

Master Module
Proteinbiochemistry and Bioinformatics
November 2025

Protein interaction networks

Katja Luck, PhD

Some organizational information

- Questions throughout the lecture are welcome
- I will ask questions, too!
- Happy to receive feedback on the lecture and practical part

Outline

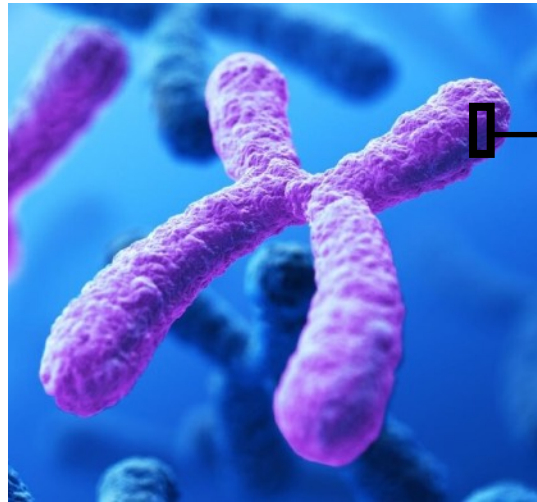
1. What are protein interactions?
2. Methods to detect protein interactions
3. Bioinformatic resources for protein interactions
4. Graph theoretical aspects of protein interaction networks
5. Visualizing and analyzing networks using Cytoscape

Master Module
Proteinbiochemistry and Bioinformatics
December 2024

Session: Protein interaction networks

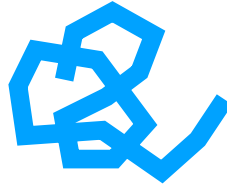
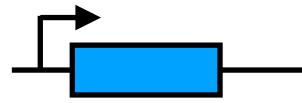
1. What are protein interactions?

Why do protein interactions matter?

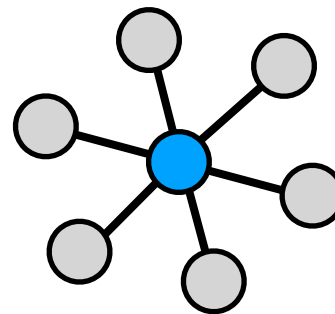


humanoriginproject.com

Gene X functions in Y - How?

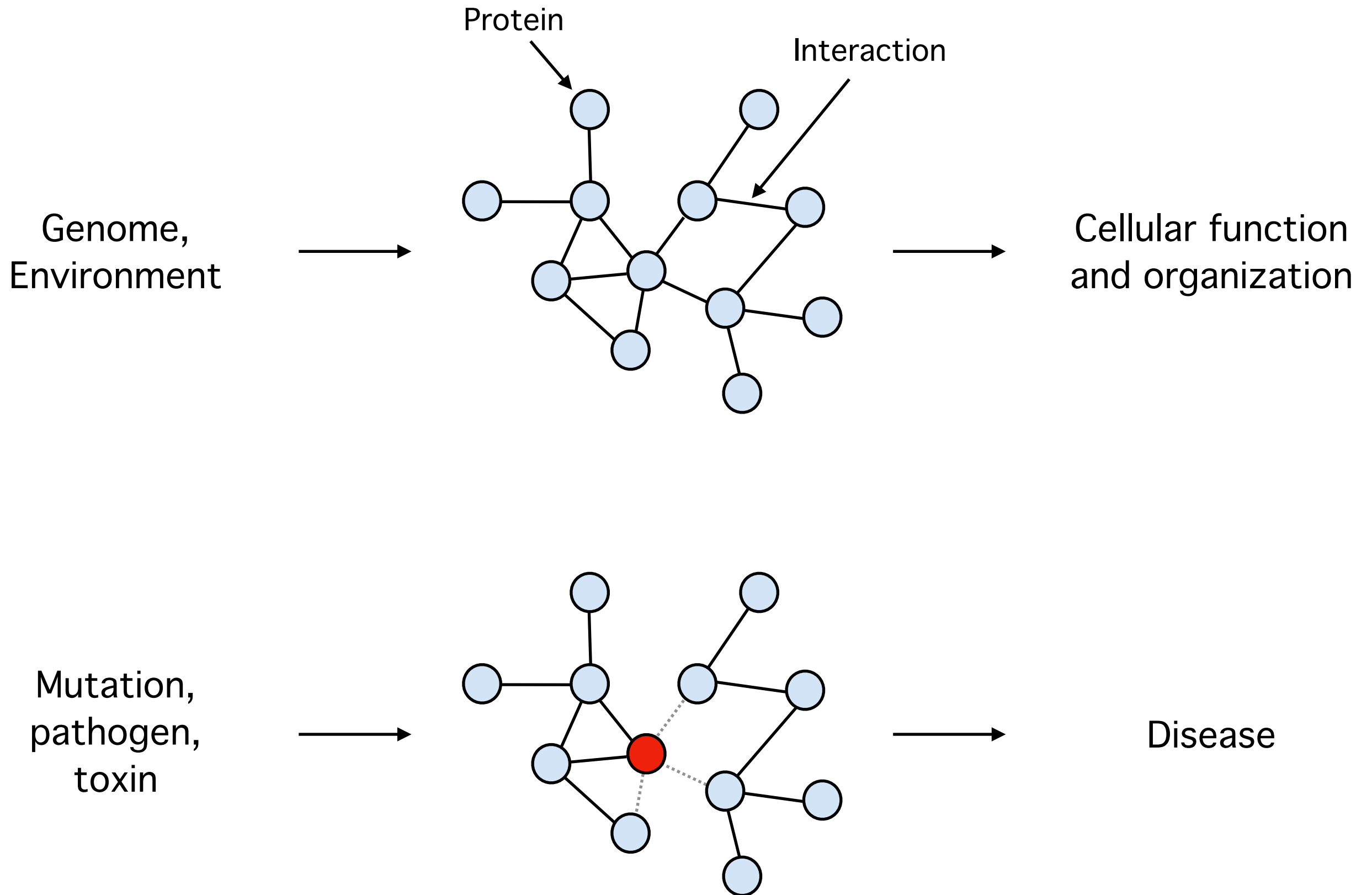


Protein X functions in Y - How?

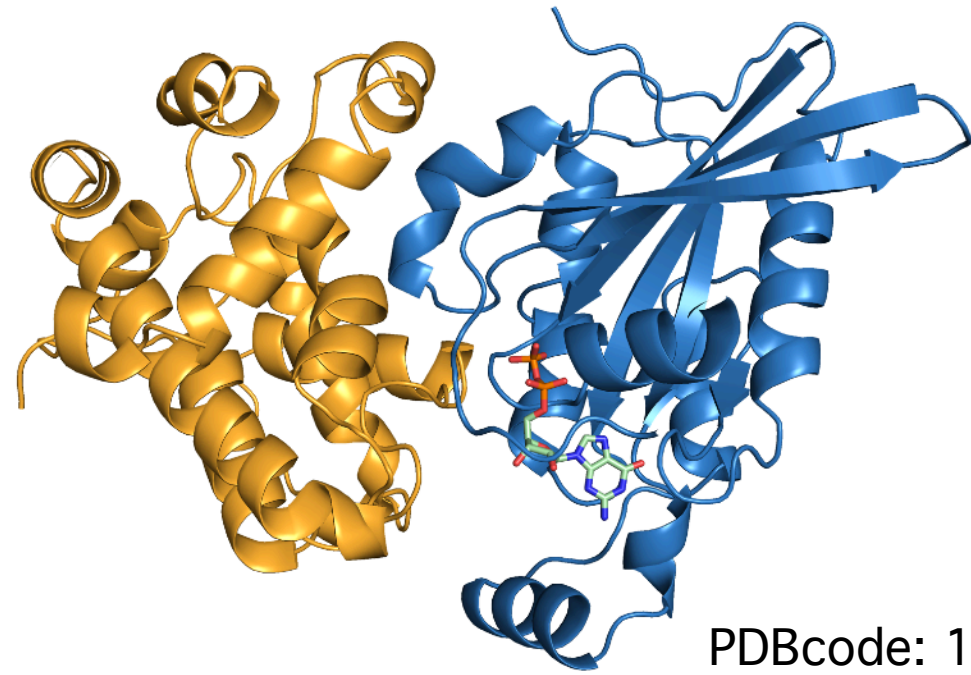
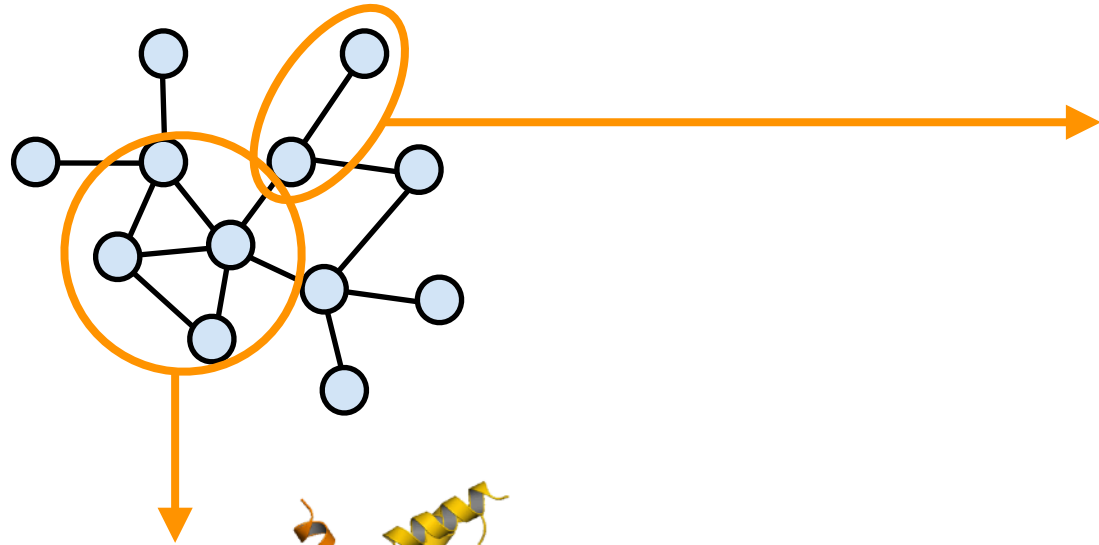


- Interactions mediate a gene's function
- Interactions inform on a gene's function

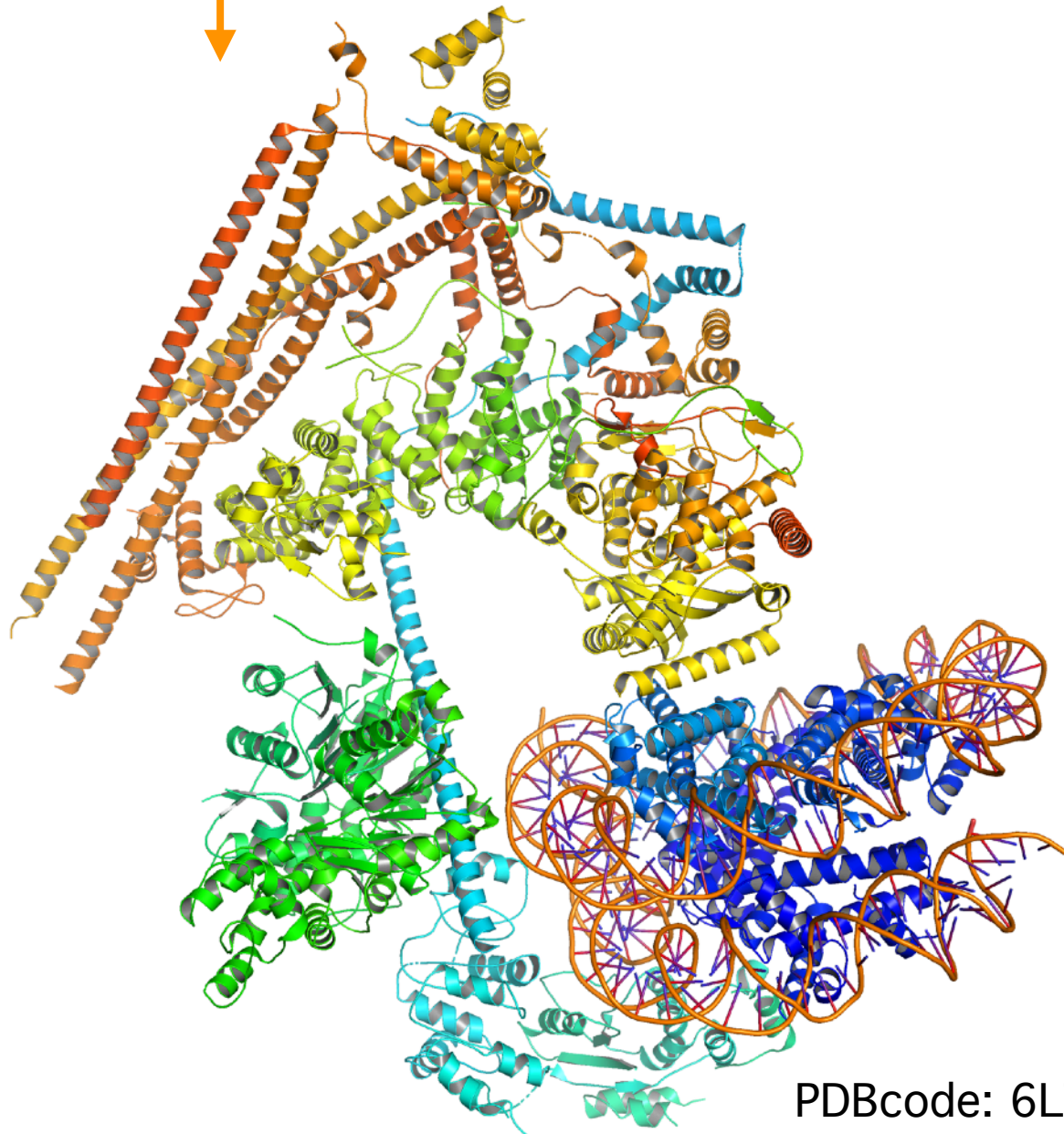
Protein interactions mediate cellular function



Protein interactions are complex

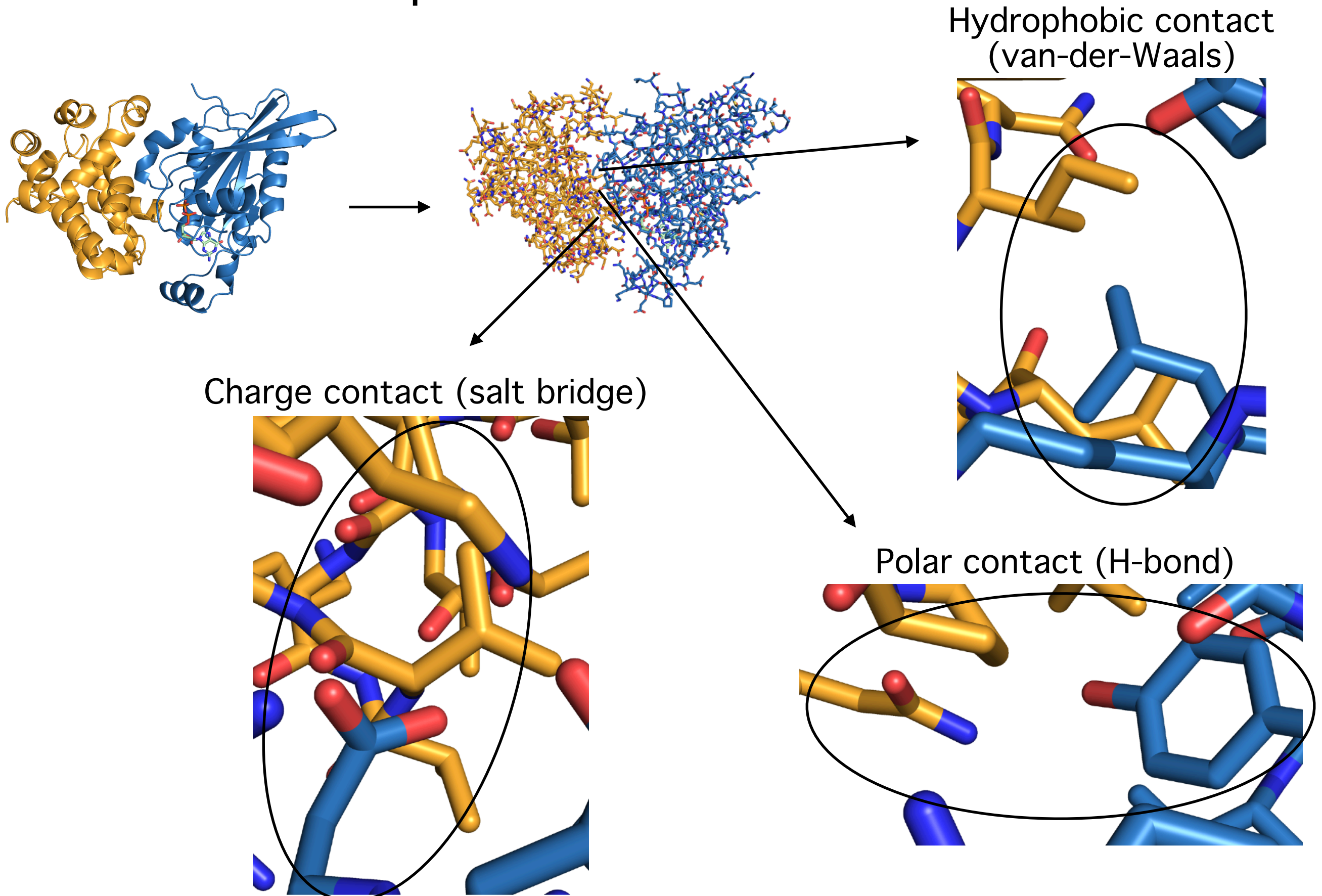


PDBcode: 1GRN



PDBcode: 6LTJ

Non-covalent contacts between amino acids mediate protein interactions



Protein interaction strength is expressed as
dissociation constant K_D

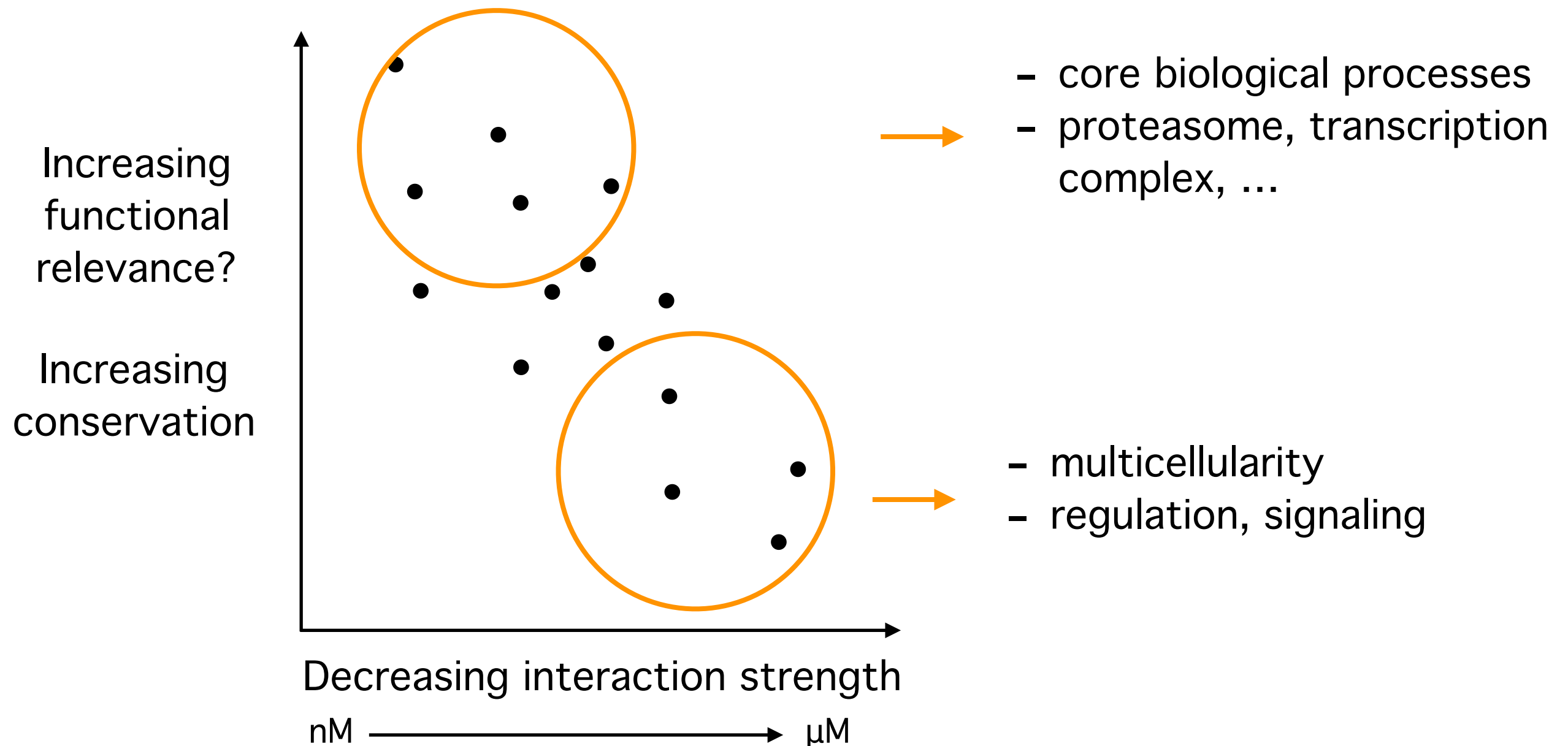


$$K_D = \frac{[A][B]}{[AB]}$$

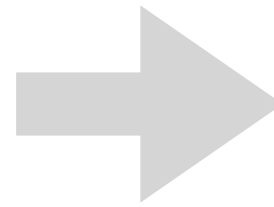
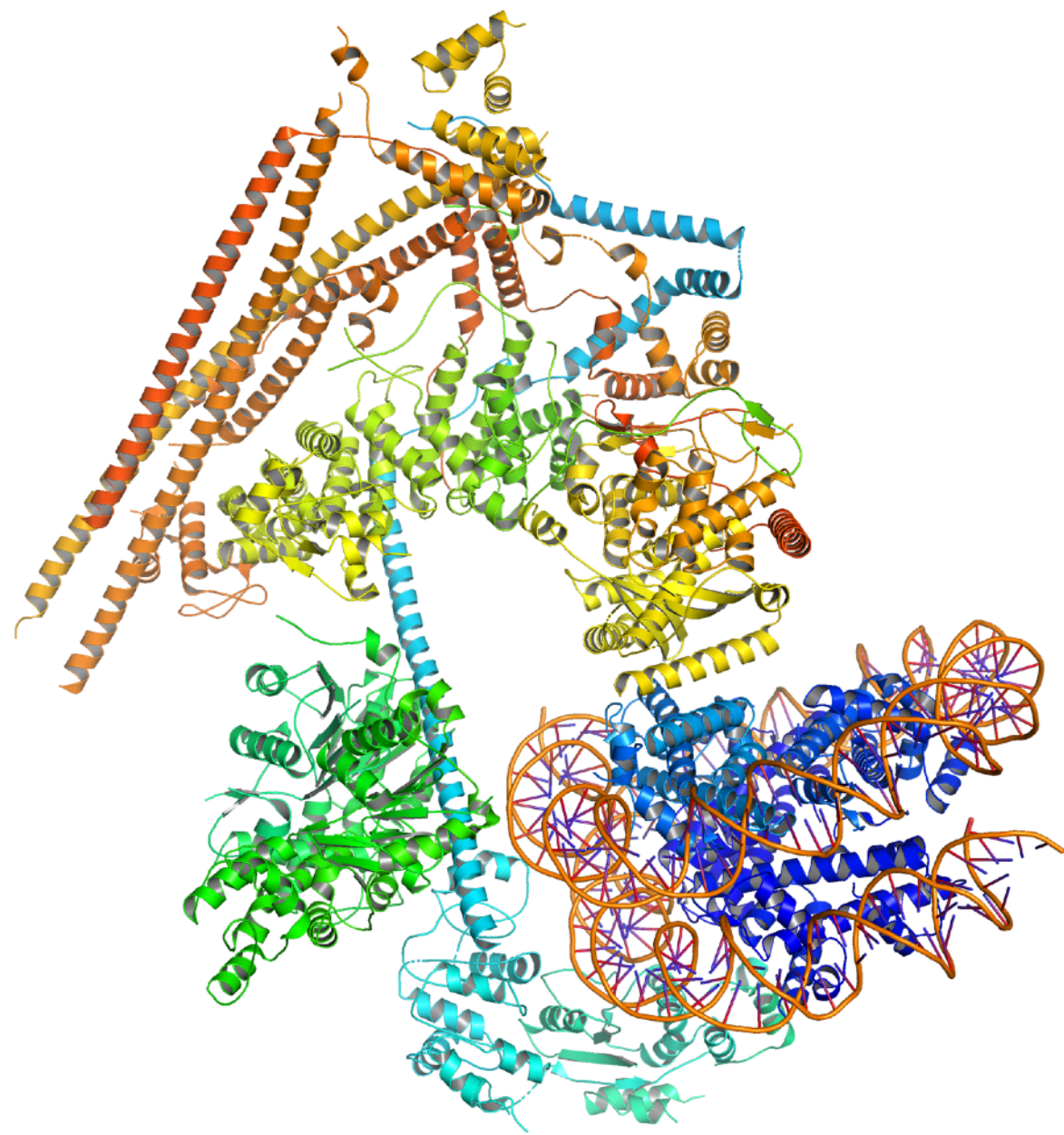
- the smaller the K_D , the stronger the interaction
- nM $\rightarrow$ very strong, μ M $\rightarrow$ rather weak
- it is a continuum!

When can we say that two proteins interact with each other?

- interaction strength (K_D) is a continuum
- there is no universal cutoff on the K_D
- discrimination into binding/no binding is assay-dependent

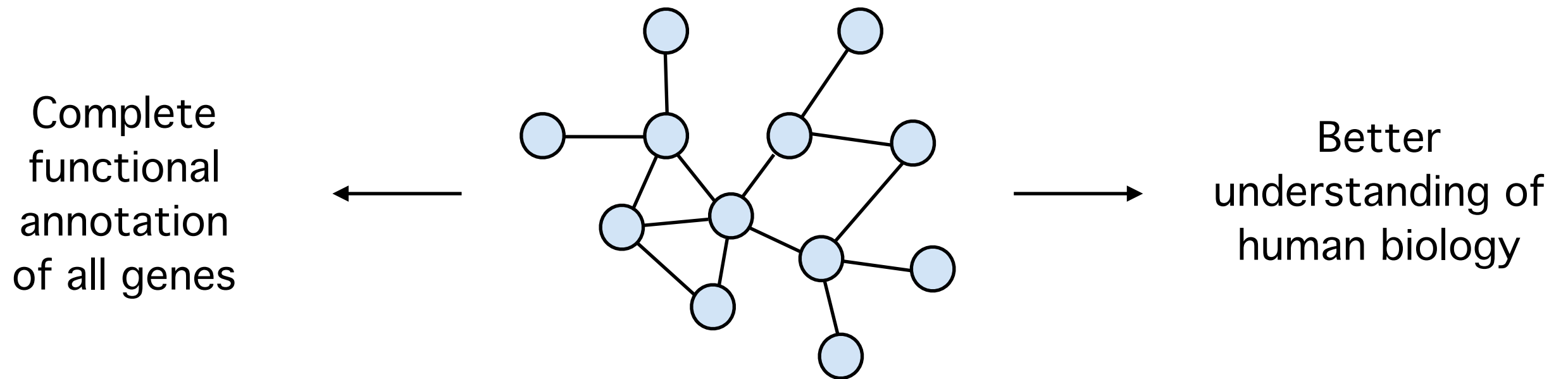


All life depends on the proper formation and dissociation of protein interactions



Mechanisms of protein interaction specificity?

If we knew all (human) protein interactions...

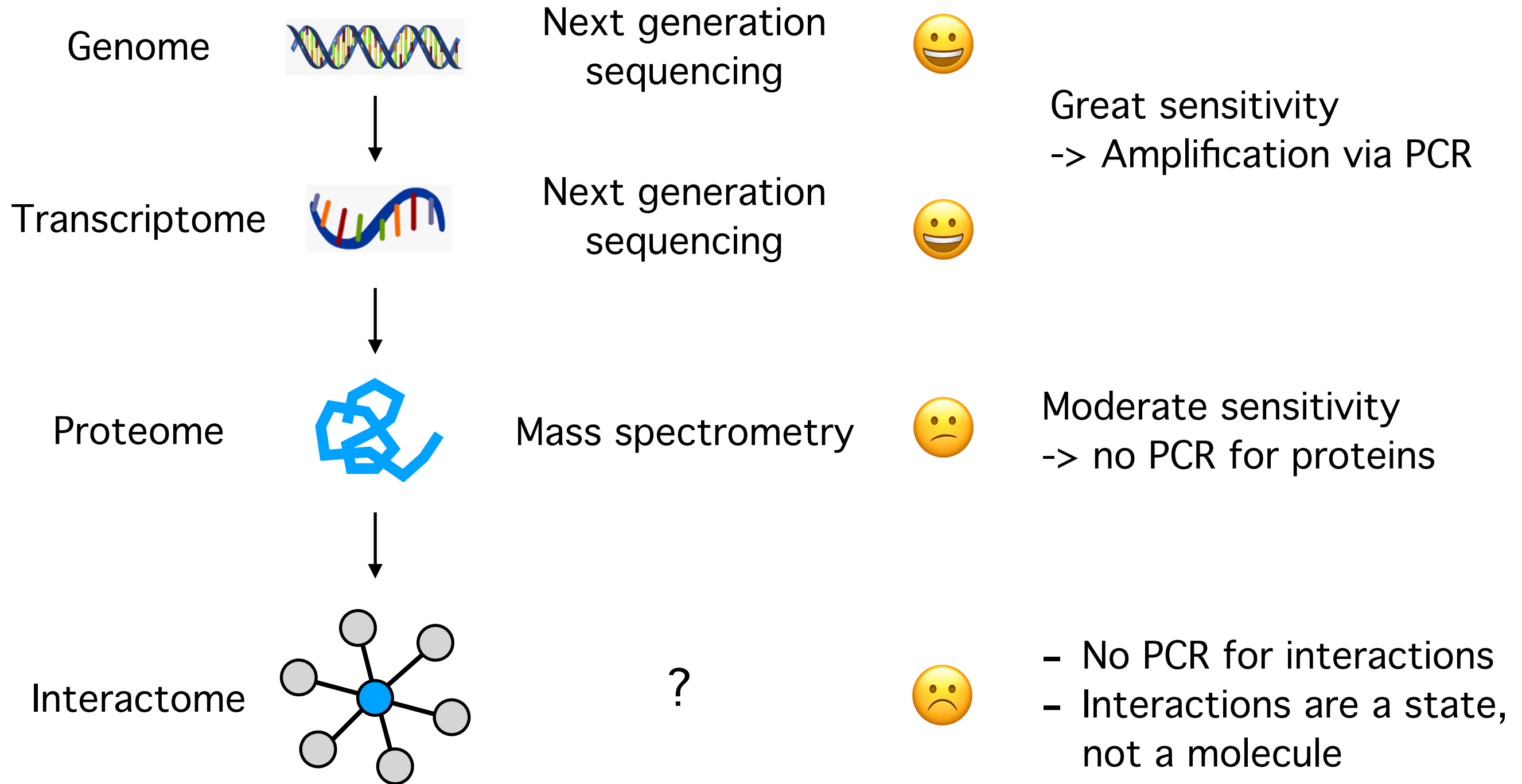


Master Module
Proteinbiochemistry and Bioinformatics
December 2024

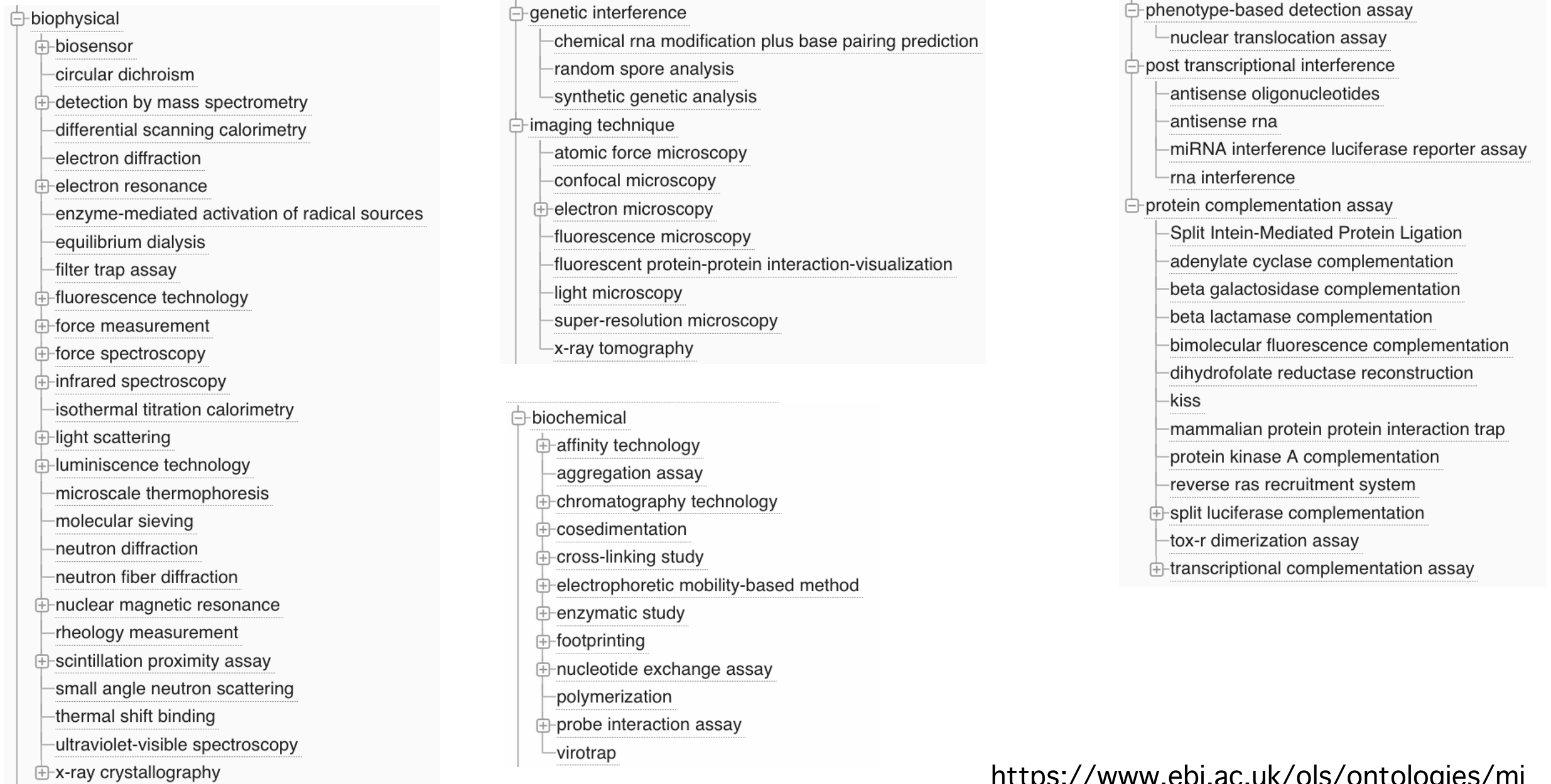
Session: Protein interaction networks

2. Methods to detect protein interactions

Why is it so hard to detect protein interactions?



Approaches to detect protein interactions



<https://www.ebi.ac.uk/ols/ontologies/mi>

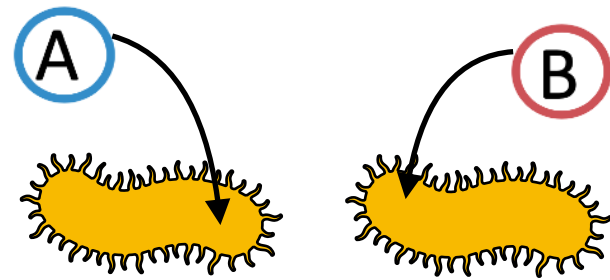
Direct assays

Binary assays

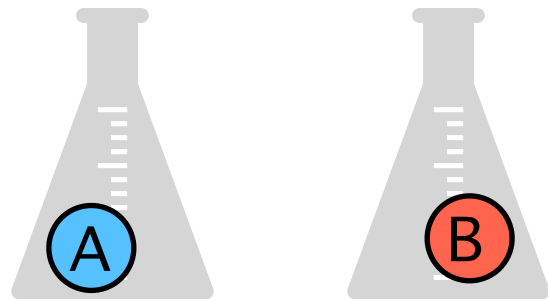
Co-complex assays

Assays to detect **direct** protein interactions

Principle



Production of
interaction
partners in
bacteria



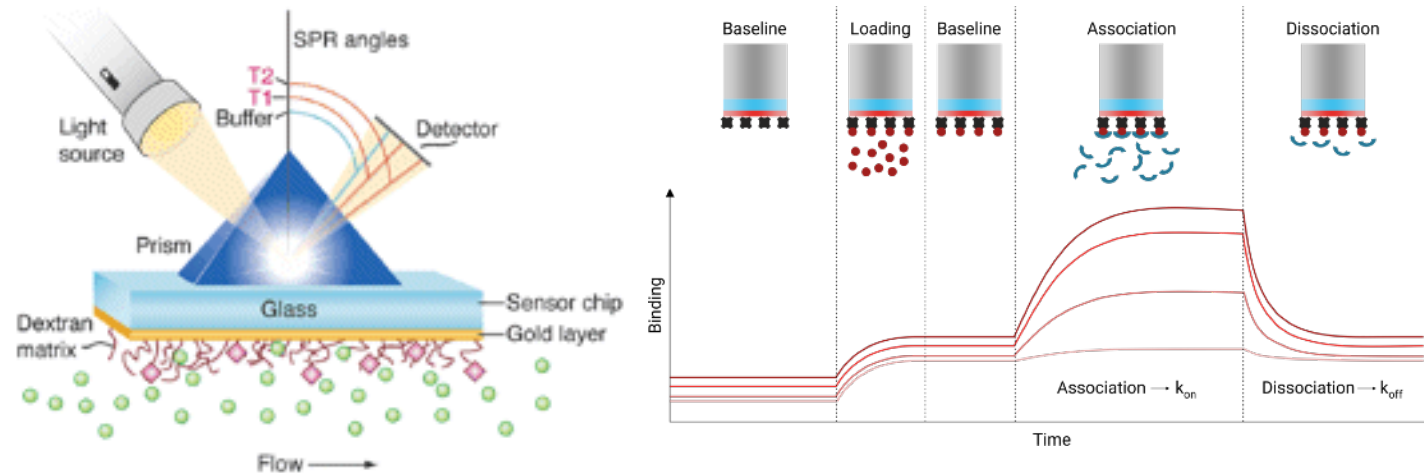
Purification of
interaction
partners



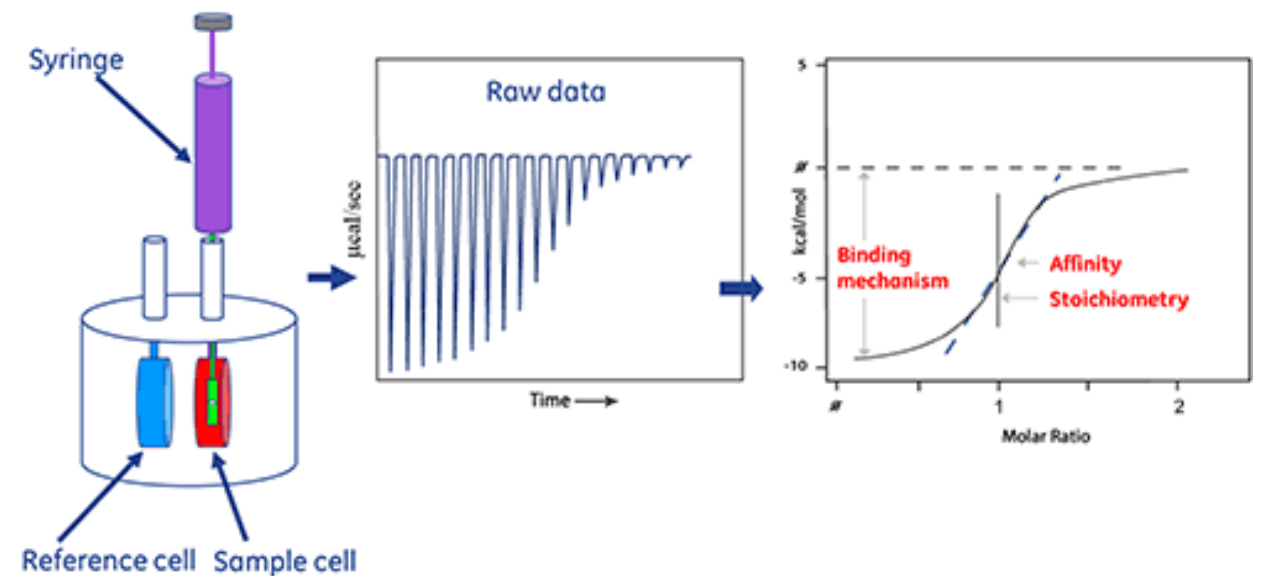
Measure

Examples of assays

Surface Plasmon Resonance (SPR)

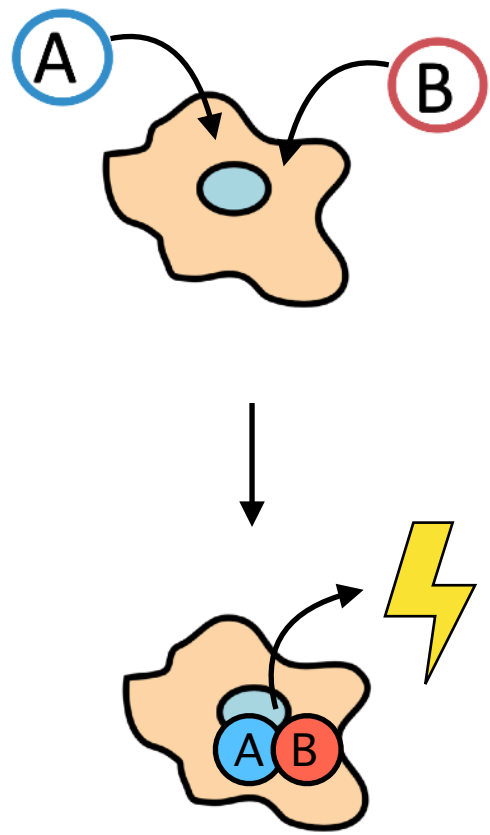


Isothermal titration calorimetry (ITC)



Assays to detect **binary** protein interactions

Principle

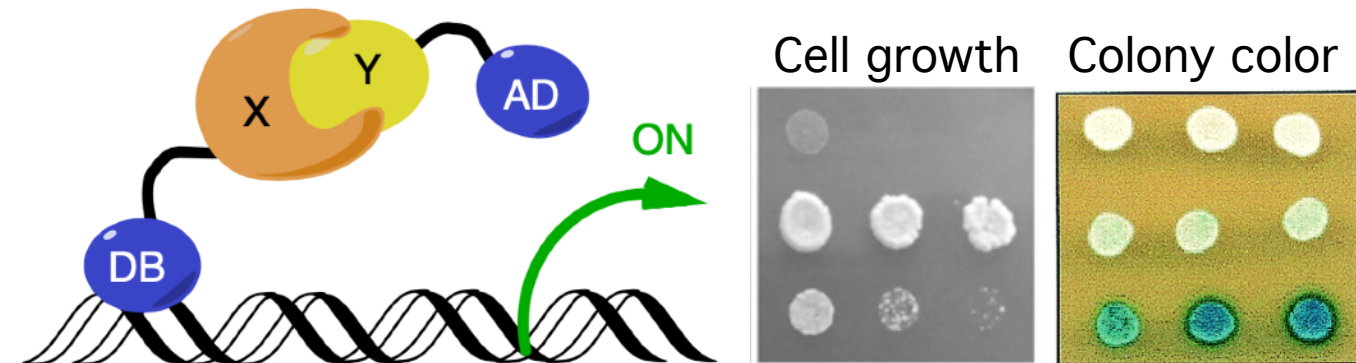


Exogenous expression of interaction partners as fusion constructs in cellular system

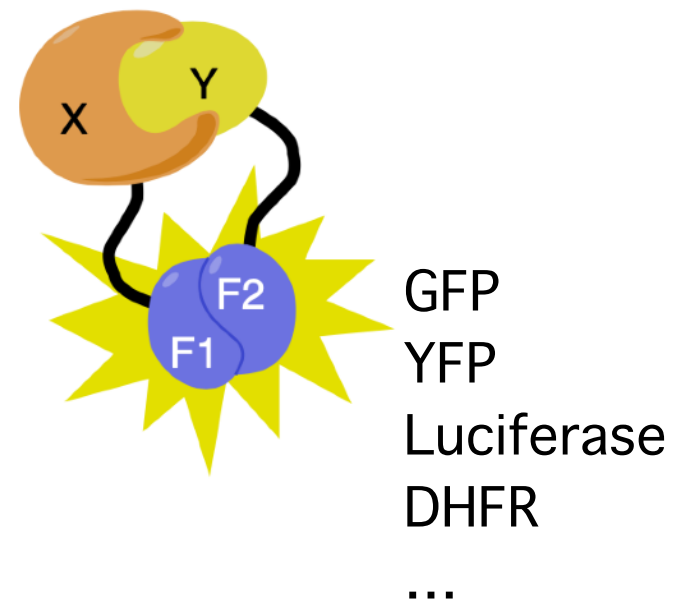
Interaction creates a signal in form of fluorescence, light, cellular growth, or others

Examples

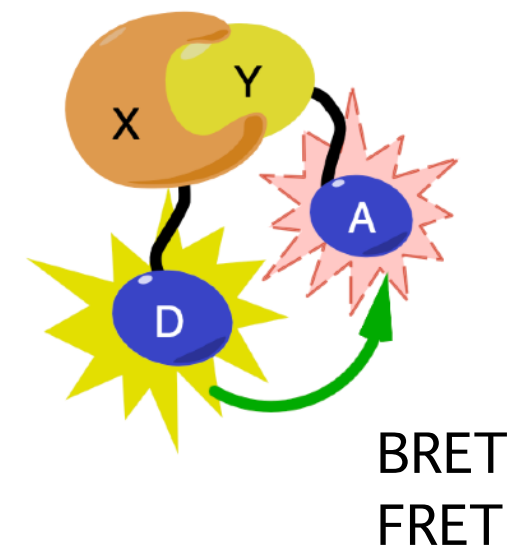
Yeast two-hybrid (Y2H)



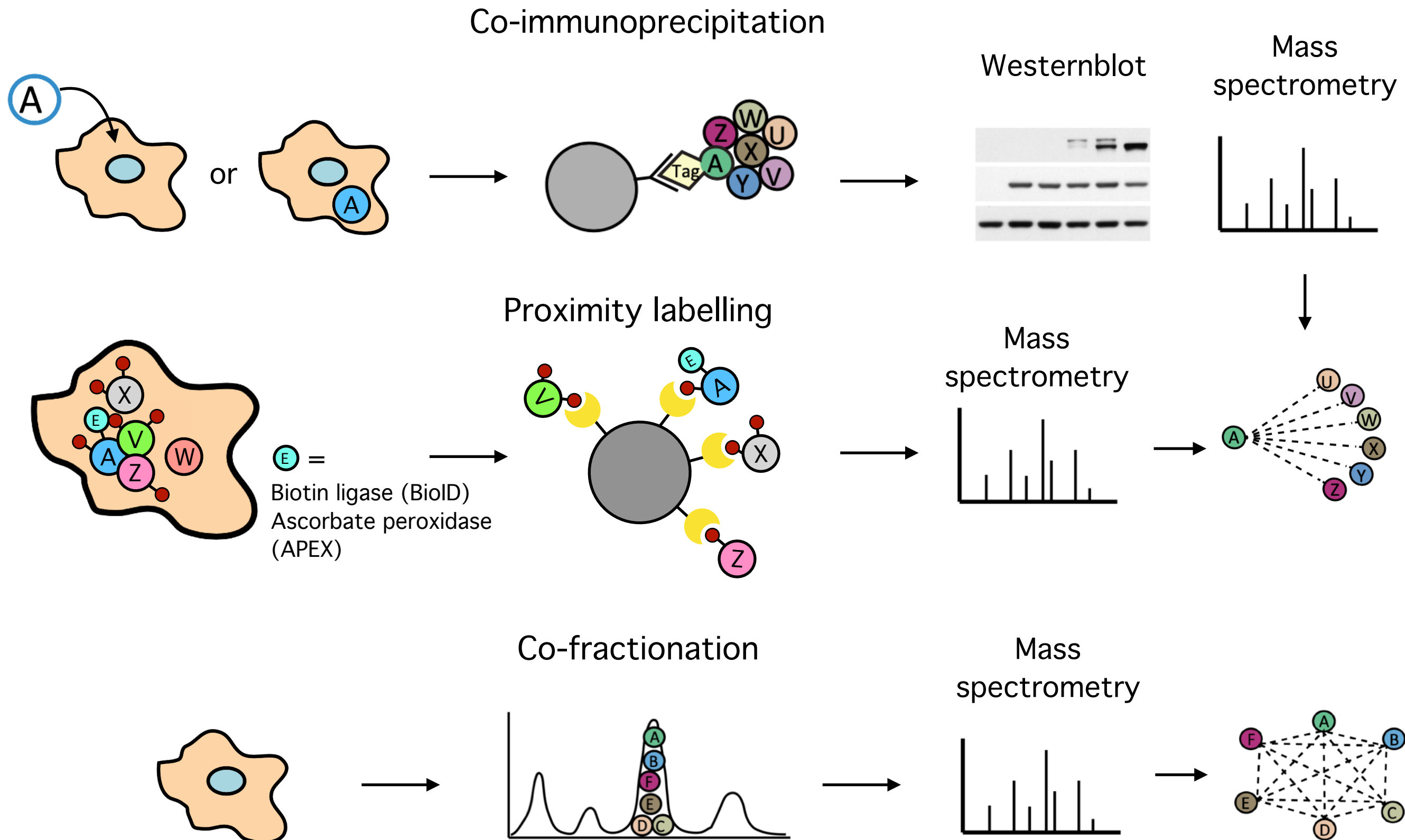
Protein complementation assays (PCA)



Energy transfer assays



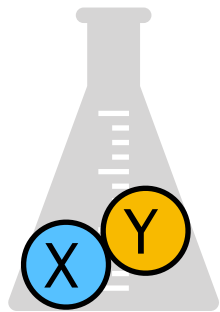
Assays to detect **co-complex** protein interactions



Different assays produce different types of protein interaction data

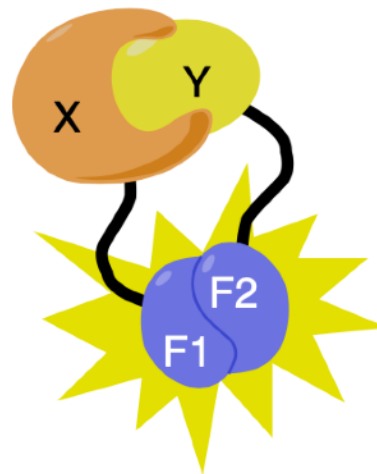
Direct assays

- Direct interactions
- Protein fragments
- With K_D
- Low-throughput



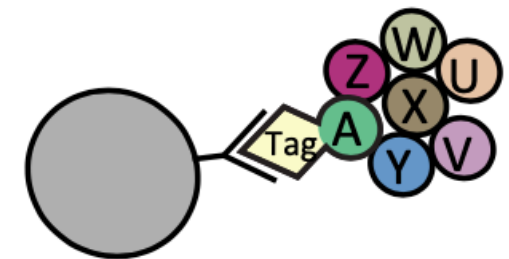
Binary assays

- Binary interactions
- Full length proteins
- No K_D
- Over-expression



Co-complex assays

- Co-complex associations
- Full length proteins
- No K_D
- Over-expression and endogenous



- All are called protein interactions
- Assays differ in which interactions they can detect

Accuracy of protein interaction assays

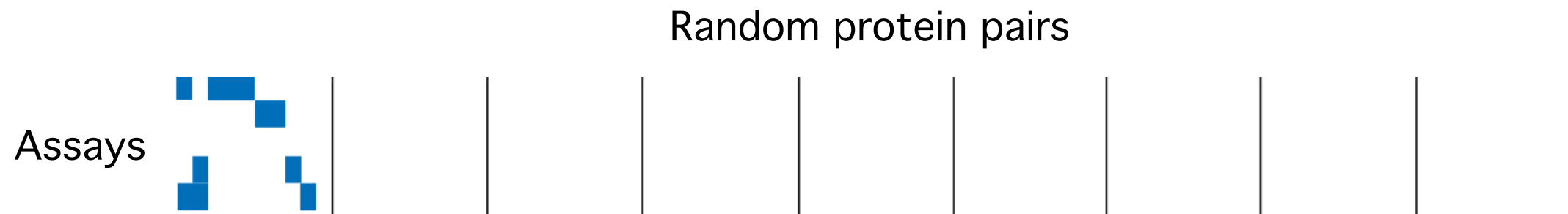
Sensitivity of protein interaction assays



Why are some interactions detected by some assays and not by others?

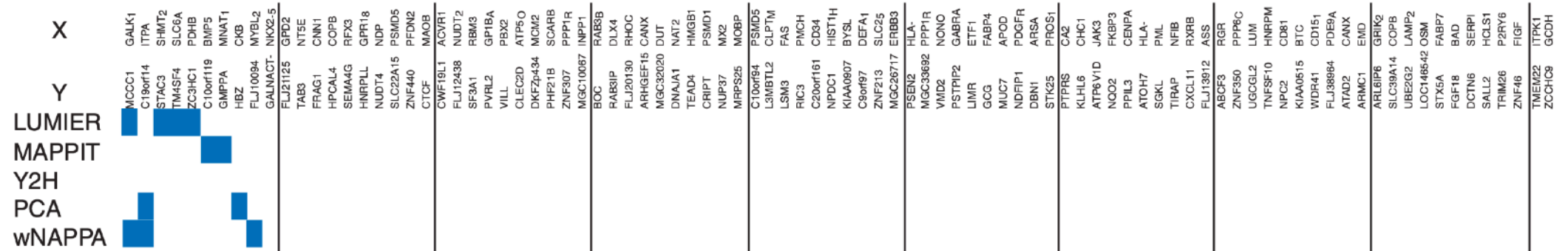
Accuracy of protein interaction assays

Specificity of protein interaction assays

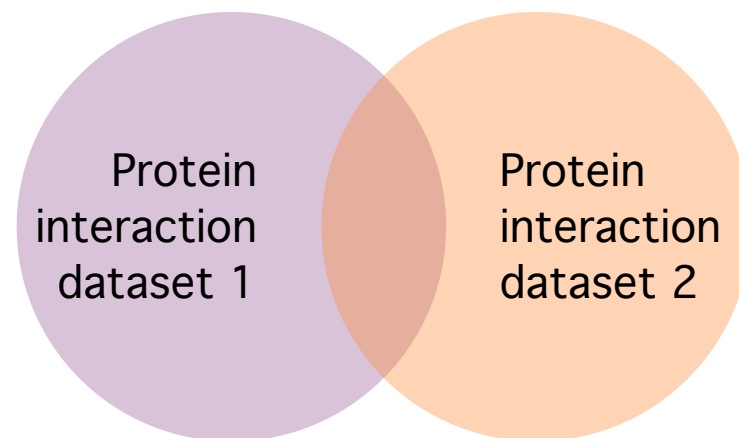


Why would an assay erroneously report a protein interaction?

Correct benchmarking of assays



Low overlap



Low sensitivity
High specificity

Methods to detected protein interactions

Summary

- Interaction strength is a continuum
- Most common methods are direct, binary, and co-complex assays
- Different methods detect different types of protein interactions
- Many interactions remain undetected
- If properly controlled interaction data can be of high quality