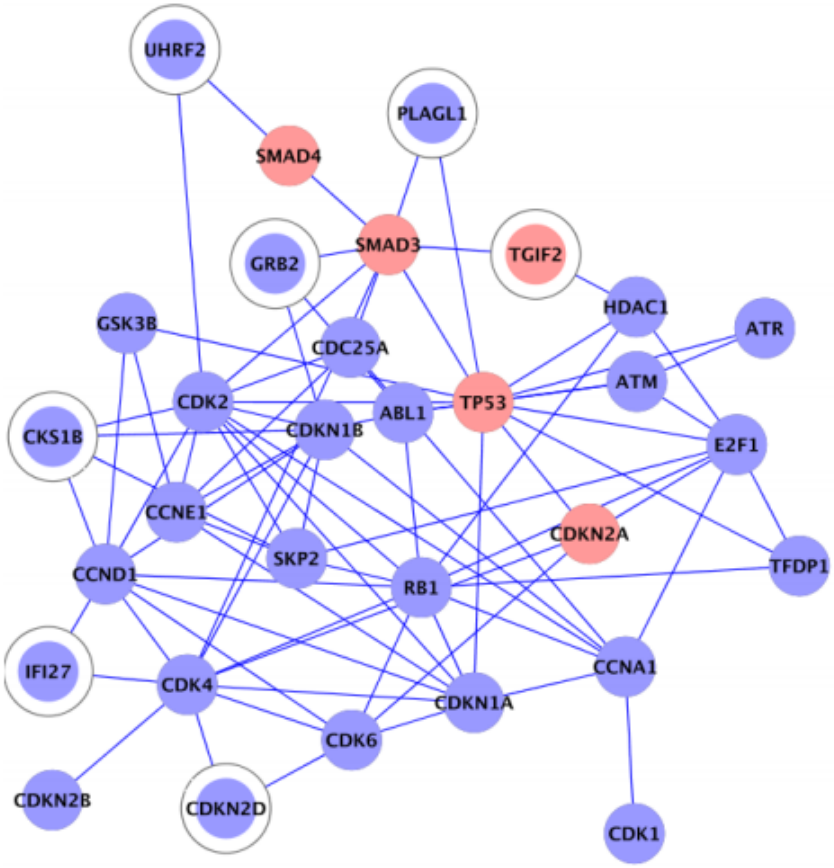
(*later confirmed: Shin et al., Autophagy, 2018)



KEGG Alzheimer disease pathway mapped on human protein interaction network

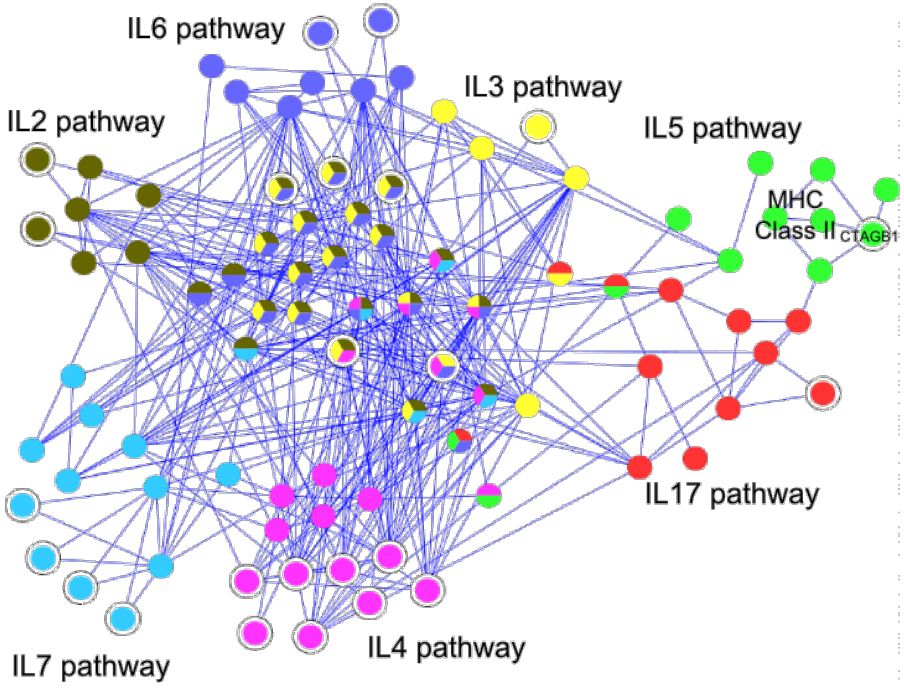
Biological applications (2): Pancreatic cancer

- "Cell cycle G1/S check point process" - extension procedure adds 7 proteins
- 6 of the added proteins are involved in cell cycle regulation
- the 7th (TGIF2) is known to be mutated in pancreatic cancer
- points to functional role of added proteins



Biological applications (3): Interleukin signaling

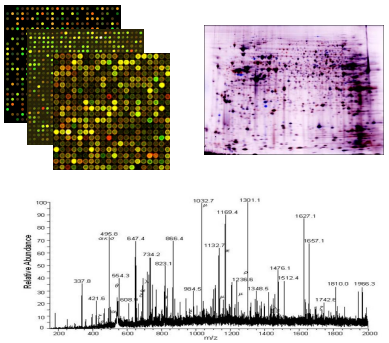
- Complex system of intracellular signaling cascades
- New putative pathway regulators identified
- New “cross-talk proteins” identified (associated with multiple pathways)

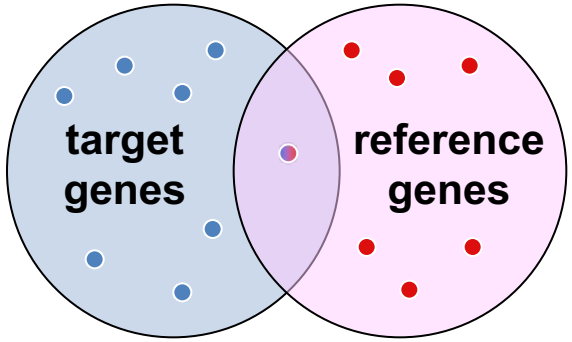


Two functions: pathway-regulation & pathway-communication?

Scoring of omics/pathway associations (EnrichNet)

Experimentally derived genes (**target genes**)





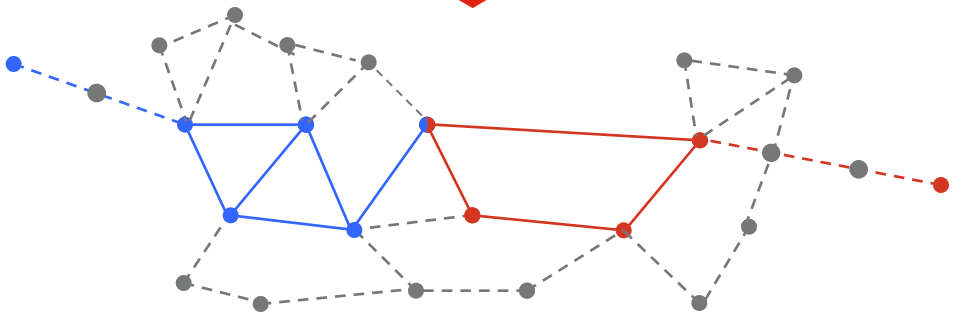
Pathway genes (**reference genes**)

KEGG Kyoto Encyclopedia of Genes and Genomes

WIKIPATHWAYS

GO the Gene Ontology

Reactome



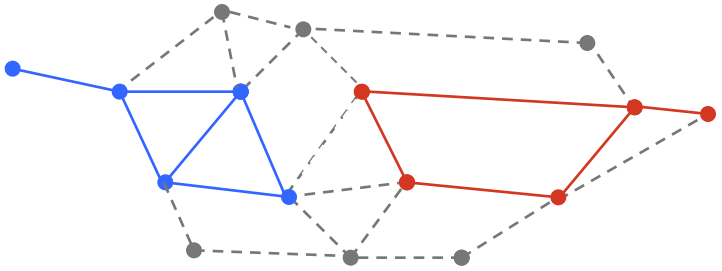
target node reference node other nodes

Network association scoring (EnrichNet)

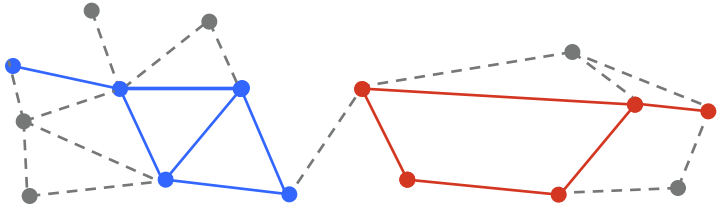
Scoring criteria:

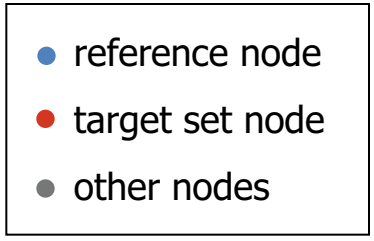
- distances between target and reference genes in network
- multiplicity of interactions between target and reference genes
- density of interactions between target and reference genes (compared to rest of the network)

Example 1: dense inter-connections



Example 2: sparse inter-connections





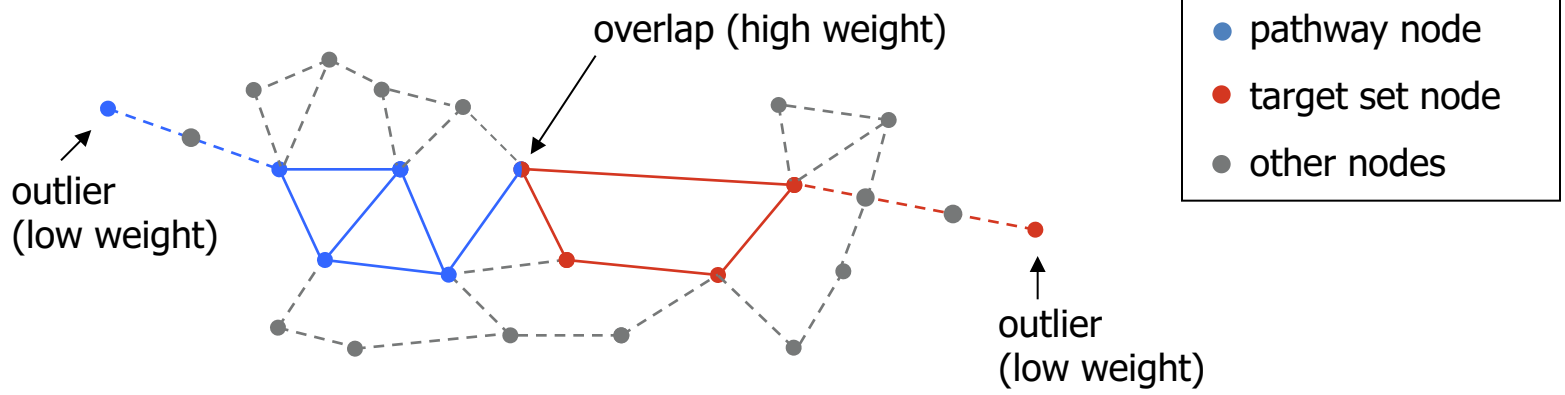
Network association scoring (EnrichNet)

Handling of overlapping genes and long distance outliers:

→ overlapping nodes and small distance node pairs → heigher weight

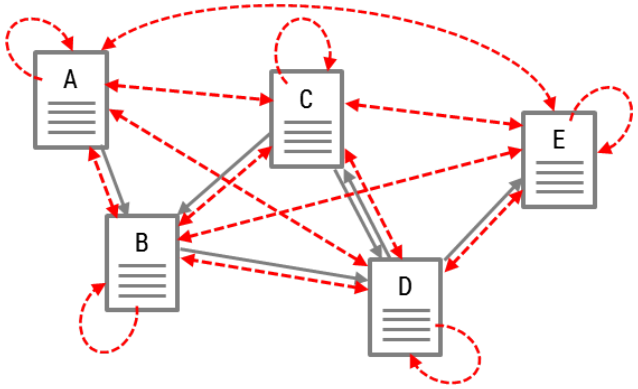
→ outlier nodes / large distance node pairs → lower weight

Example:

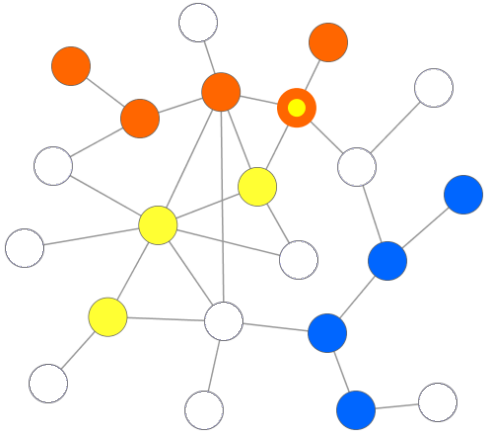


Network association scoring (EnrichNet)

Algorithm: Google‘s “Personalized Page Rank“



Transfer approach to molecular networks



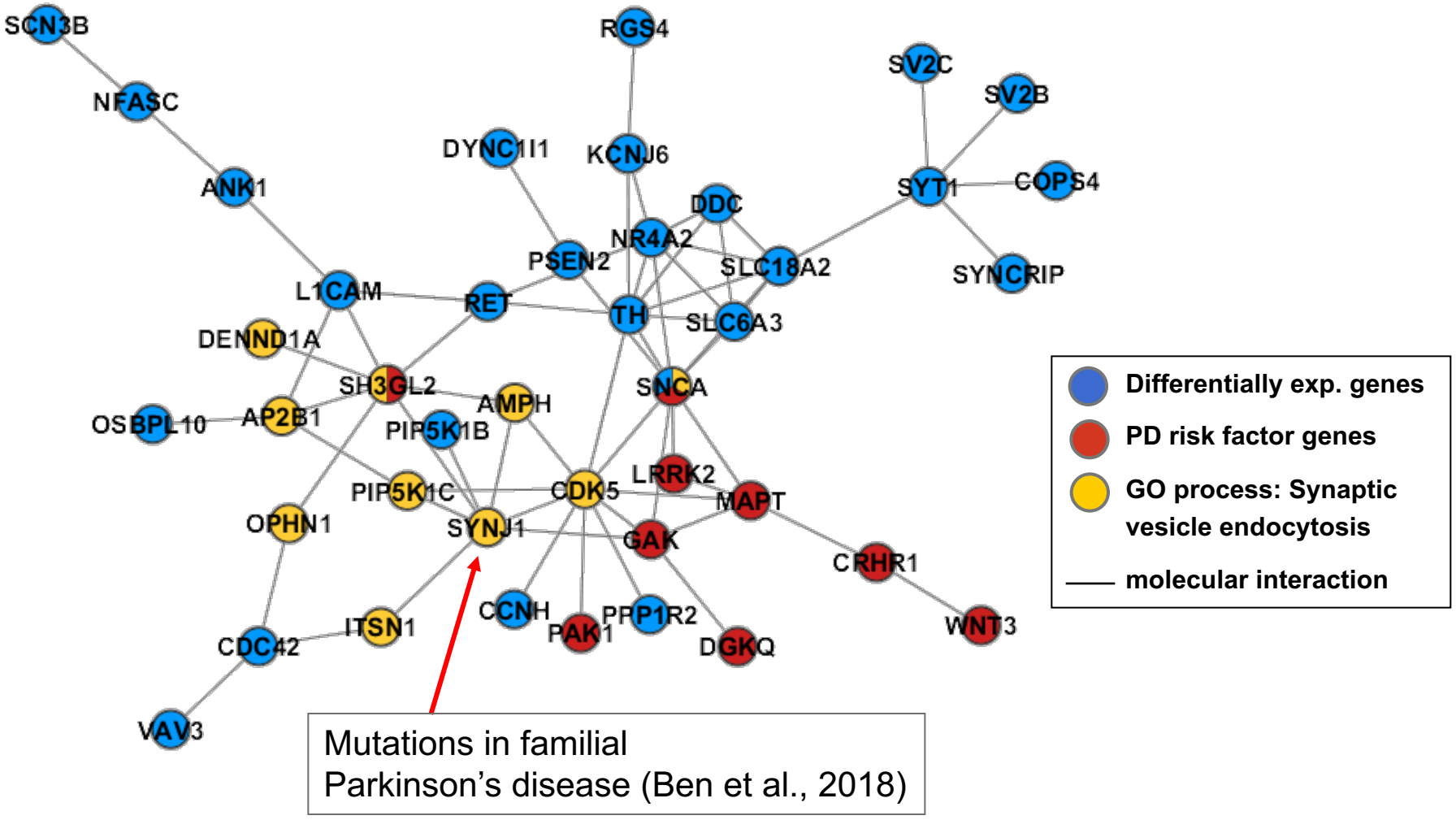
web-page
Real hyperlink
virtuel hyperlink

target genes
target/pathway overlap
pathway 1
pathway 2
interaction

Output: Relevance scores for each web-page (in relation to other web-pages)

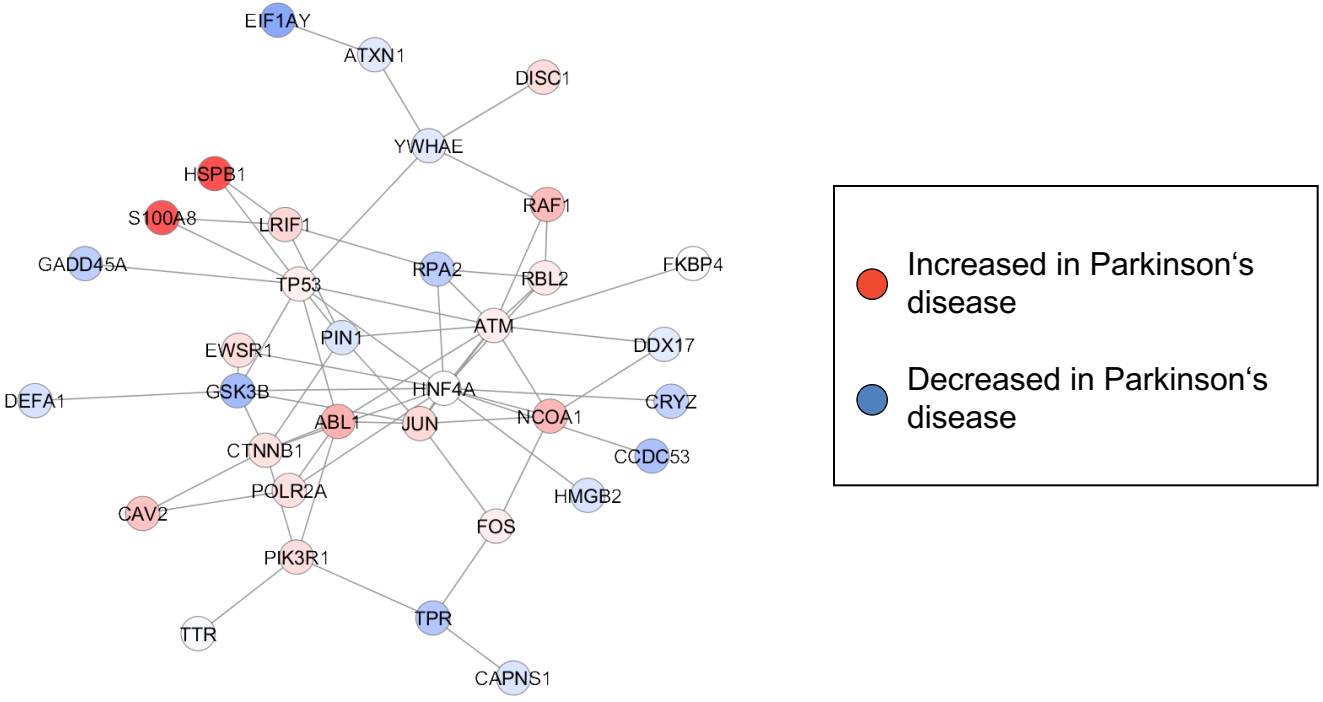
Output: Relevance scores for each pathway (in relation to a target set of genes)

Example Result: Parkinson‘s disease



Pathway-independent network analysis

Motivation: Disease perturbations may cluster in network regions outside of known pathways. Finding these clusters may lead to more robust biomarker models.



Question: How can we find clustered gene/protein groups efficiently, accounting for their diagnostic predictivity and connectedness in the network?

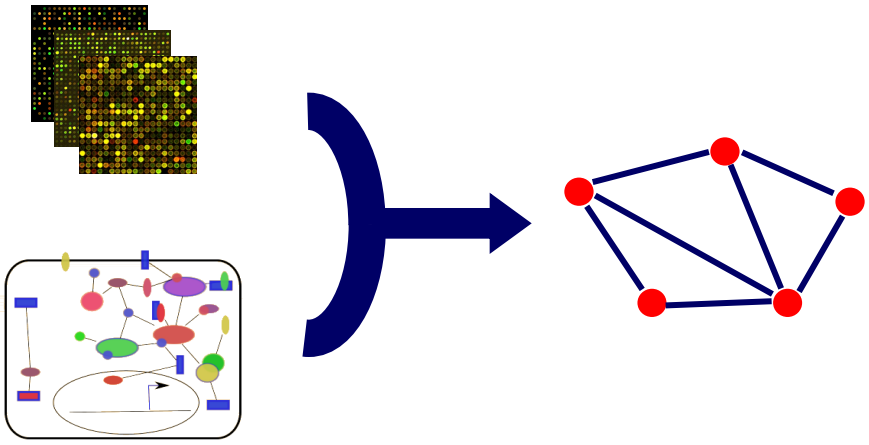
Network analysis software (GenePEN)

Input:

- Omics dataset (table with rows = genes/biomolecules, columns = samples)
- Class labels (e.g. “patient” or “control”)
- Table of interactions between the biomolecules (e.g. protein-protein interactions)

Output:

- A subset of discriminative biomolecules (rows) representing a connected component in the network that provides a predictive signature to classify new samples



Network analysis approach (GenePEN)

Idea: Find genes maximizing two quantities:

- the diagnostic prediction accuracy of their omics biomarker signature
- the connectedness of the selected genes in the network

→ formulate a corresponding scoring function (details not shown):

$$\min_{w} loss(w) + \lambda \cdot penalty(w)$$

loss-function (minimize error) trade-off parameter penalty-function (network grouping)

→ Minimize the function to find a good gene selection

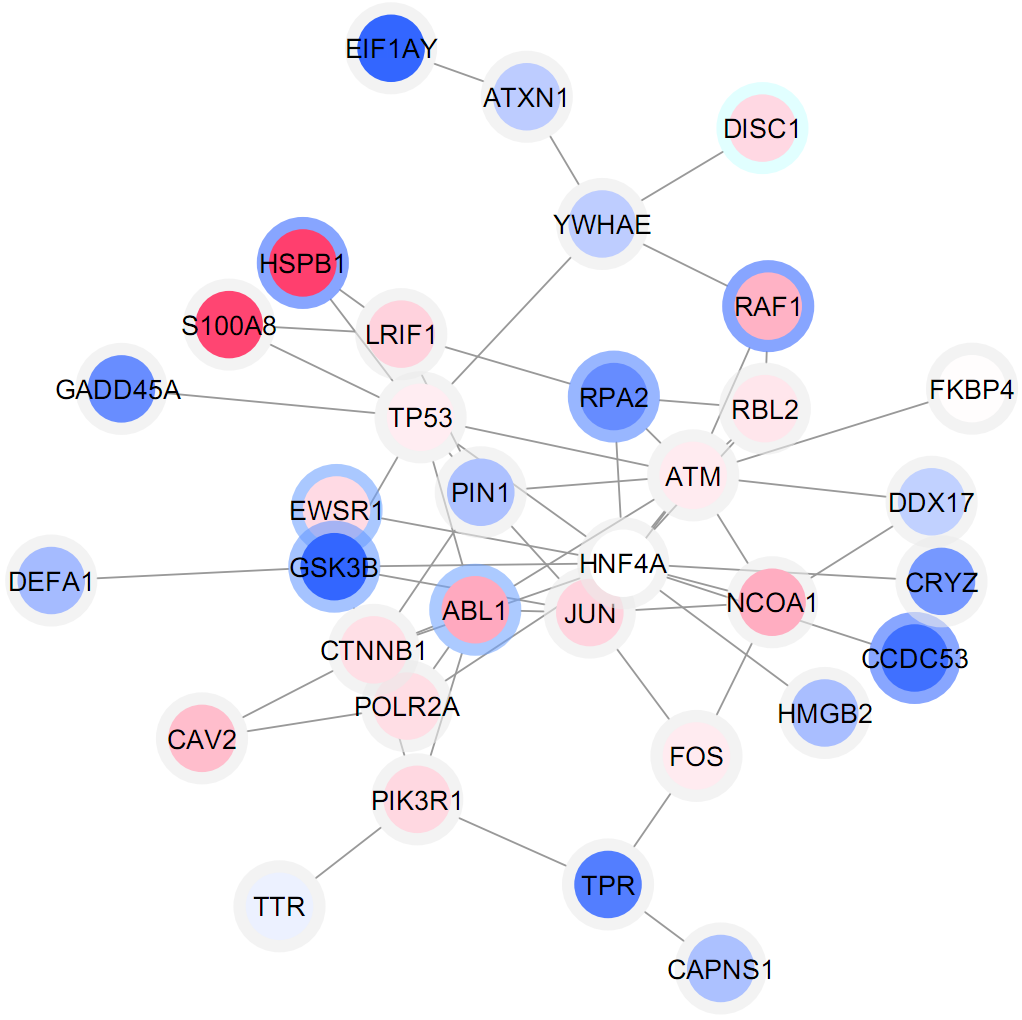
Application to Parkinson‘s disease (GenePEN)

Network alteration in Parkinson’s disease:

- red = over-expressed in PD
  blue = under-expressed in PD
  node borders = significance of alteration (from gray to blue with increasing significance)

- significant genes are over-represented in the sub-network (p = 0.01)

- GSK3B, the top significant gene in the sub-network, contains polymorphisms associated with Parkinson’s disease



Conclusion & Summary

- Why study diseases using network analysis?
  → to identify common mechanisms and combinatorial changes

- Three approaches presented:
  1) Automated network extension of disease pathways
  2) Scoring disease/pathway associations using network information
  3) Pathway-independent network analysis using machine learning

- **Future**: Time series data analysis of causal network perturbation

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