

# BGGN 213

## Structural Bioinformatics II

Lecture 12

Barry Grant  
UC San Diego

<http://thegrantlab.org/bggn213>

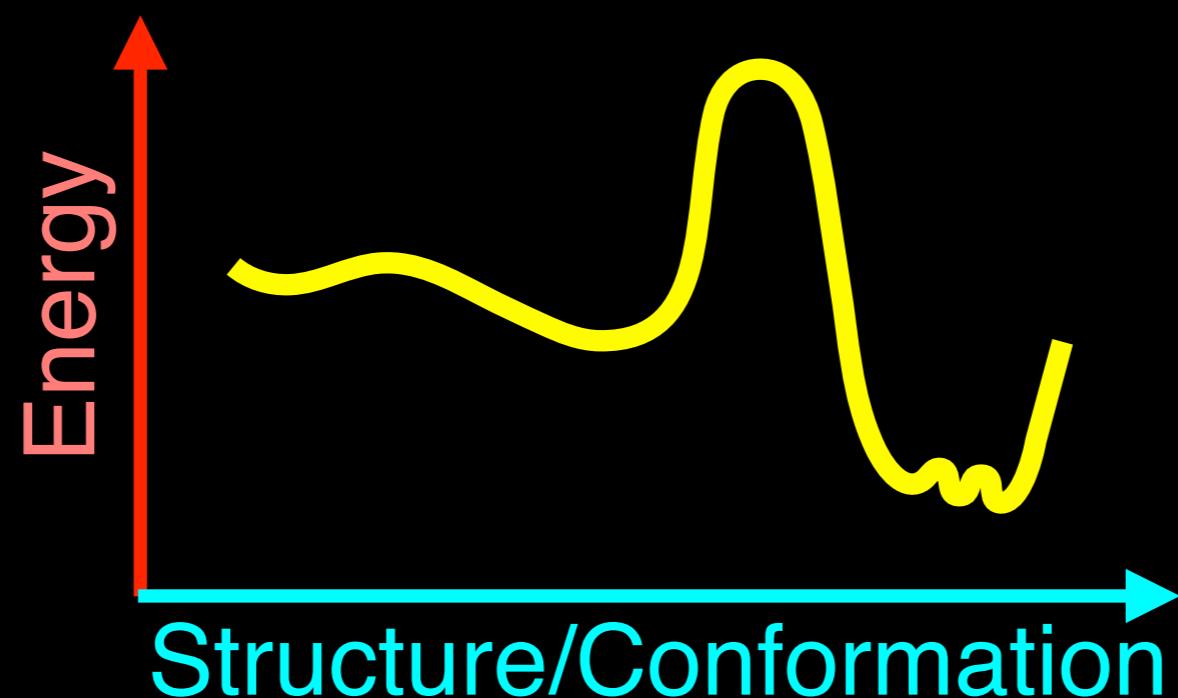
Download MGL Tools: See class website!

# Next Up:

- **Overview of structural bioinformatics**
  - Motivations, goals and challenges
- **Fundamentals of protein structure**
  - Structure composition, form and forces
- **Representing, interpreting & modeling protein structure**
  - Visualizing and interpreting protein structures
  - Analyzing protein structures
  - Modeling energy as a function of structure
  - Drug discovery & Predicting functional dynamics

# Key concept:

Potential functions describe a systems  
energy as a function of its structure



Two main approaches:

- (1). Physics-Based
- (2). Knowledge-Based

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(1). Physics-Based

(2). Knowledge-Based

For **physics** based potentials  
energy terms come from physical theory

$$V(R) = E_{\text{bonded}} + E_{\text{non.bonded}}$$

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Sum of **bonded** and **non-bonded**  
atom-type and position based terms

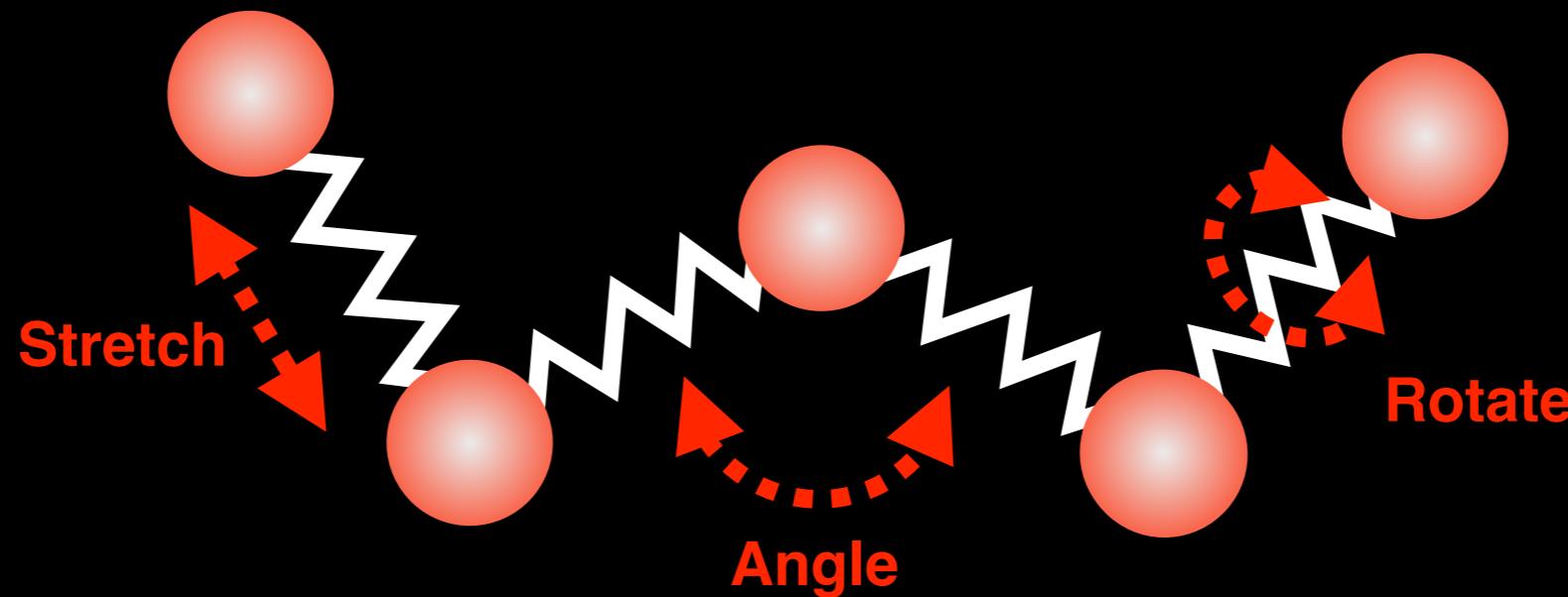
$$V(R) = E_{\text{bonded}} + E_{\text{non.bonded}}$$

$E_{\text{bonded}}$  is itself a sum of three terms:

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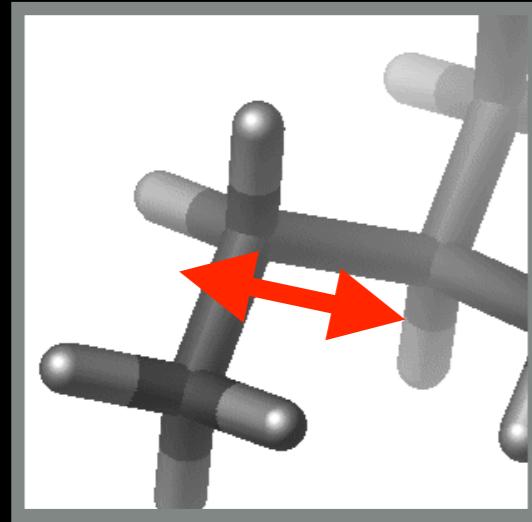
$$E_{\text{bond.stretch}} + E_{\text{bond.angle}} + E_{\text{bond.rotate}}$$



$$V(R) = E_{bonded} + E_{non.bonded}$$

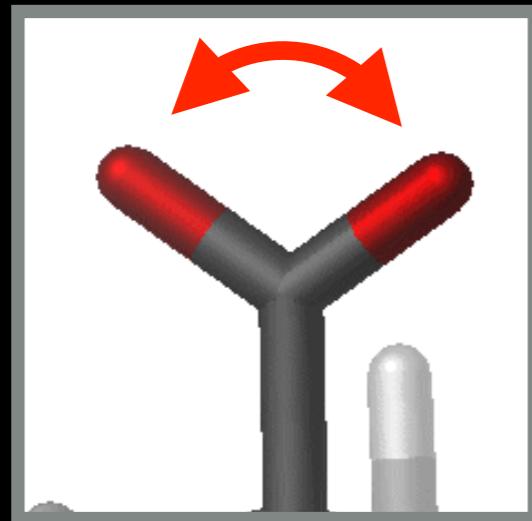
$E_{bonded}$  is itself a sum of three terms:

$$E_{bond.stretch} + E_{bond.angle} + E_{bond.rotate}$$



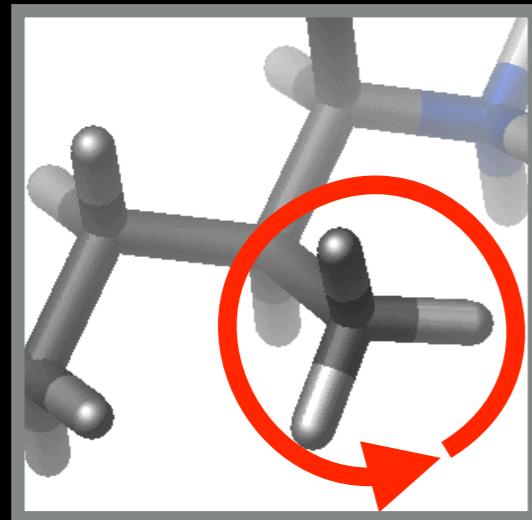
Bond Stretch

$E_{bond.stretch}$



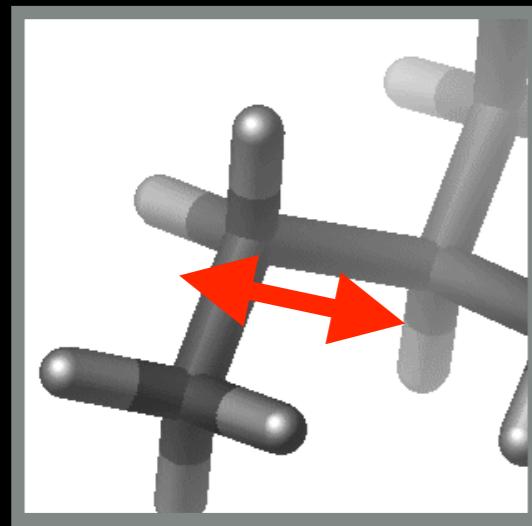
Bond Angle

$E_{bond.angle}$



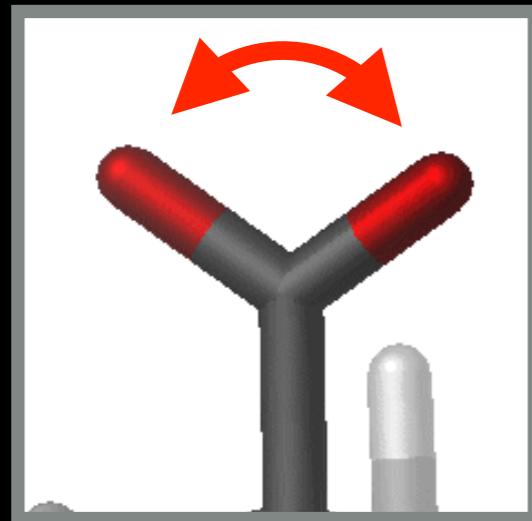
Bond Rotate

$E_{bond.rotate}$



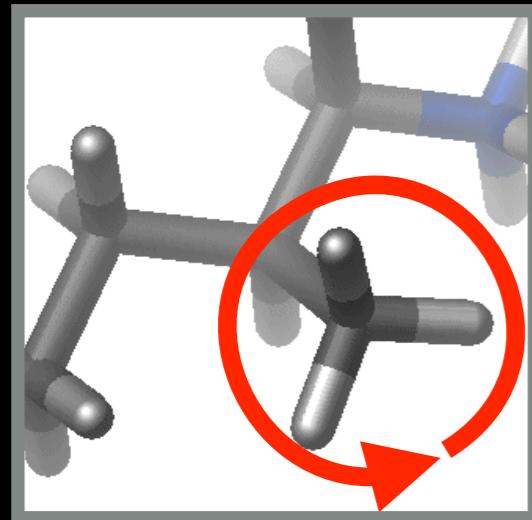
## Bond Stretch

$$\sum_{bonds} K_i^{bs}(b_i - b_o)$$



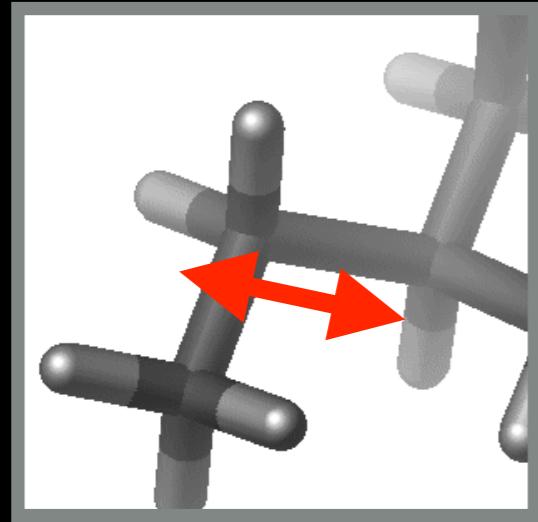
## Bond Angle

$$\sum_{angles} K_i^{ba}(\theta_i - \theta_o)$$



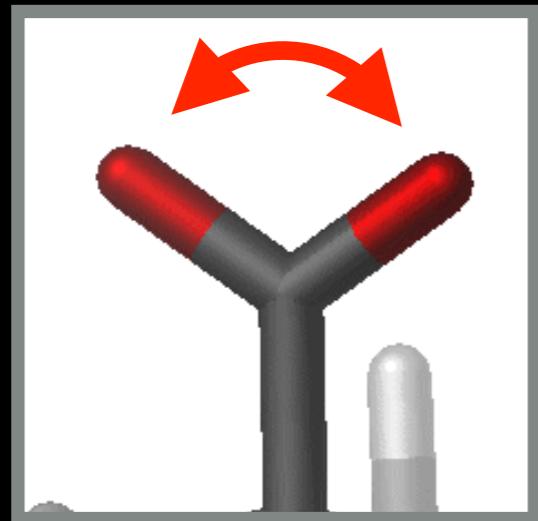
## Bond Rotate

$$\sum_{dihedrals} K_i^{br}[1 - \cos(n_i\phi_i - \phi_o)]$$



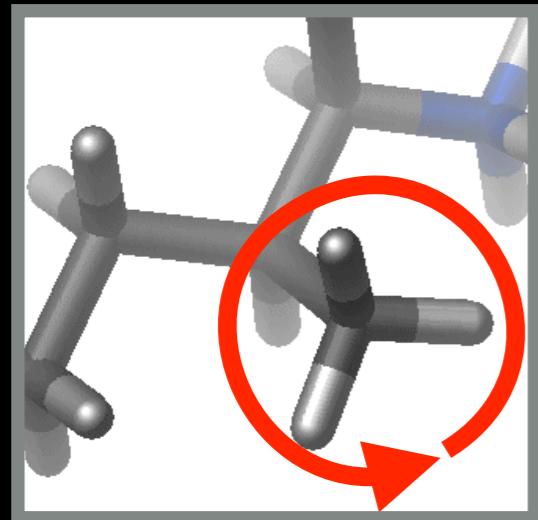
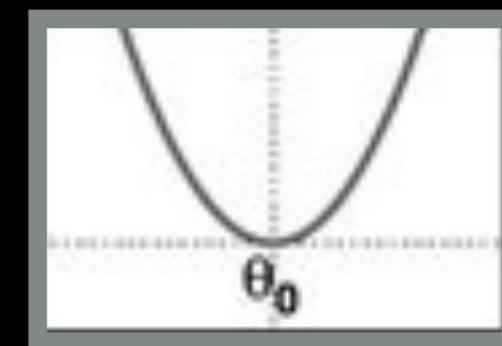
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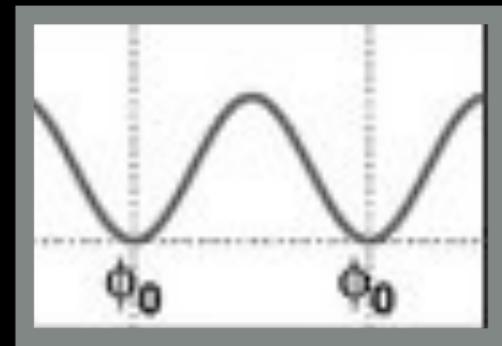
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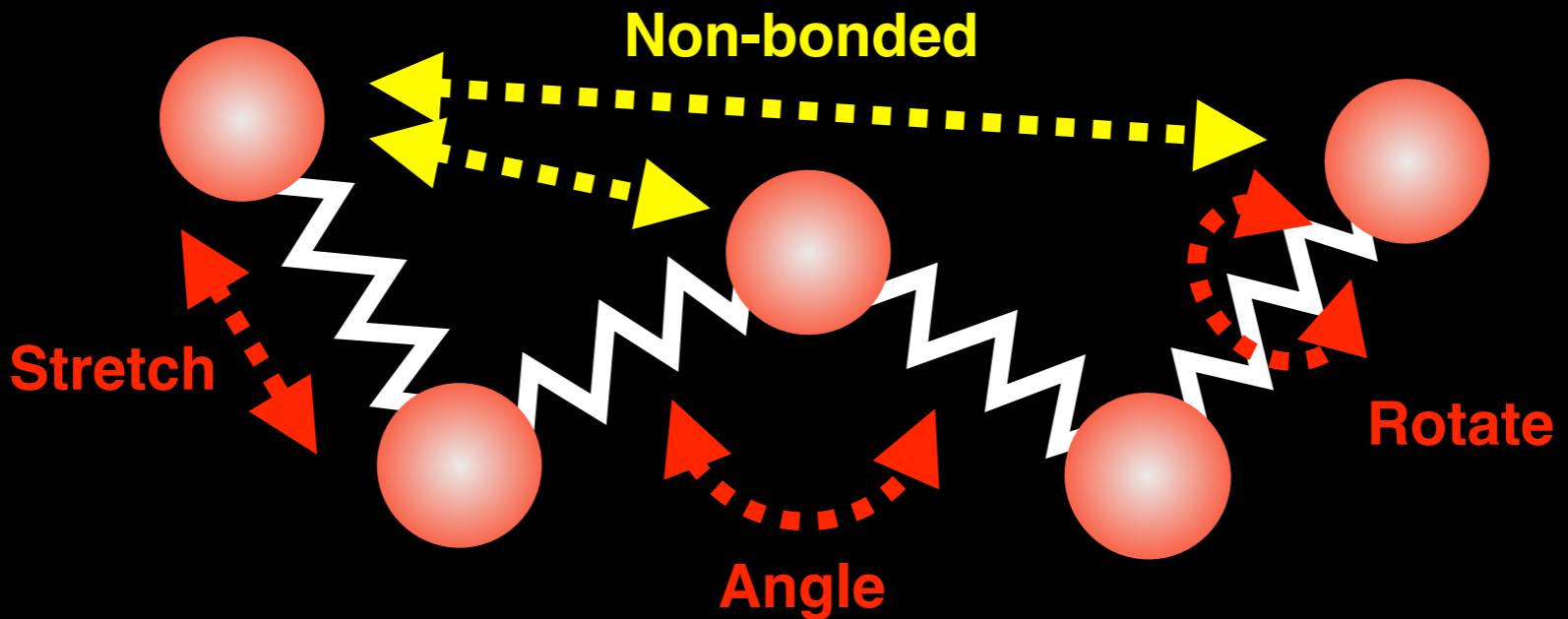
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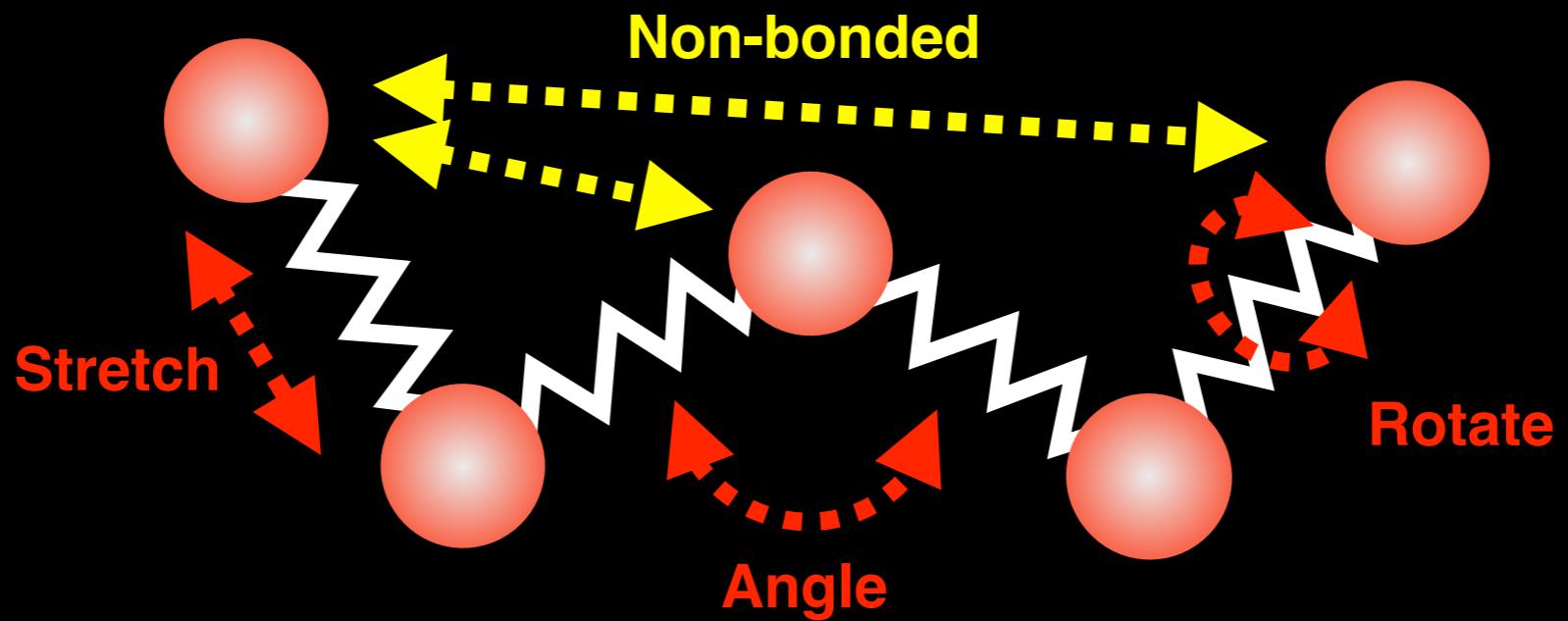
$$E_{van.der.Waals} + E_{electrostatic}$$



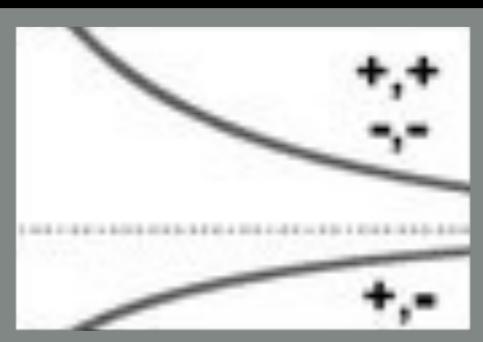
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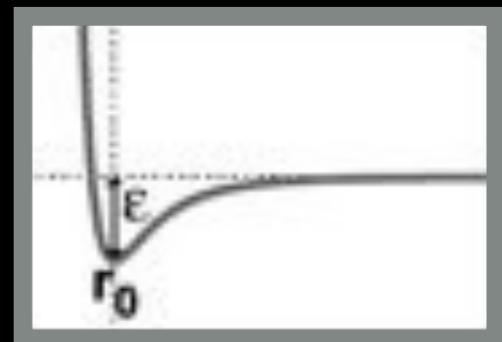
$$E_{van.der.Waals} + E_{electrostatic}$$



$$E_{electrostatic} = \sum_{pairs.i.j} \frac{q_i q_j}{\epsilon r_{ij}}$$



$$E_{van.der.Waals} = \sum_{pairs.i.j} \left[ \epsilon_{ij} \left( \frac{r_{o.ij}}{r_{ij}} \right)^{12} - 2 \epsilon_{ij} \left( \frac{r_{o.ij}}{r_{ij}} \right)^6 \right]$$



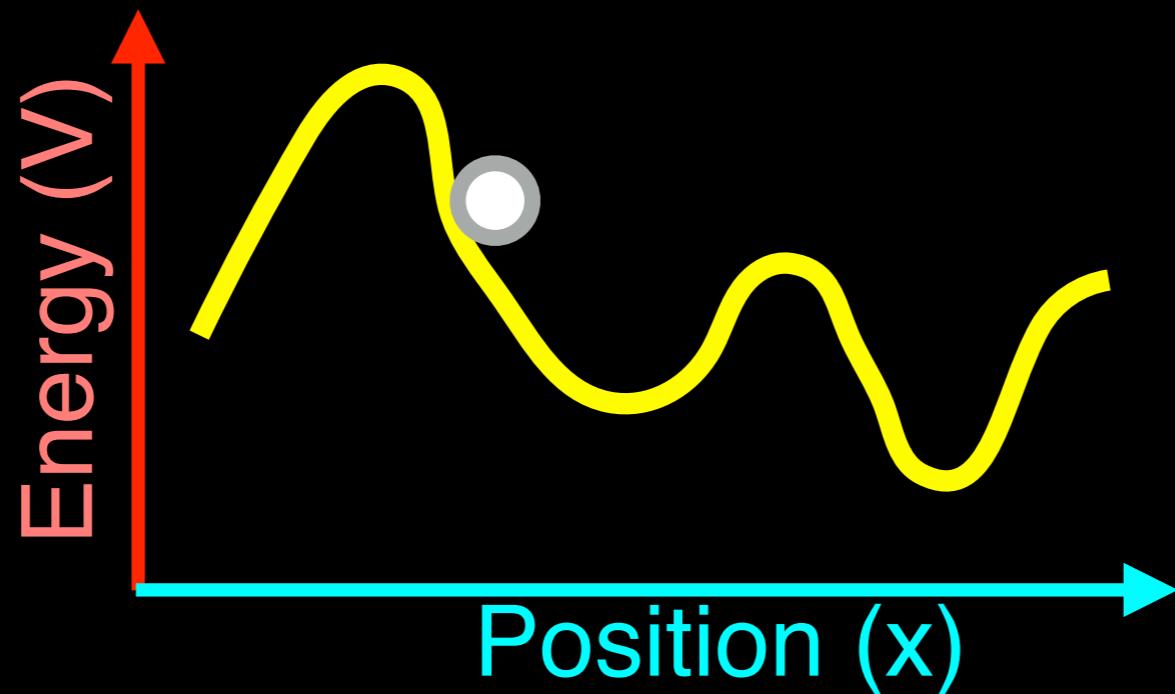
# Total potential energy

The potential energy can be given as a sum of terms for: Bond stretching, Bond angles, Bond rotations, van der Walls and Electrostatic interactions between atom pairs

$$V(R) = E_{\text{bonded}} + E_{\text{non.bonded}}$$
$$\left. \begin{aligned} &E_{\text{bond.stretch}} \\ &+ E_{\text{bond.angle}} \\ &+ E_{\text{bond.rotate}} \\ &+ E_{\text{van.der.Waals}} \\ &+ E_{\text{electrostatic}} \end{aligned} \right\} E_{\text{bonded}}$$
$$\left. \begin{aligned} & \\ & \\ & \\ & \\ & \end{aligned} \right\} E_{\text{non.bonded}}$$

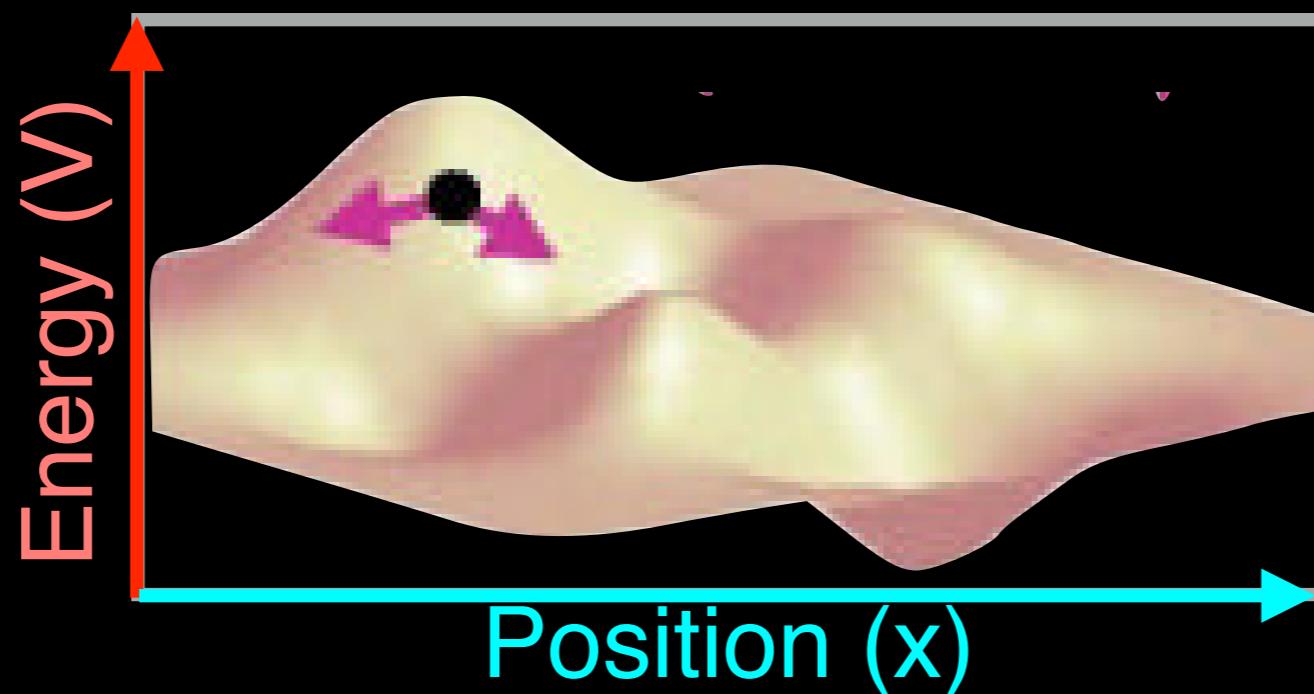
# Potential energy surface

Now we can calculate the **potential energy surface** that fully describes the energy of a molecular system as a function of its geometry



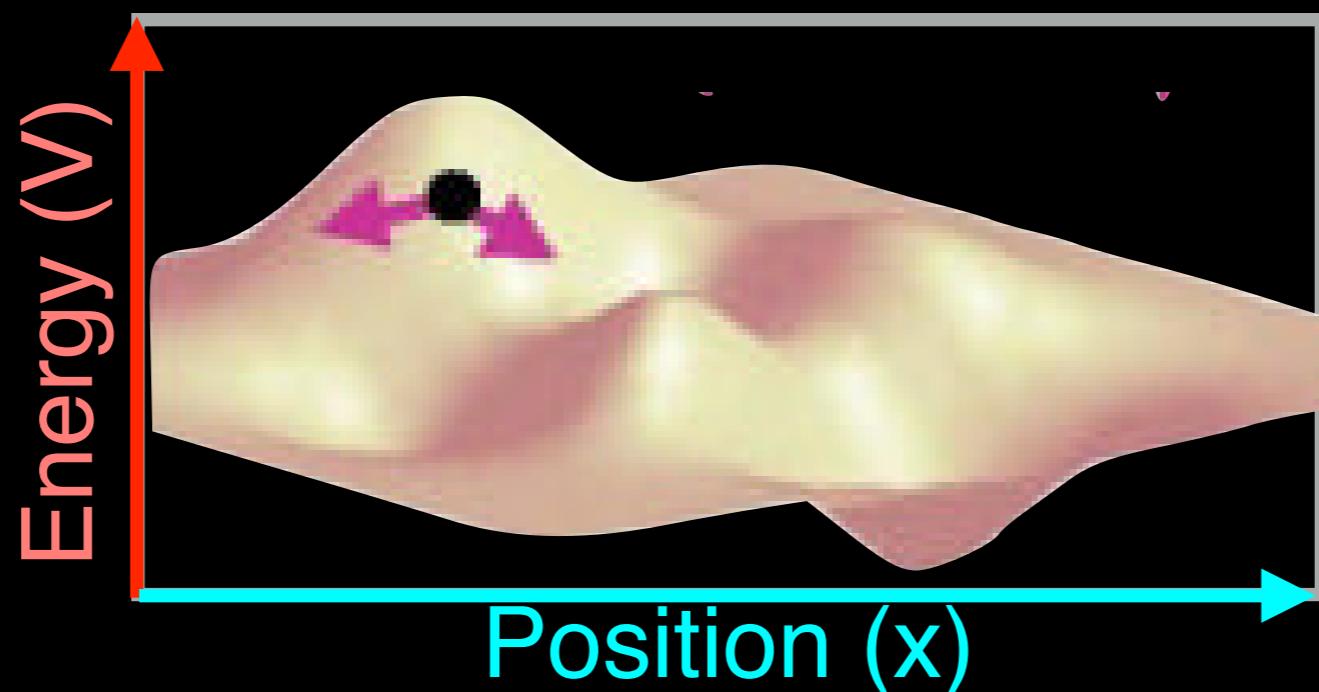
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# Key concept:

Now we can calculate the **potential energy surface** that fully describes the energy of a molecular system as a function of its geometry



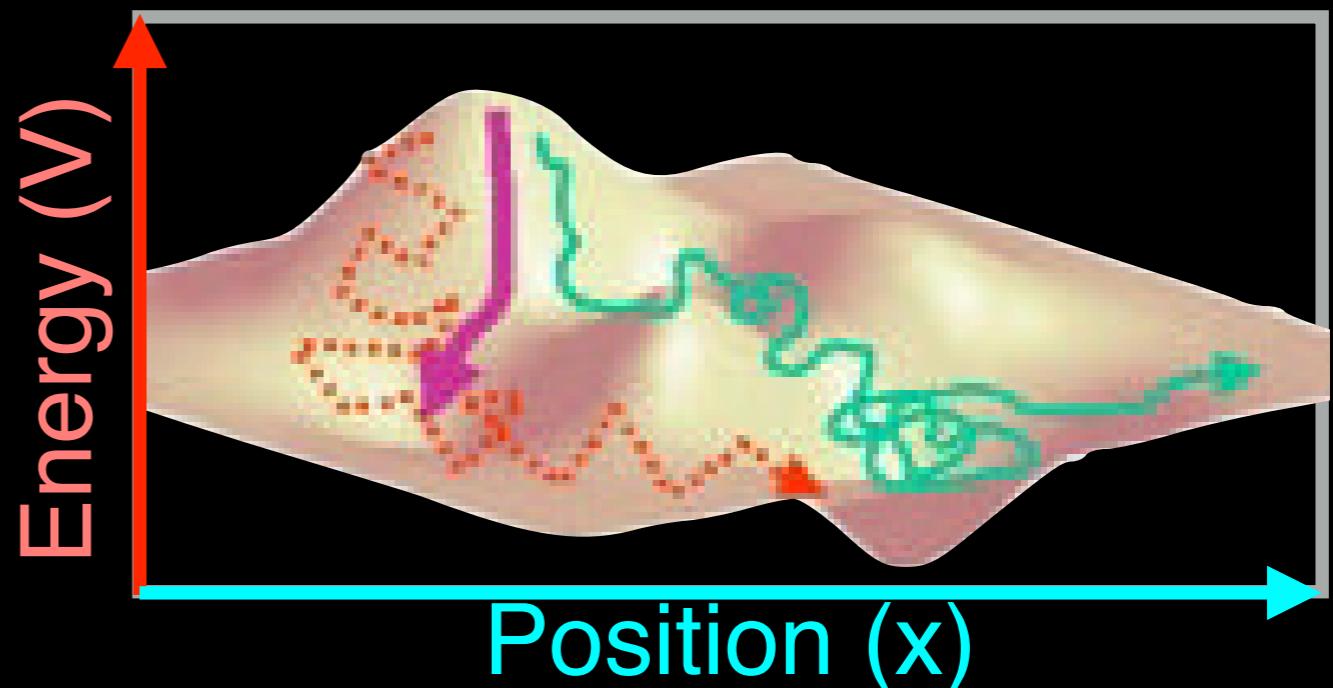
- The **forces** are the gradients of the energy

$$F(x) = - dV/dx$$

# Moving Over The Energy Surface

- **Energy Minimization**  
drops into local minimum
- **Molecular Dynamics**  
uses thermal energy to  
move smoothly over  
surface
- **Monte Carlo Moves** are  
random. Accept with  
probability:

$$\exp(-\Delta V/dx)$$



# PHYSICS-ORIENTED APPROACHES

## Weaknesses

Fully physical detail becomes computationally intractable

Approximations are unavoidable

(Quantum effects approximated classically, water may be treated crudely)

Parameterization still required

## Strengths

Interpretable, provides guides to design

Broadly applicable, in principle at least

Clear pathways to improving accuracy

## Status

Useful, widely adopted but far from perfect

Multiple groups working on fewer, better approxs

Force fields, quantum  
entropy, water effects

Moore's law: hardware improving

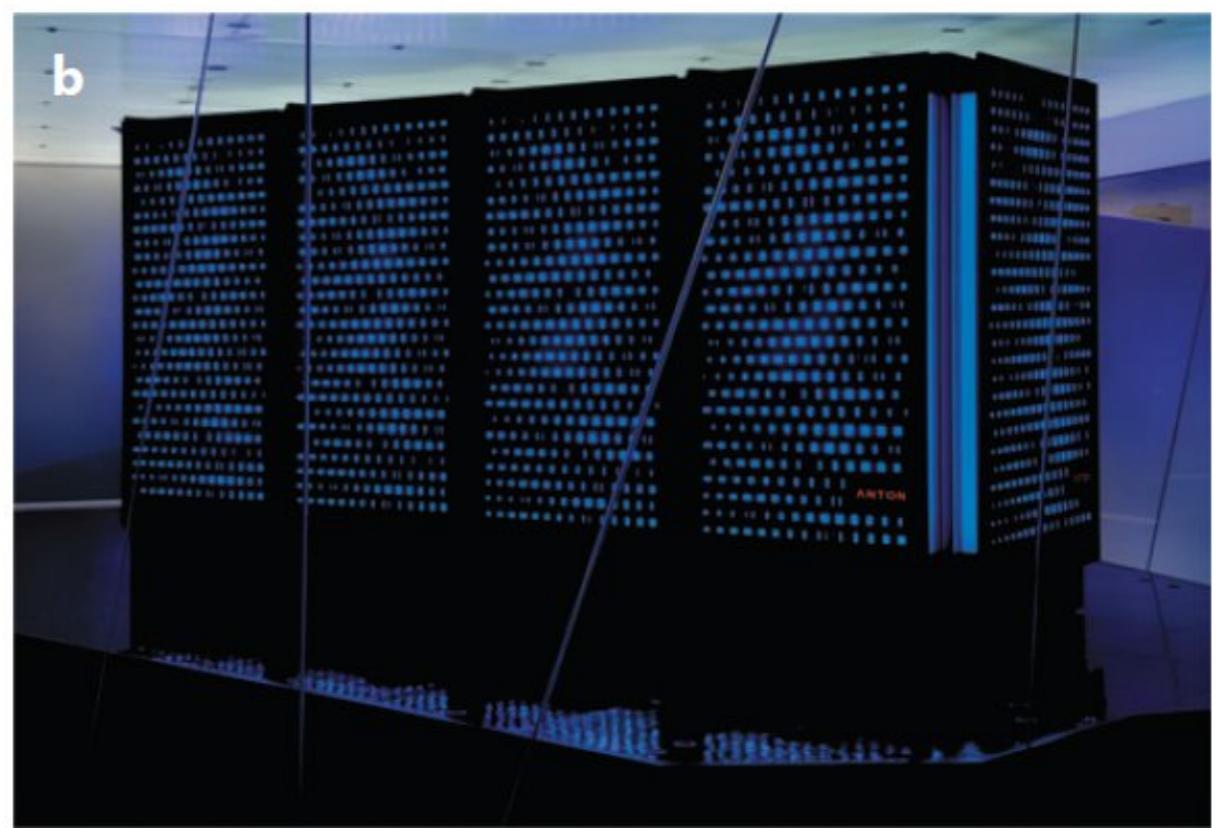
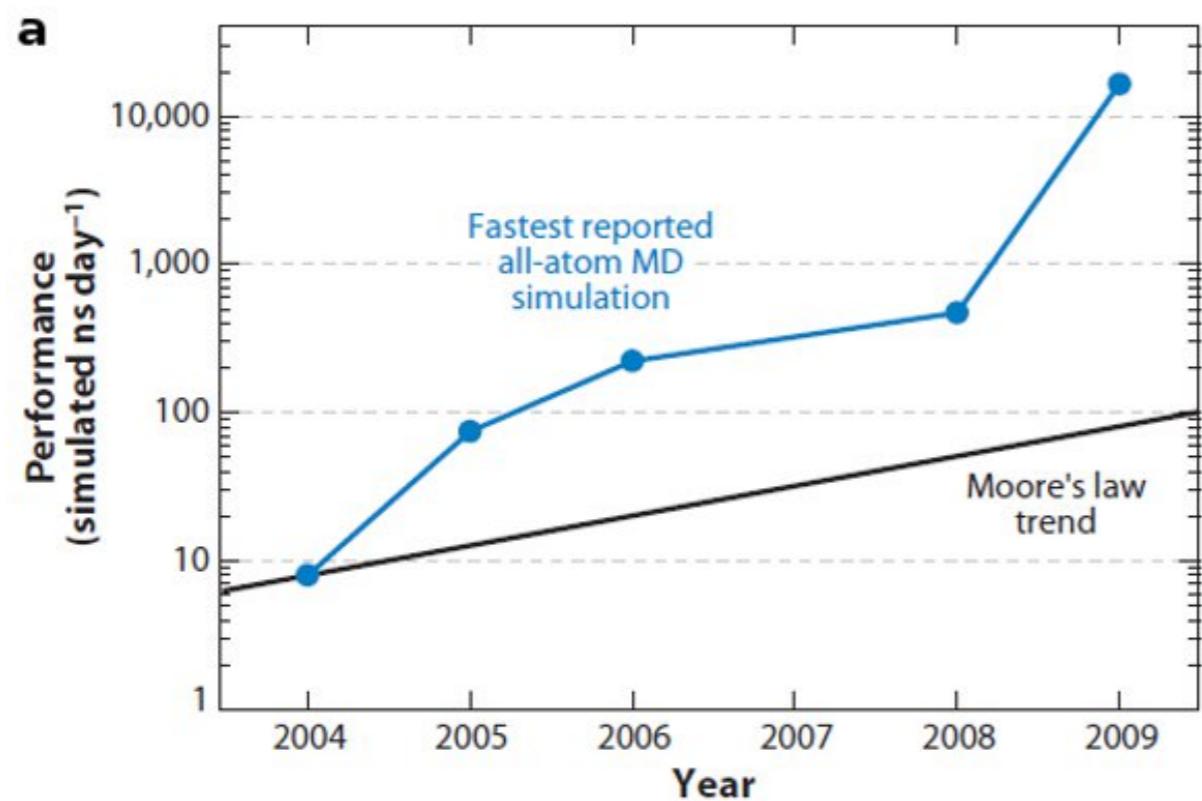
## HOW COMPUTERS HAVE CHANGED

DATE	COST	SPEED	MEMORY	SIZE
1967	\$40M	0.1 MHz	1 MB	WALL
2013	\$4,000	1 GHz	10 GB	LAPTOP
CHANGE	10,000	10,000	10,000	10,000

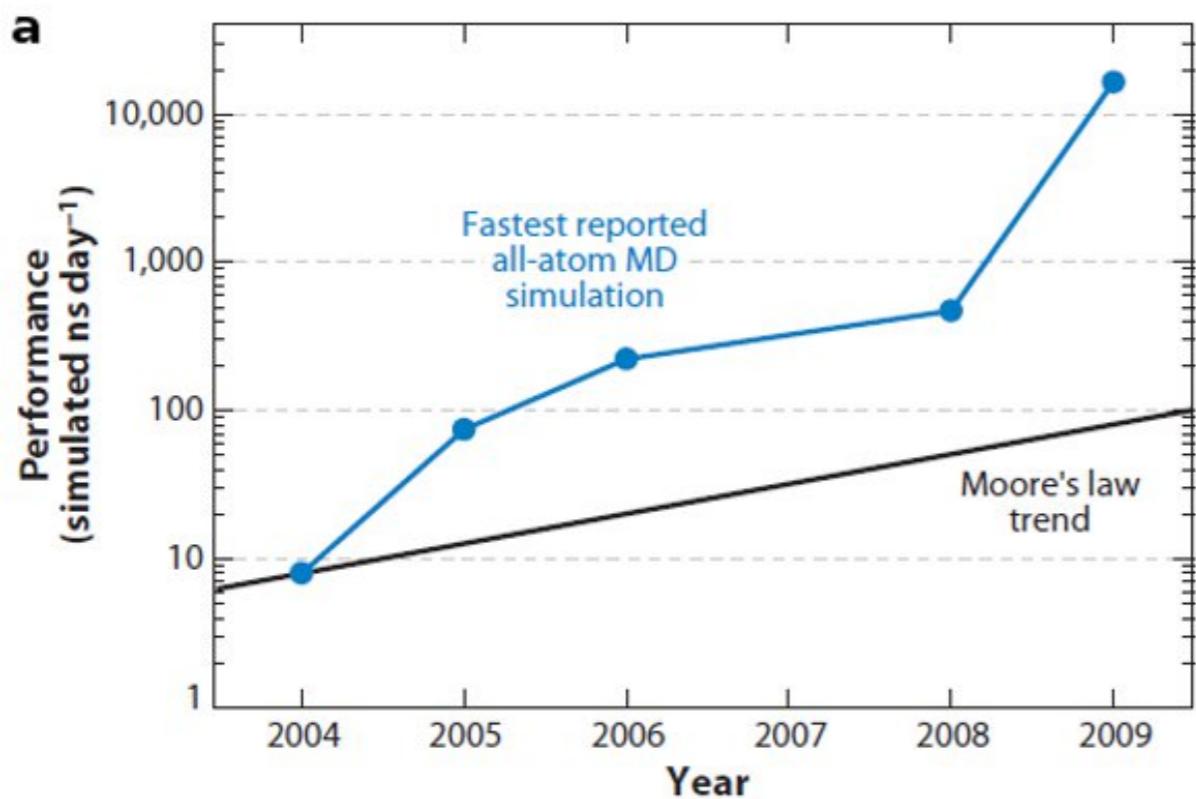
If cars were like computers then a new Volvo would cost \$3, would have a top speed of 1,000,000 Km/hr, would carry 50,000 adults and would park in a shadow.



# SIDE-NOTE: GPUS AND ANTON SUPERCOMPUTER



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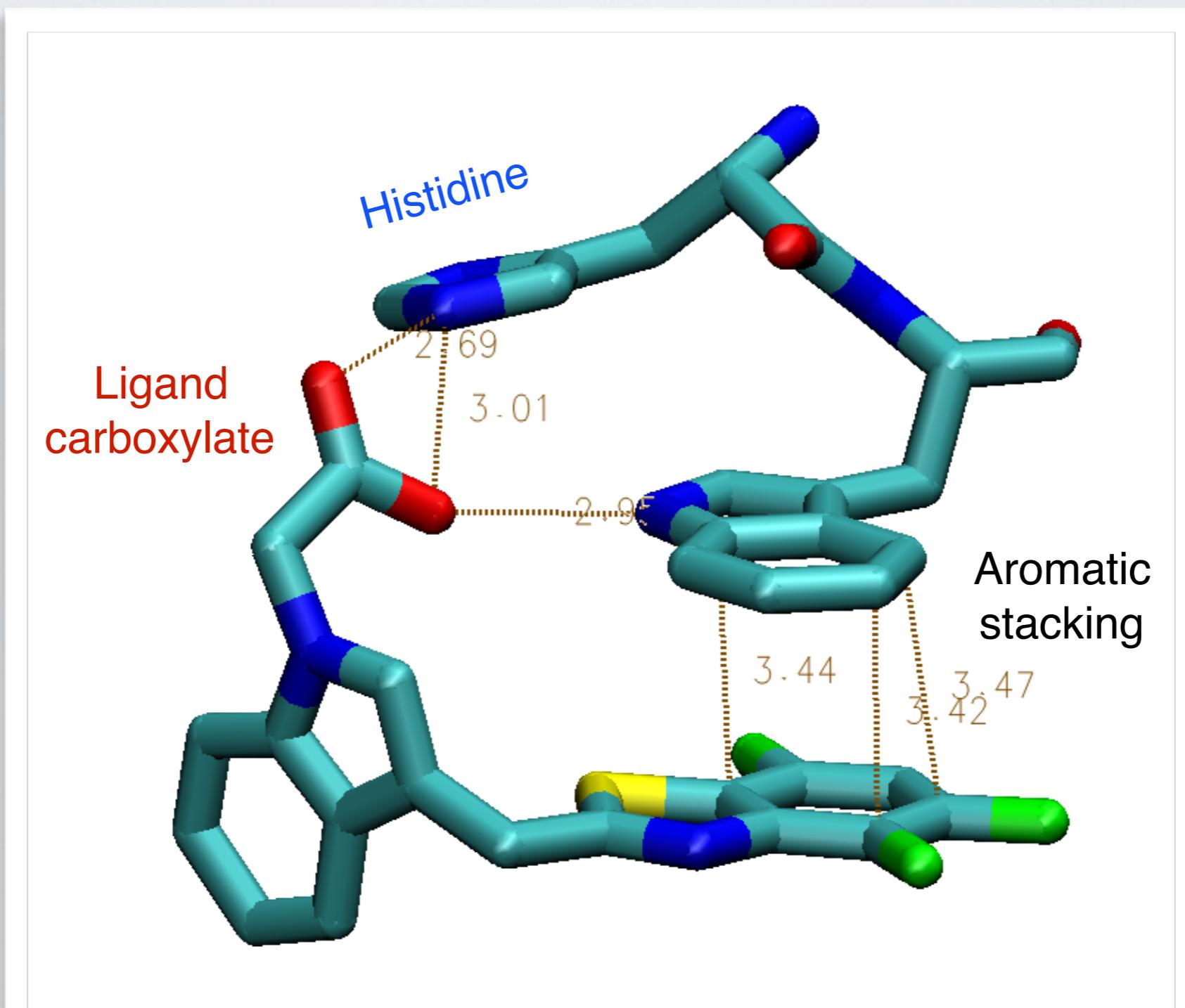


# POTENTIAL FUNCTIONS DESCRIBE A SYSTEMS **ENERGY** AS A FUNCTION OF ITS **STRUCTURE**

Two main approaches:

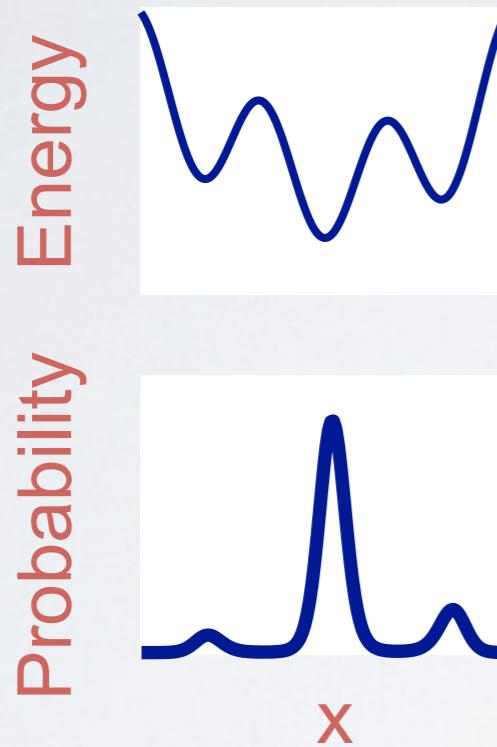
- (1). Physics-Based
- (2). Knowledge-Based

# KNOWLEDGE-BASED DOCKING POTENTIALS



# ENERGY DETERMINES **PROBABILITY** (STABILITY)

Basic idea: Use probability as a proxy for energy



Boltzmann:

$$p(r) \propto e^{-E(r)/RT}$$

Inverse Boltzmann:

$$E(r) = -RT \ln[p(r)]$$

**Example: ligand carboxylate O to protein histidine N**

Find all protein-ligand structures in the PDB with a ligand carboxylate O

1. For each structure, histogram the distances from O to every histidine N
2. Sum the histograms over all structures to obtain  $p(r_{O-N})$
3. Compute  $E(r_{O-N})$  from  $p(r_{O-N})$

# KNOWLEDGE-BASED POTENTIALS

## Weaknesses

Accuracy limited by availability of data

## Strengths

Relatively easy to implement

Computationally fast

## Status

Useful, far from perfect

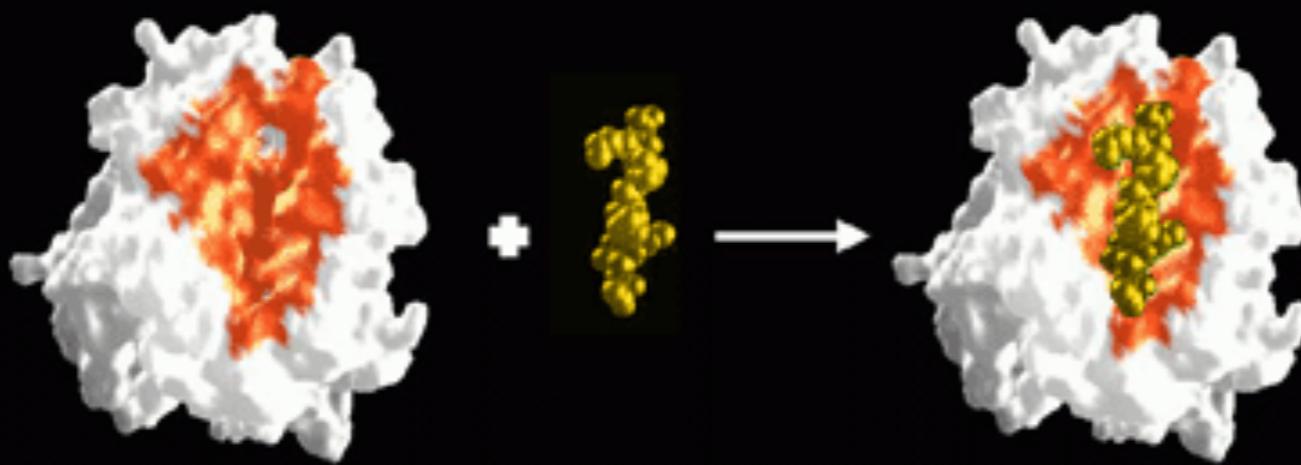
May be at point of diminishing returns

(not always clear how to make improvements)

- Break -

**Download MGL Tools:** See class website!

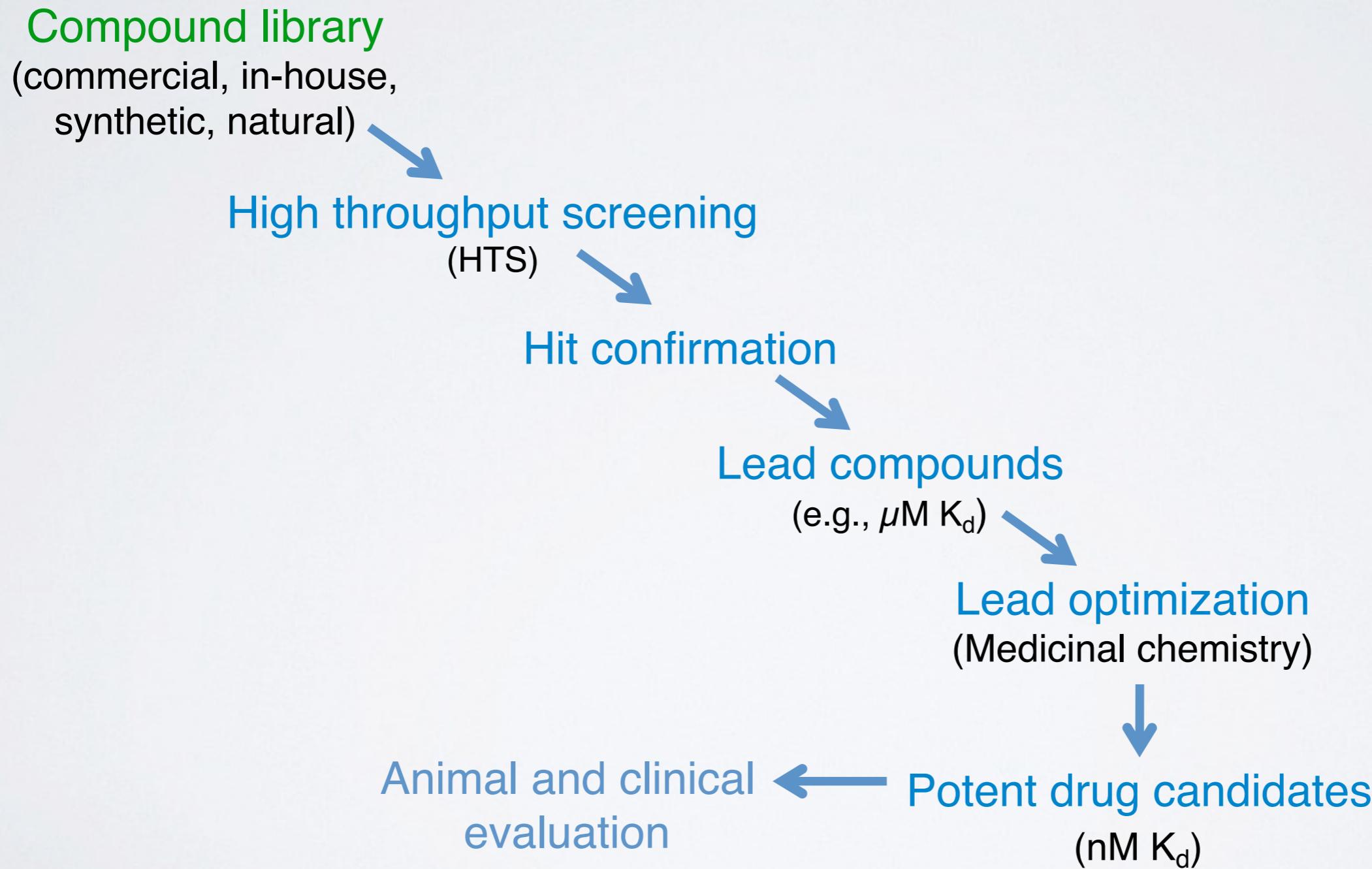
# Computer Aided Drug Discovery



# Next Up:

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# THE TRADITIONAL EMPIRICAL PATH TO DRUG DISCOVERY



# COMPUTER-AIDED DRUG DISCOVERY

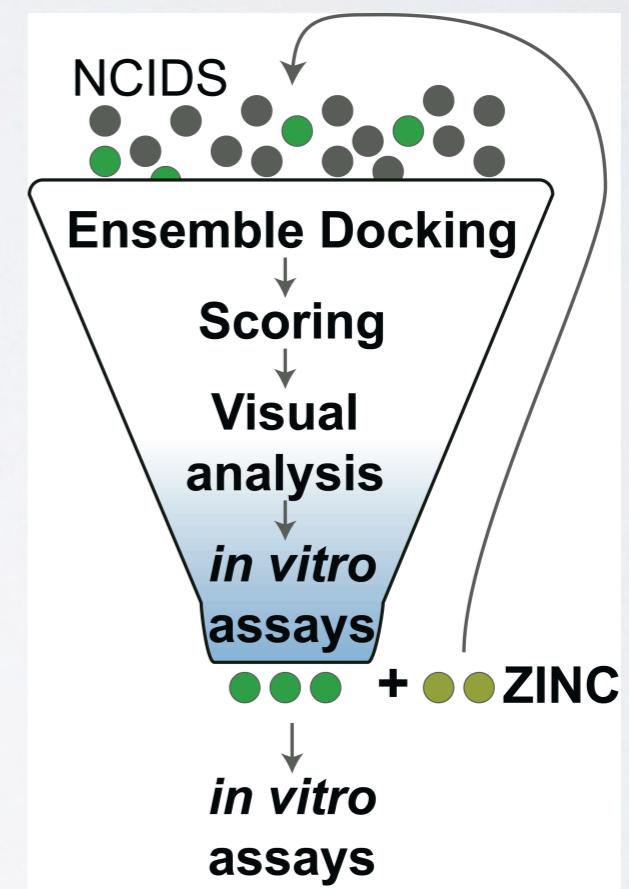
Aims to reduce number of compounds synthesized and assayed

Lower costs

Reduce chemical waste

Facilitate faster progress

N.B. Comparable experimental screens often out of reach of academia (facilities, cost)

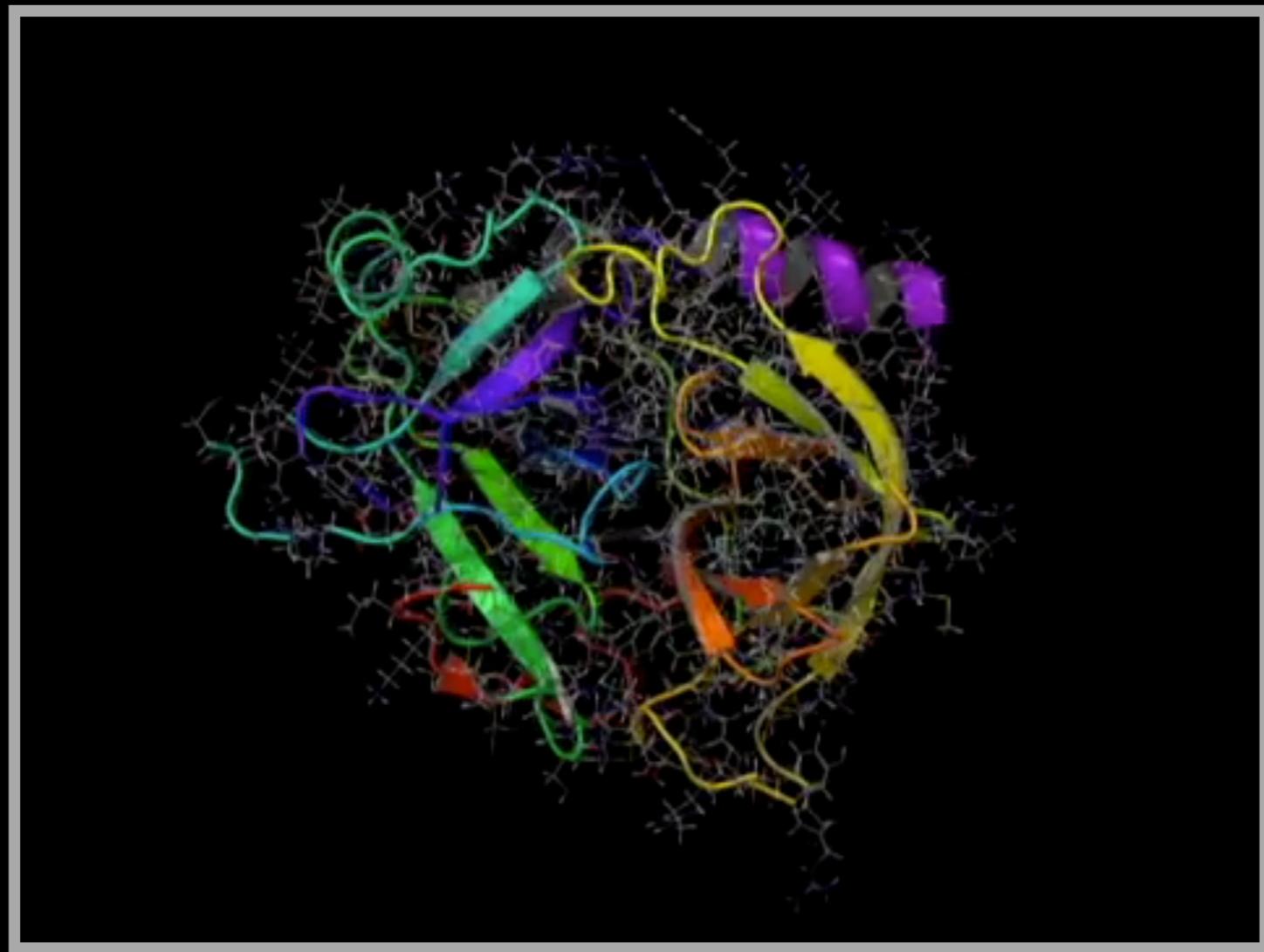


# Applications...

- Discriminate between good and poor binders, or provide a priority ranking to a collection of ligands
- Provide in-depth mechanistic characterization of specific ligand or group of ligands
- Provide valuable guidance for medicinal chemists trying to synthesize ligands with improved properties (affinities and potencies)

Q. “How can we modify an already active ligand to make it even more active?”

# Computational Ligand Docking



- Screening and ranking compounds as potential ligands (a.k.a. **virtual screening**)
- Improving "lead" compounds (a.k.a. **ligand optimization**, more on this later...)
  - This is a common practice among seasoned computational chemists

Two main approaches:

- (1). Receptor/Target-Based
- (2). Ligand/Drug-Based

Two main approaches:

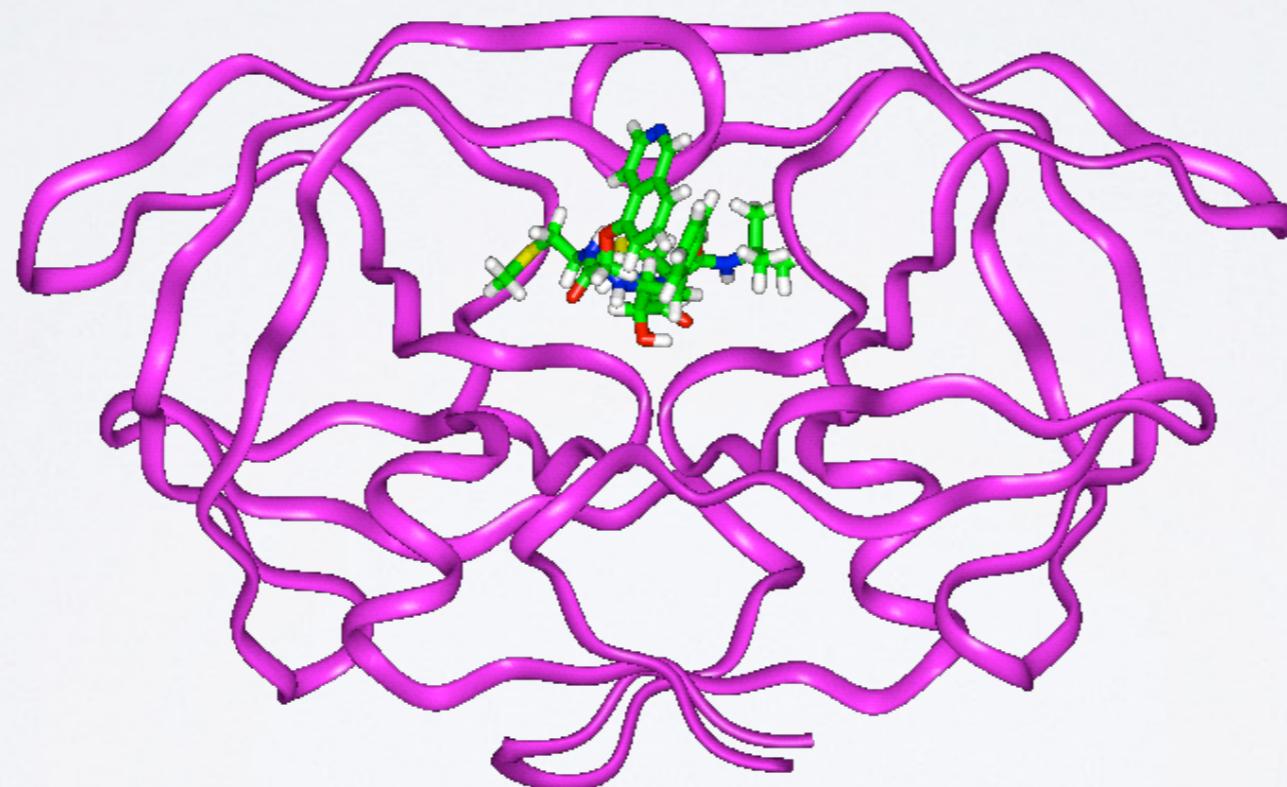
(1). Receptor/Target-Based

(2). Ligand/Drug-Based

# **SCENARIO I:**

## RECEPTOR-BASED DRUG DISCOVERY

Structure of Targeted Protein Known: **Structure-Based Drug Discovery**



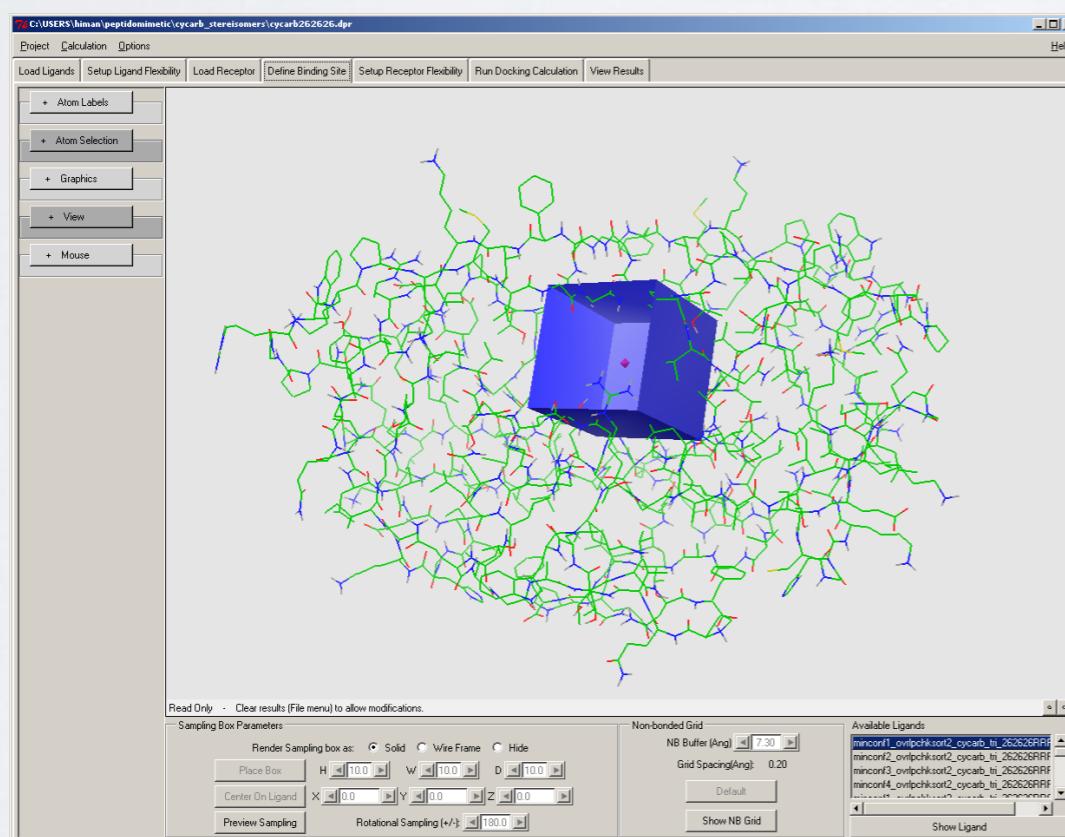
HIV Protease/KNI-272 complex

# PROTEIN-LIGAND DOCKING

## Structure-Based Ligand Design

### Docking software

Search for structure of lowest energy



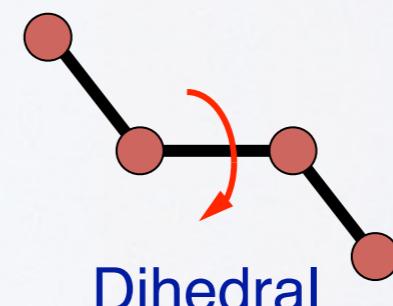
Potential function  
Energy as function of structure



VDW

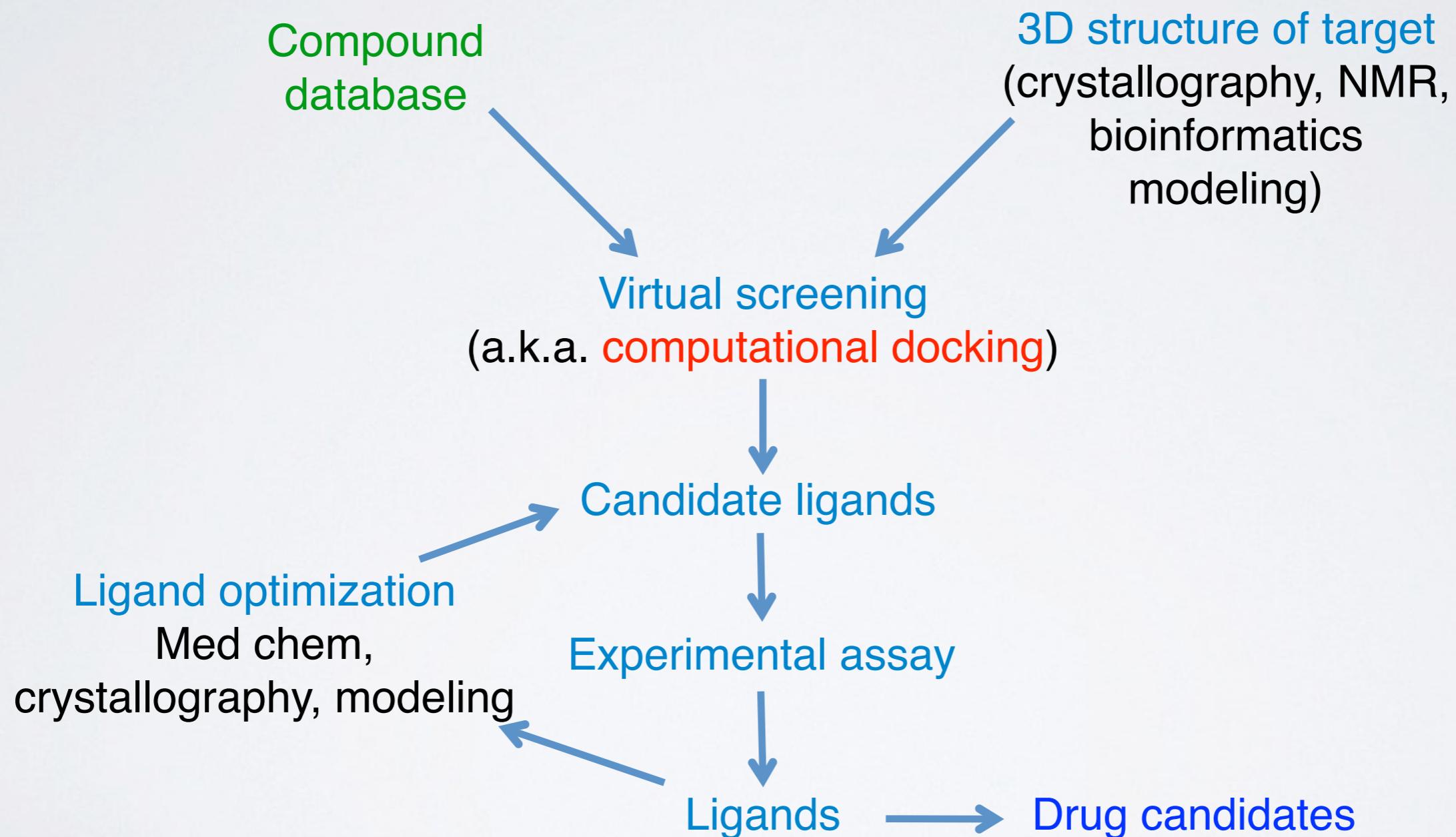


Screened Coulombic



Dihedral

# STRUCTURE-BASED VIRTUAL SCREENING



# COMPOUND LIBRARIES

**MAYBRIDGE**  
Part of Thermo Fisher Scientific

Search center  
Building Blocks  
Screening  
Select the catalog you want to query  
  
Building Blocks  
Screening

**Maybridge HitFinder™**  
This pre-selected diverse screening library makes identifying potential drug leads easy, convenient and cost effective.

Maximise quality hits from your screens  
• The HitFinder™ Collection comprises 14,400 premier compounds representing the drug-like diversity of the Maybridge Screening Collection, offering easy and rapid lead identification.  
• Selections are made using a clustering algorithm employing standard Daylight Fingerprints with the Tanimoto similarity index clustering at 0.71 similarity\*.

Reduced time to optimize any hit  
• All screening compounds fit Lipinski guidelines for "Drug-likeness\*\*", and all have purity greater than 90%.  
• Compounds have been selected to be non-reactive, ensuring fewer false positives and higher quality results.  
• When you are ready to optimize your drug lead, our range of over 6000 advanced novel Maybridge Building Blocks gives high chemical diversity for accelerating your drug design process.

**Ready to Screen**  
  
All HitFinder™ plates are securely sealed and carry both a clear plate number and bar-code for convenient use and storage.

To download the HitFinder™ database files [click here](#).  
To download the PDF file [click here](#).

**NIH MOLECULAR LIBRARIES**  
SMALL MOLECULE REPOSITORY

**A Roadmap Initiative**

**Welcome**

NIH Molecular Libraries Small Molecule Repository collects samples for high throughput biological screening and distributes them to the NIH Molecular Libraries Probe Production Centers Network. [Learn more](#).

MLSMR is a key component of the [Molecular Libraries Initiative](#), an [NIH Roadmap](#) project supporting [New Pathways to Discovery](#) in the 21<sup>st</sup> century. The project is funded in whole with Federal funds from the [National Institutes of Health](#), Department of Health and Human Services, under Contract No. HHS-N-278-2004-41001C.

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**PMLSC**  
**BIG DISCOVERIES FROM SMALL MOLECULES**

Welcome

The Pittsburgh Molecular Library Screening Center (PMLSC) comprises investigators at the University of Pittsburgh and Carnegie Mellon University. Its mission is to assist scientists and the National Institutes of Health to thoughtfully interrogate small molecule libraries using optical-based High Throughput and High Content assays.

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**Commercial**  
(in-house pharma)

**Government (NIH)**

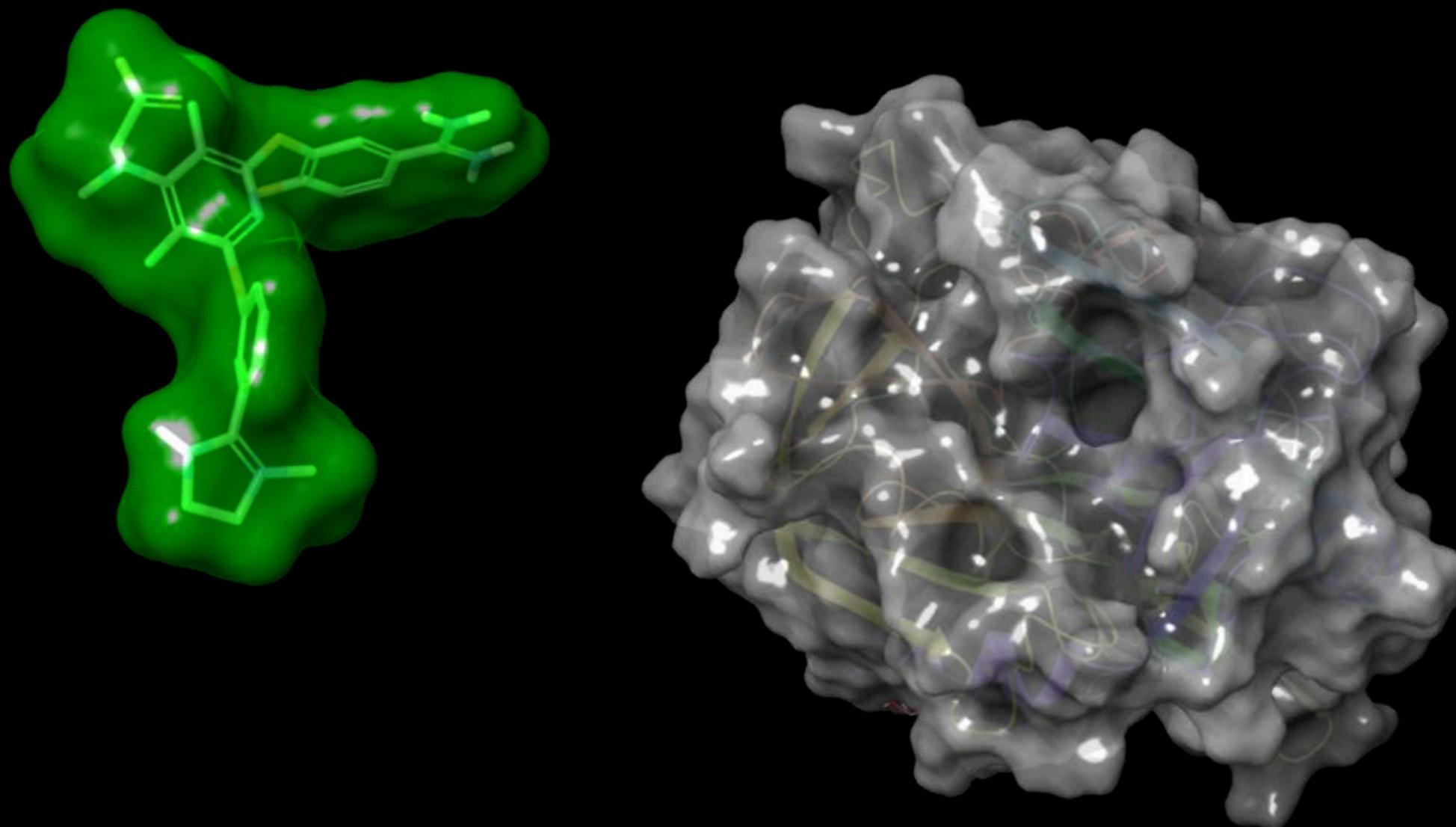
**Academia**

# Docking at its core is a shape matching problem

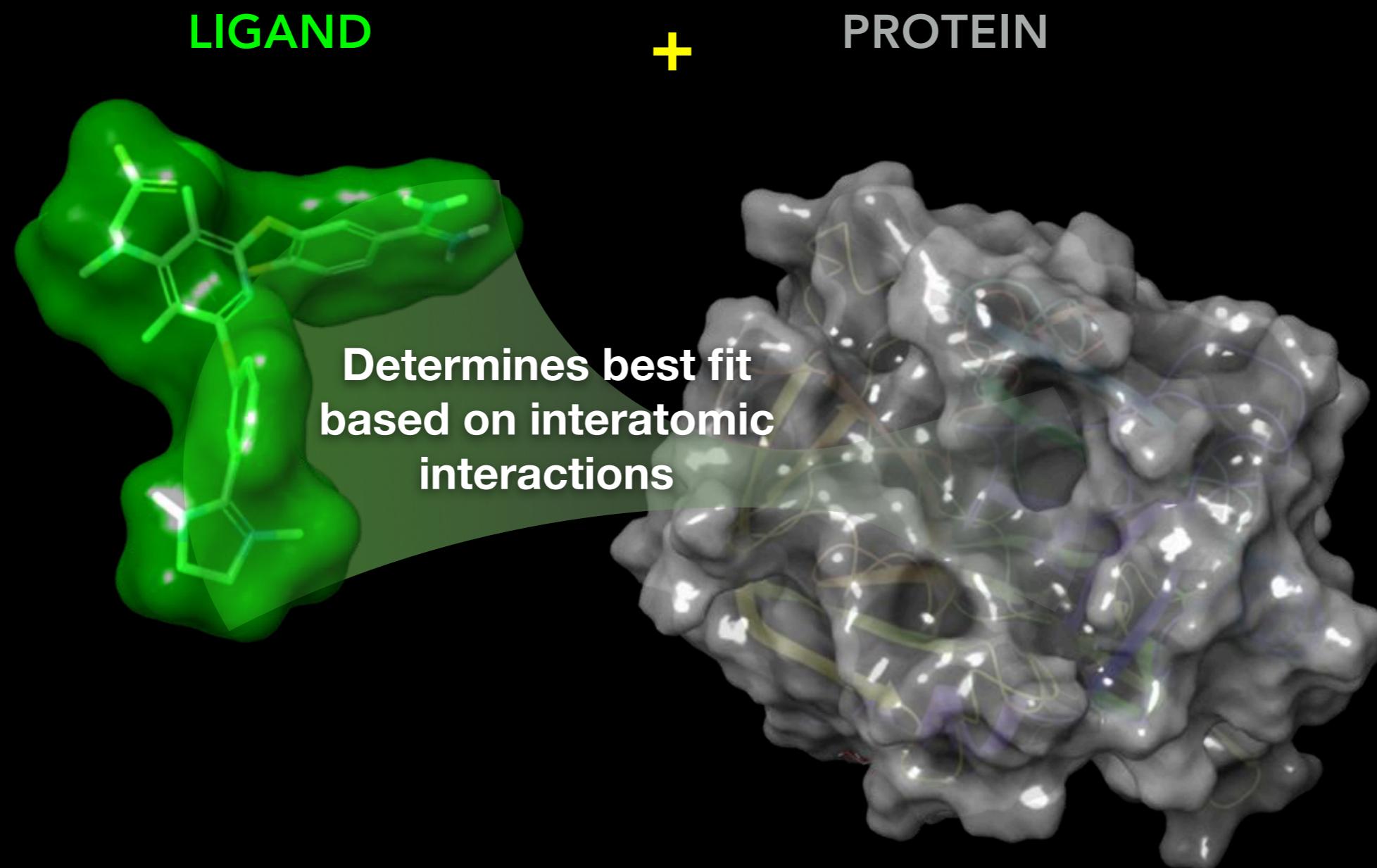
LIGAND

+

PROTEIN



# Docking at its core is a shape matching problem



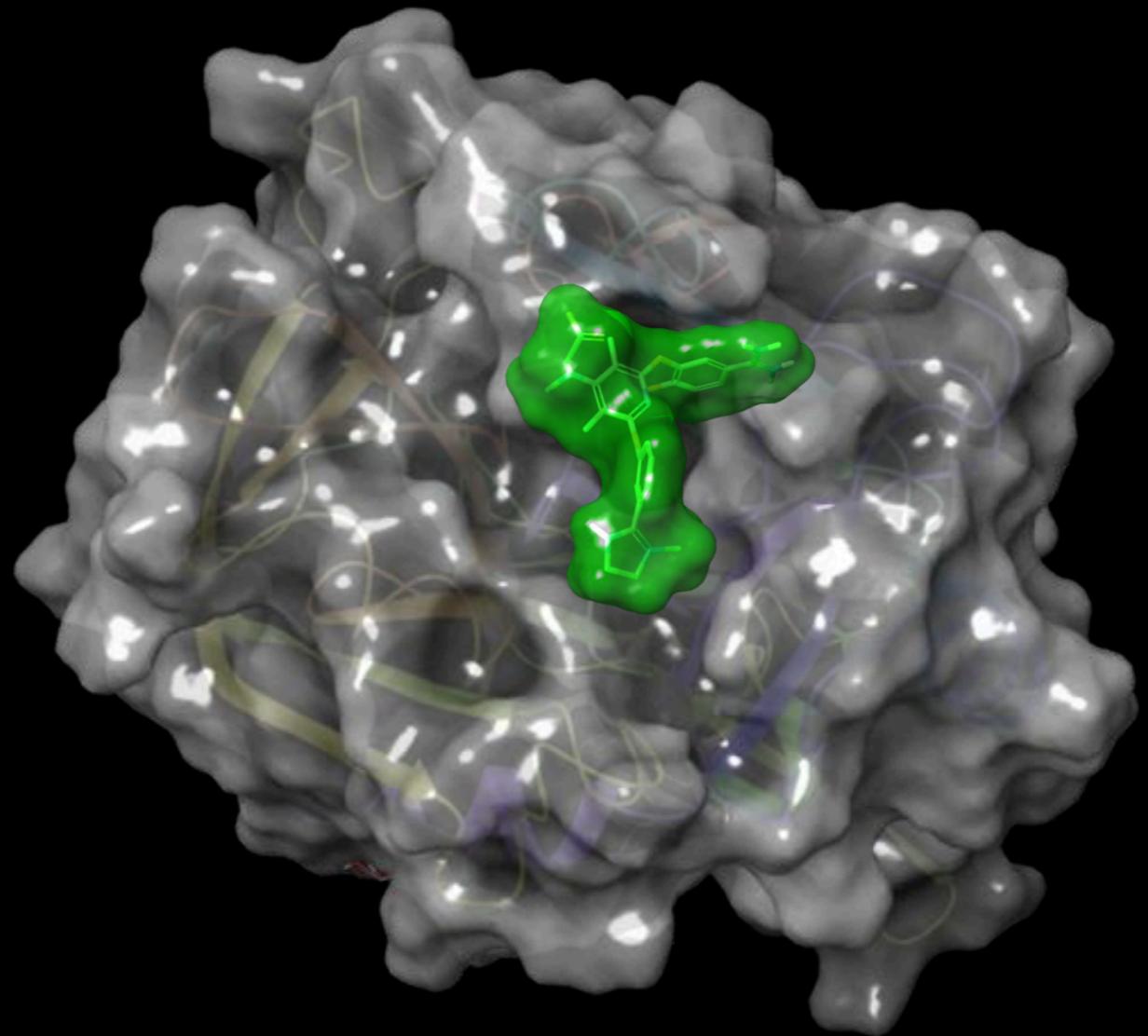
$$V(R) = E_{bonded} + E_{non.bonded}$$

## Bonding Interactions

- Bond length
- Bond angels
- Torsions

## Non-Bonding Interactions

- van der Waal's interactions
- H-bonds
- Charge-Charge interactions
- pi-pi, pi-cation, etc.



PROTEIN-**LIGAND**  
complex

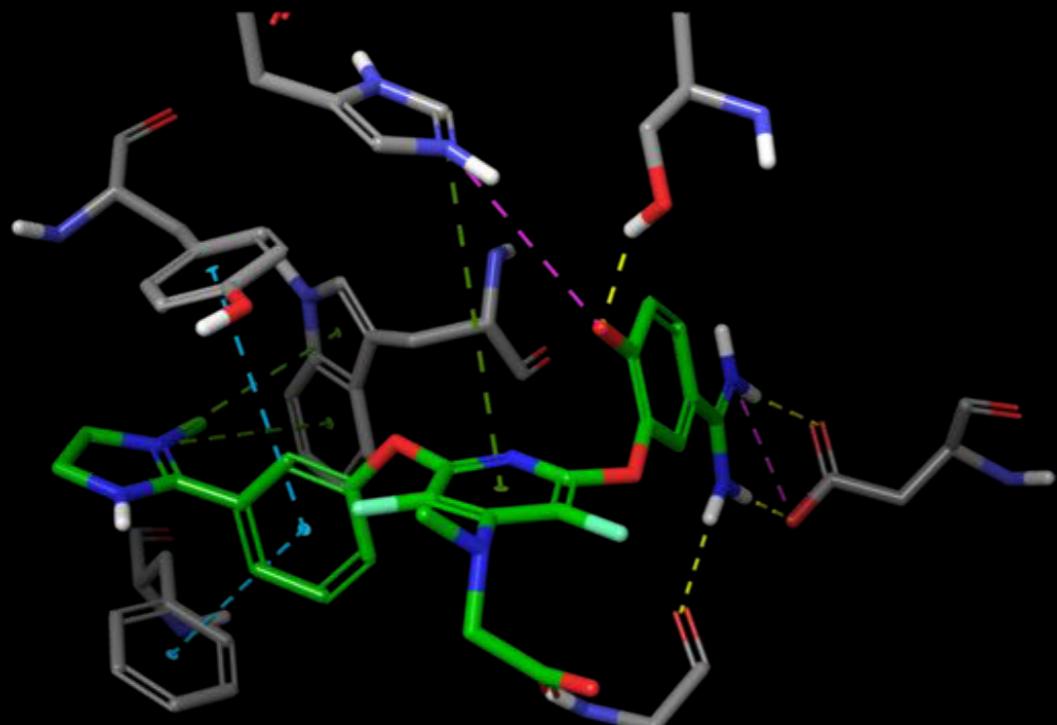
Do it Yourself!

# Hand-on time!

[https://bioboot.github.io/bggn213\\_F19/lectures/#12](https://bioboot.github.io/bggn213_F19/lectures/#12)

You can use the classroom computers or your own laptops. If you are using your laptops then you will need to install **MGLTools**

# A Docking Program Generates a...



## 1. Binding Pose

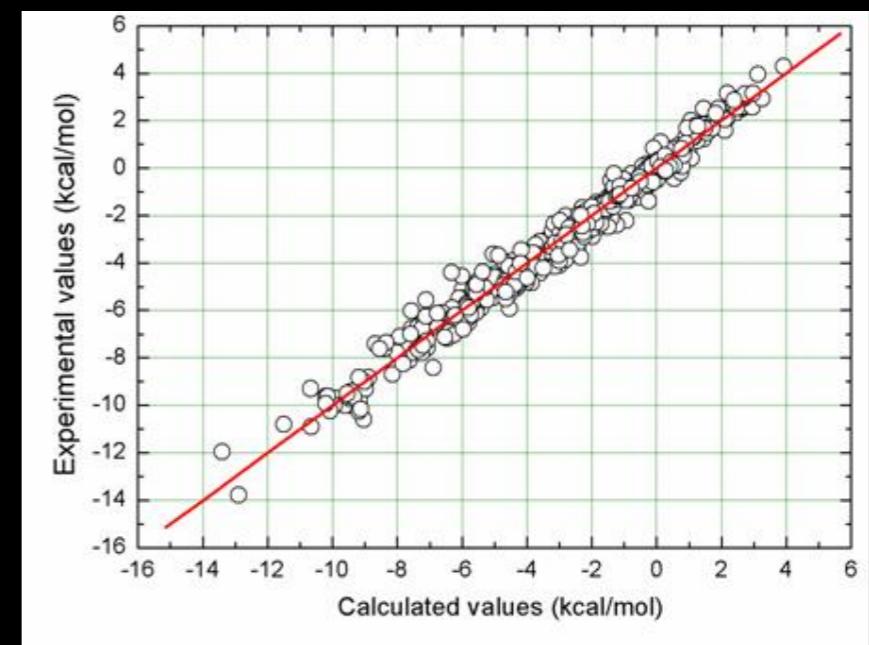
A model of the orientation of the ligand in the binding site of the receptor.

## 2. Docking Score

A numerical value representing the quality of the pose. Often presented as binding energy.

# Scoring functions enable different docking results to be compared

- Scoring functions aim to estimate ligand binding affinity, or the free energy of binding ( $\Delta G$ ), so that different poses can be compared
  - The poses with the most negative values are predicted to have the tightest interactions
- Scoring functions are constructed from a weighted sum of all possible molecular interactions that contribute to binding
  - Including H-bonds, van der Waals forces, electrostatic interactions, etc. and penalties for steric clashes and loss of entropy
- Scoring systems are optimized and validated by fitting to experimental values for known receptor-ligand interactions



# COMMON SIMPLIFICATIONS USED IN PHYSICS-BASED DOCKING

Quantum effects approximated classically

Protein often held rigid

Configurational entropy neglected

Influence of water treated crudely

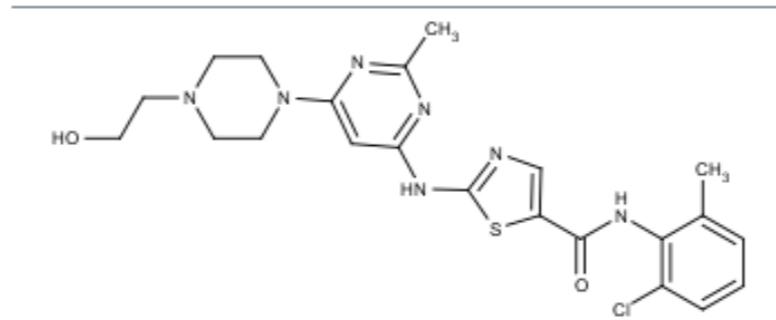
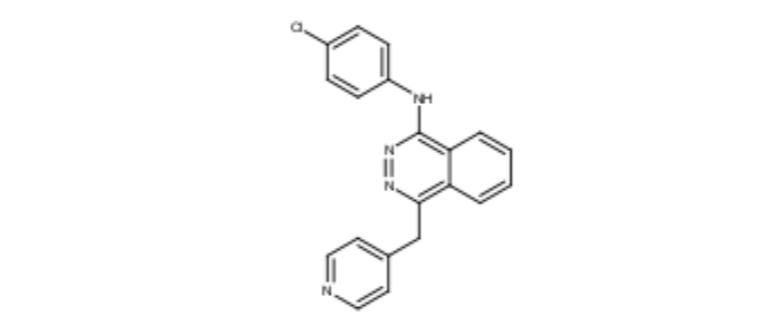
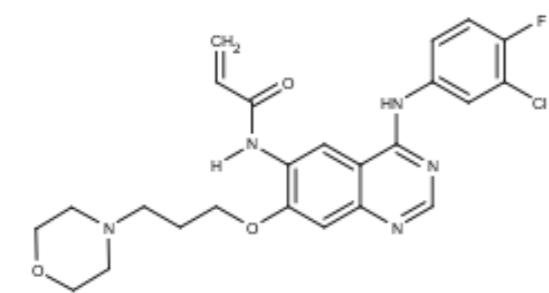
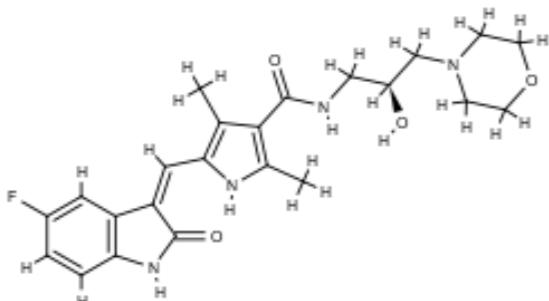
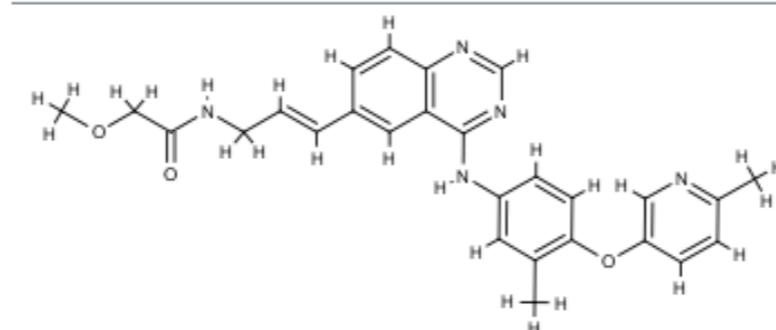
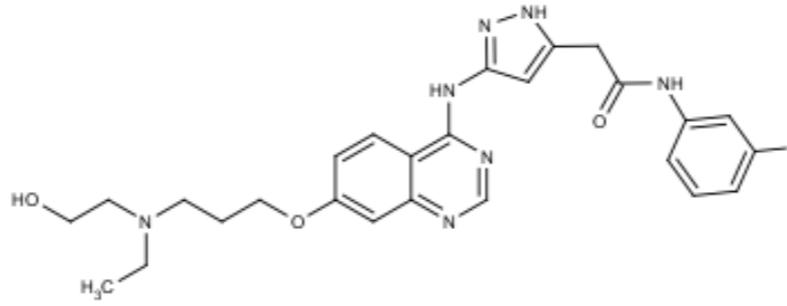
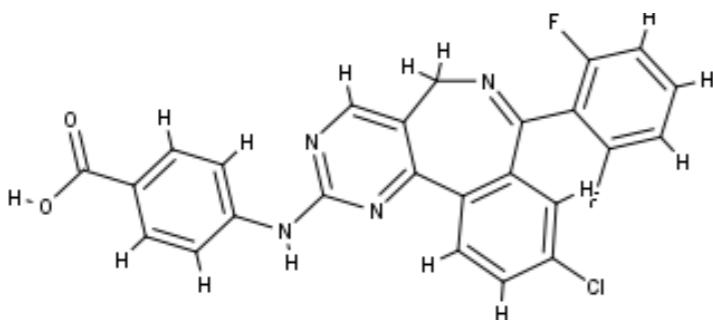
Two main approaches:

- (1). Receptor/Target-Based
- (2). Ligand/Drug-Based

# Scenario 2

## Structure of Targeted Protein Unknown: Ligand-Based Drug Discovery

e.g. MAP Kinase Inhibitors



Using knowledge of  
existing inhibitors to  
discover more

# Why Look for Another Ligand if You Already Have Some?

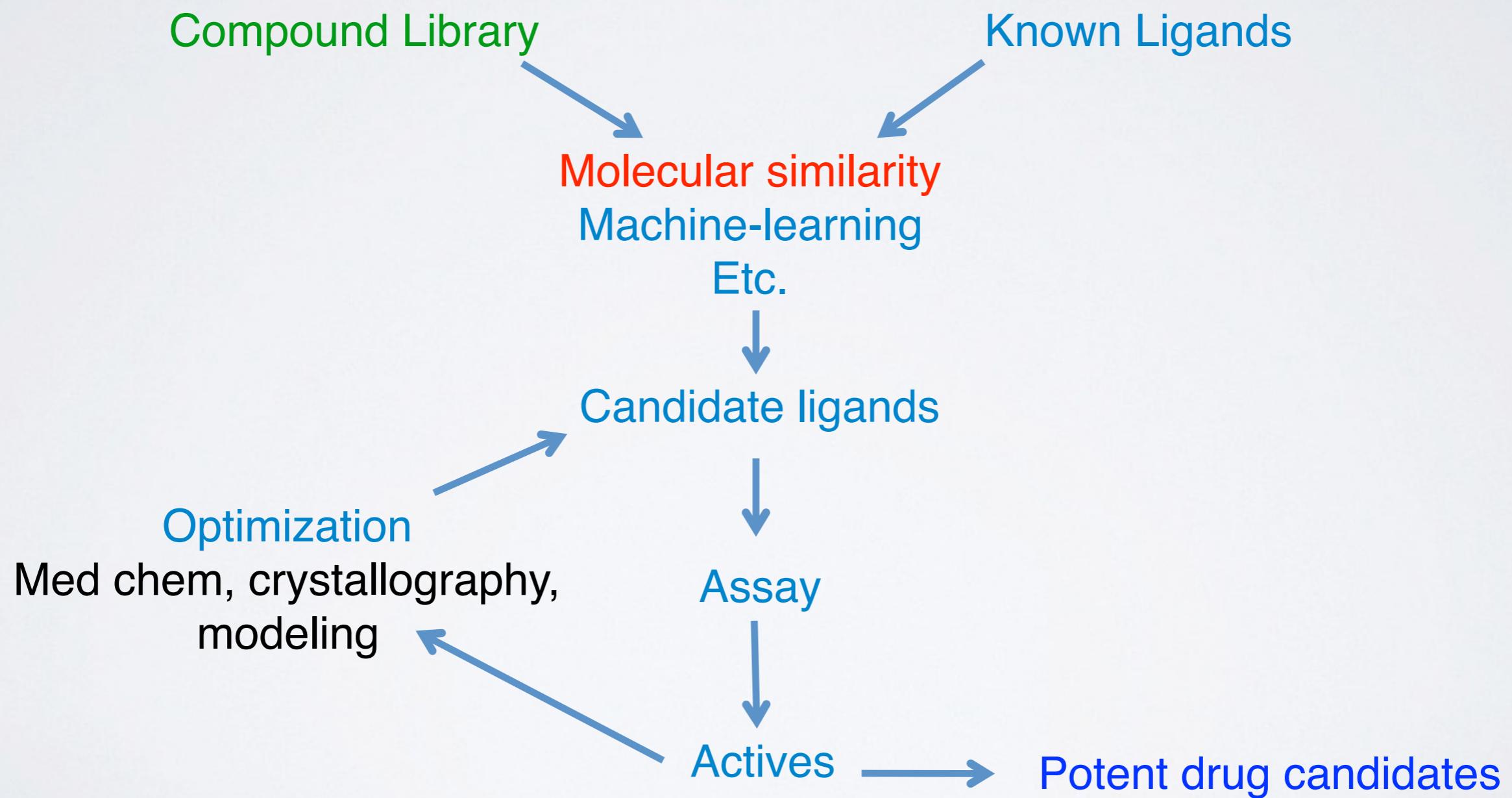
Experimental screening generated some ligands, but they don't bind tightly enough

A company wants to work around another company's chemical patents

An high-affinity ligand is toxic, is not well-absorbed, difficult to synthesize etc.

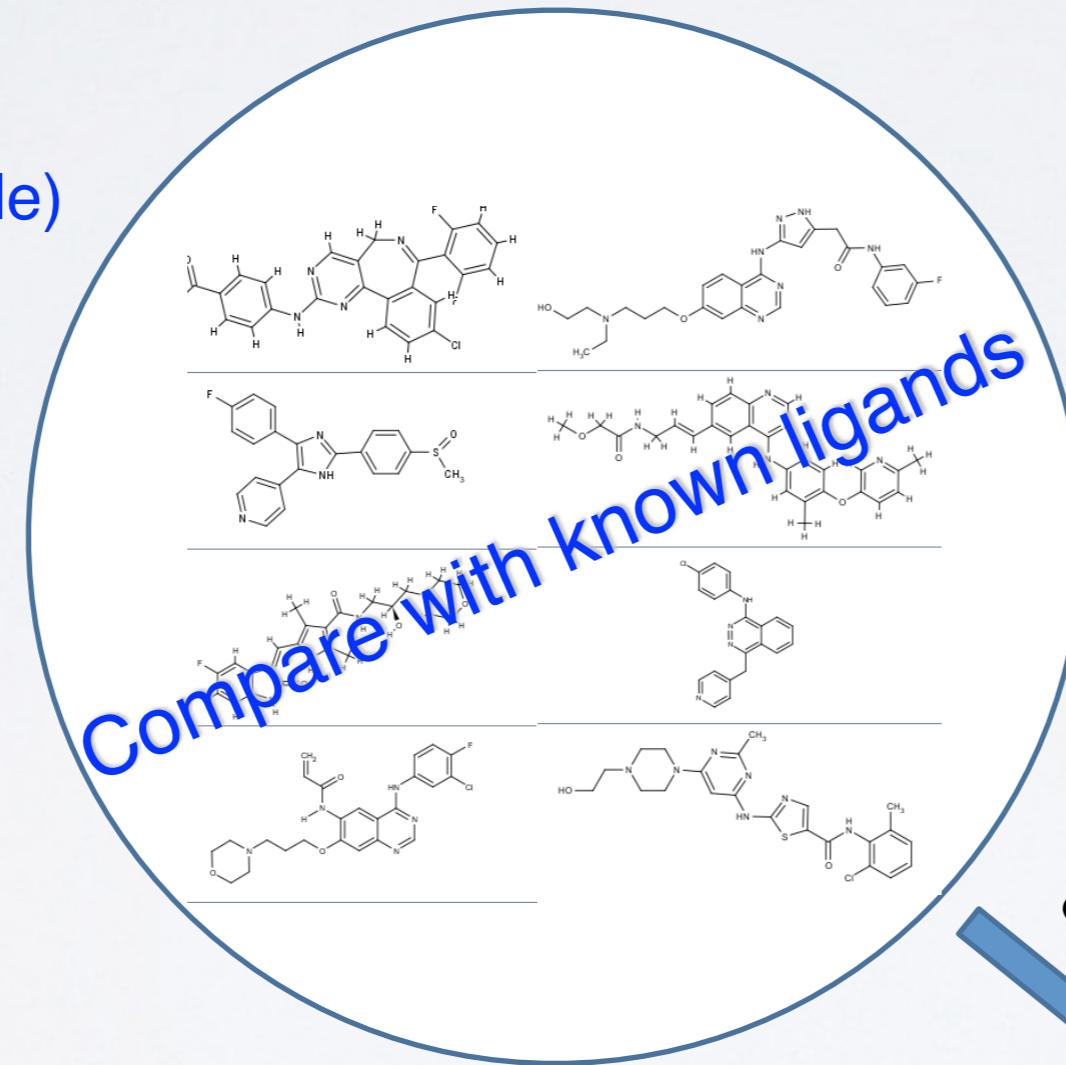
Drug resistance variants of the receptor have emerged...

# LIGAND-BASED VIRTUAL SCREENING



# CHEMICAL SIMILARITY LIGAND-BASED DRUG-DISCOVERY

Compounds  
(available/synthesizable)



Different

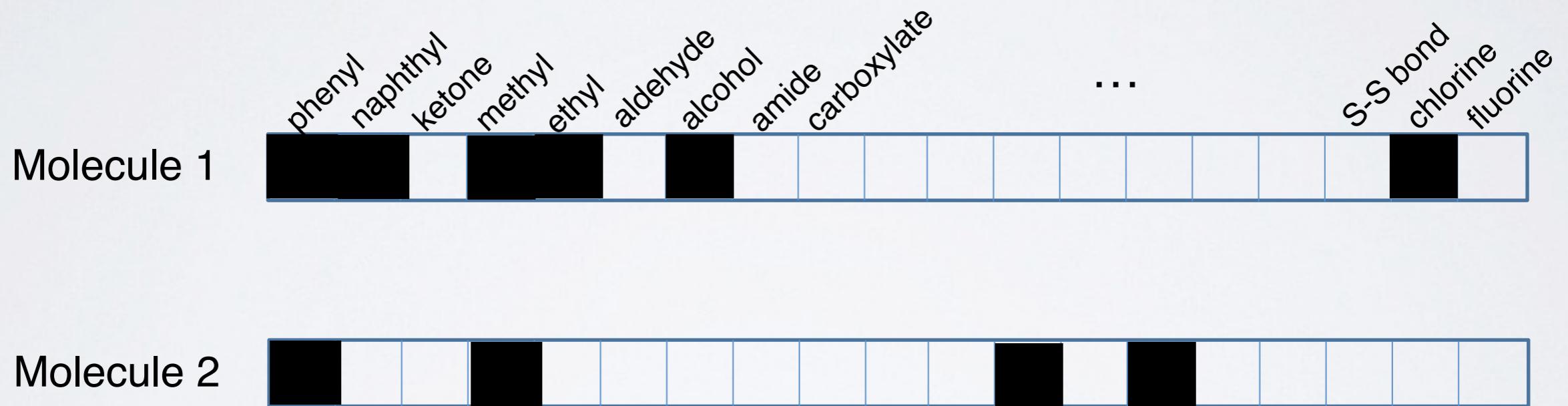
Don't bother

Similar

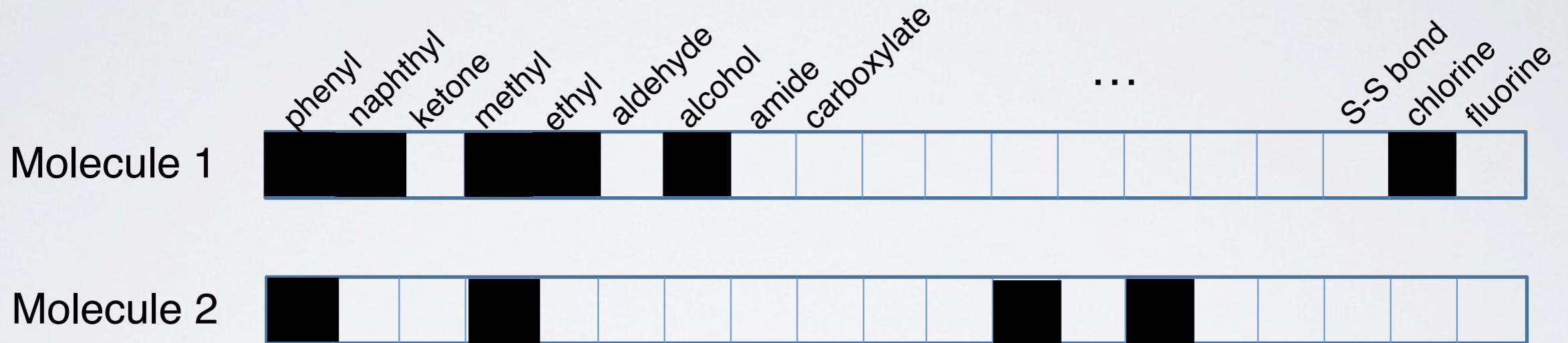
Test experimentally

# CHEMICAL FINGERPRINTS

## BINARY STRUCTURE KEYS



# CHEMICAL SIMILARITY FROM FINGERPRINTS



Tanimoto Similarity  
(or Jaccard Index), T

$$T \equiv \frac{N_I}{N_U} = 0.25$$

Intersection



$N_I=2$

Union

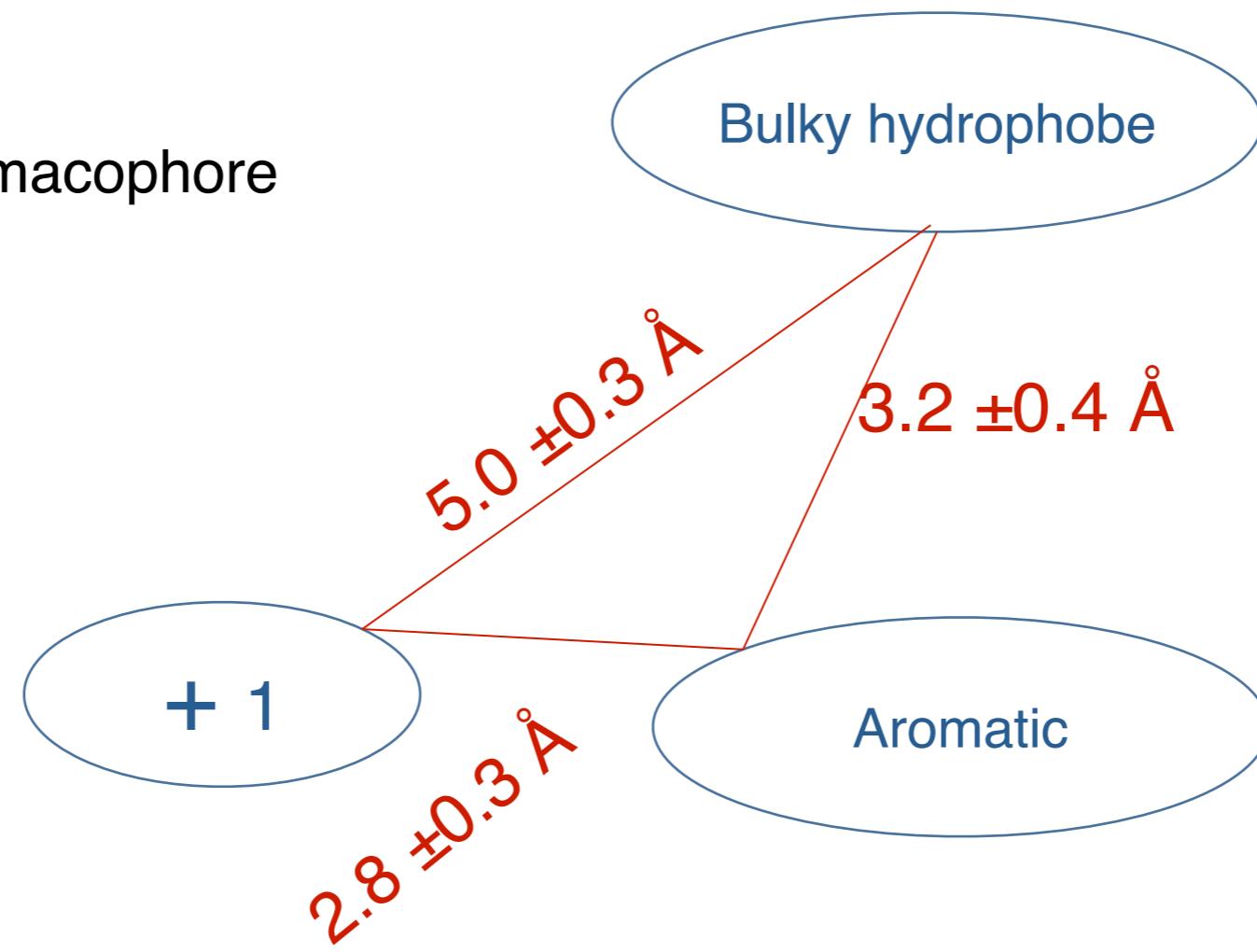


$N_U=8$

# Pharmacophore Models

Φάρμακο (drug) + Φορά (carry)

A 3-point pharmacophore



# Molecular Descriptors

## More abstract than chemical fingerprints

### Physical descriptors

molecular weight

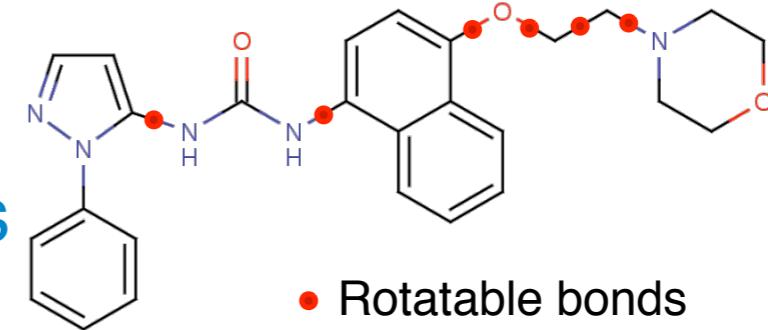
charge

dipole moment

number of H-bond donors/acceptors

number of rotatable bonds

hydrophobicity (log P and clogP)



- Rotatable bonds

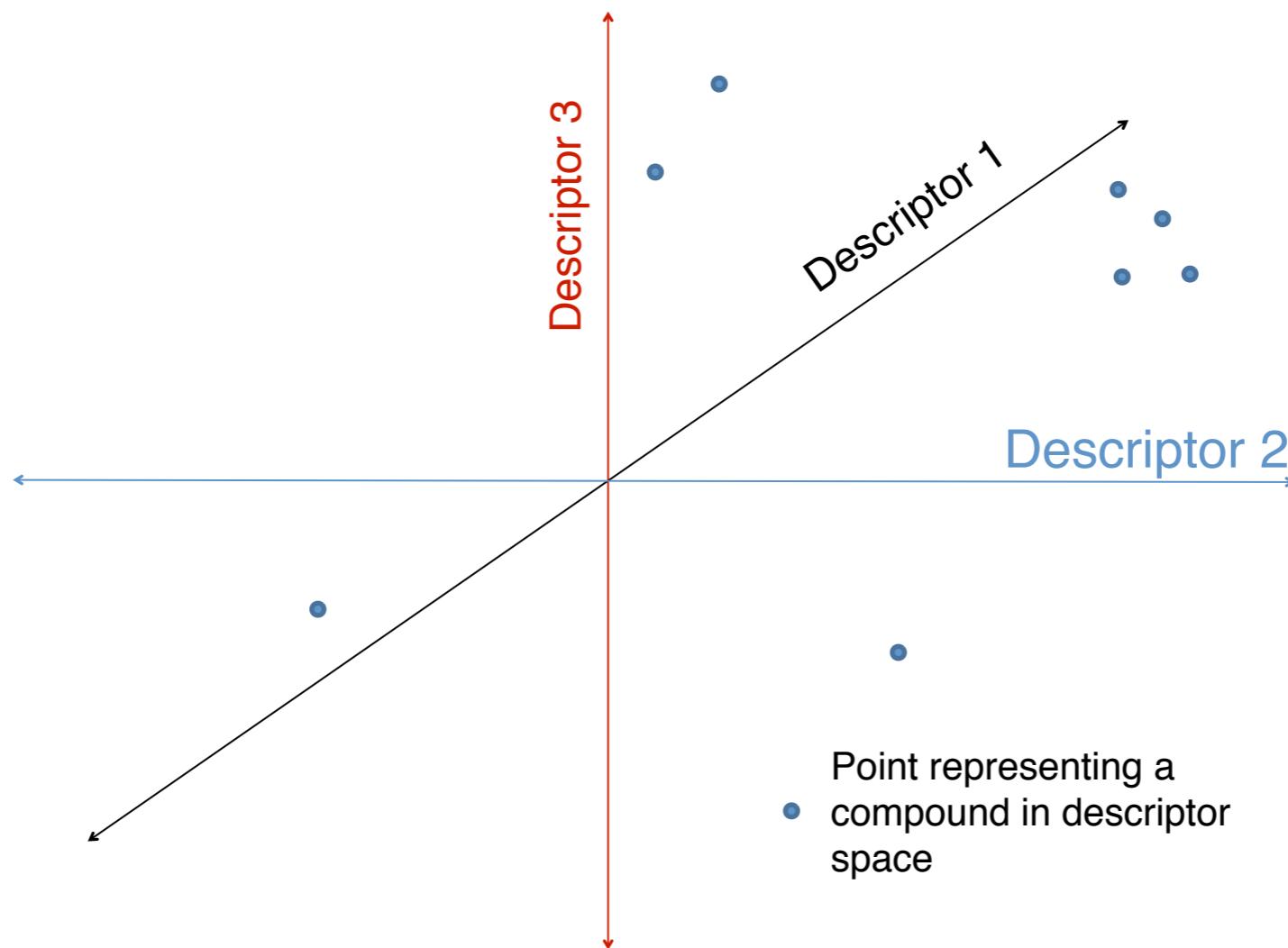
Topological  
branching index  
measures of linearity vs interconnectedness

Etc. etc.

# A High-Dimensional “Chemical Space”

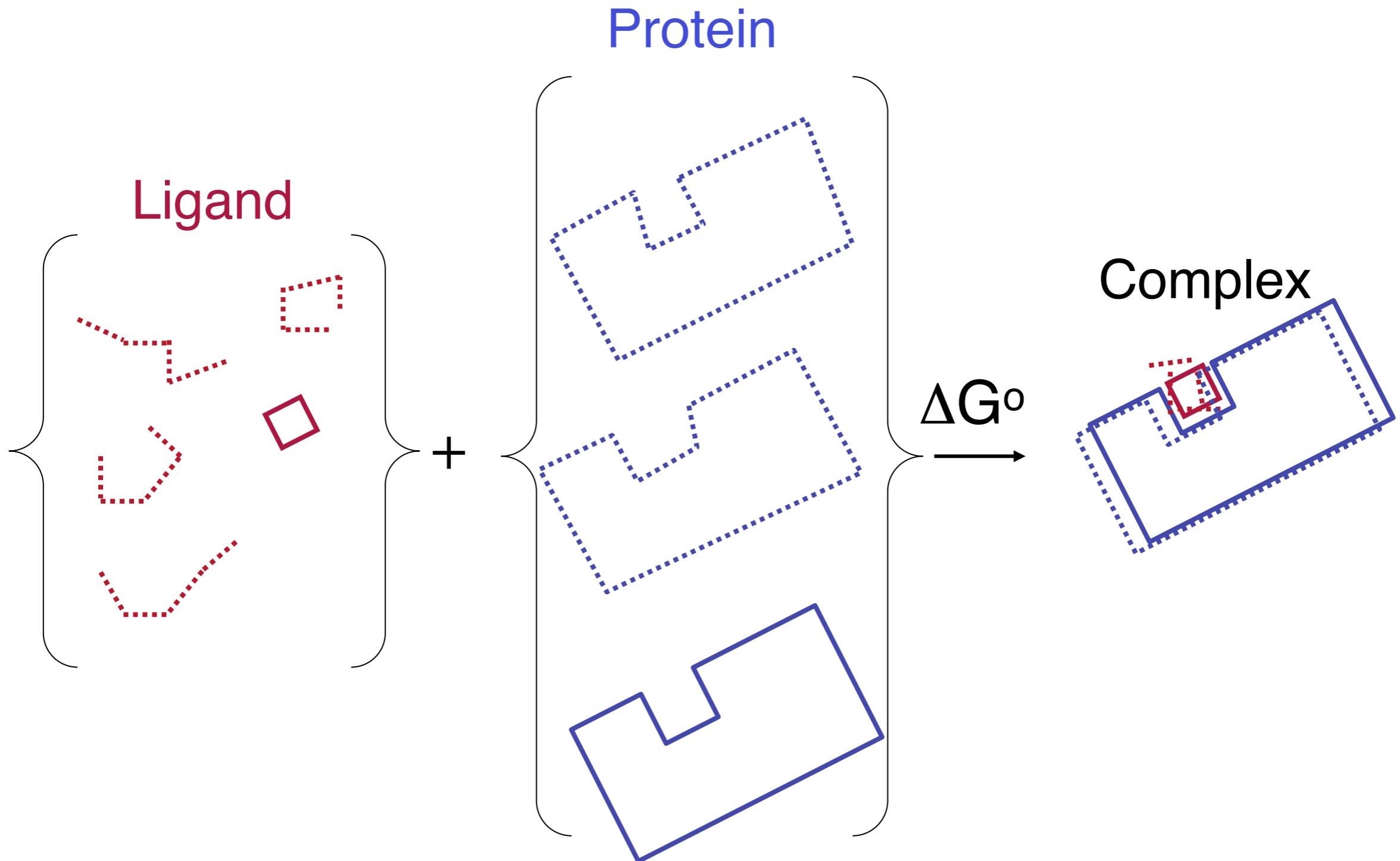
Each compound is a point in an n-dimensional space

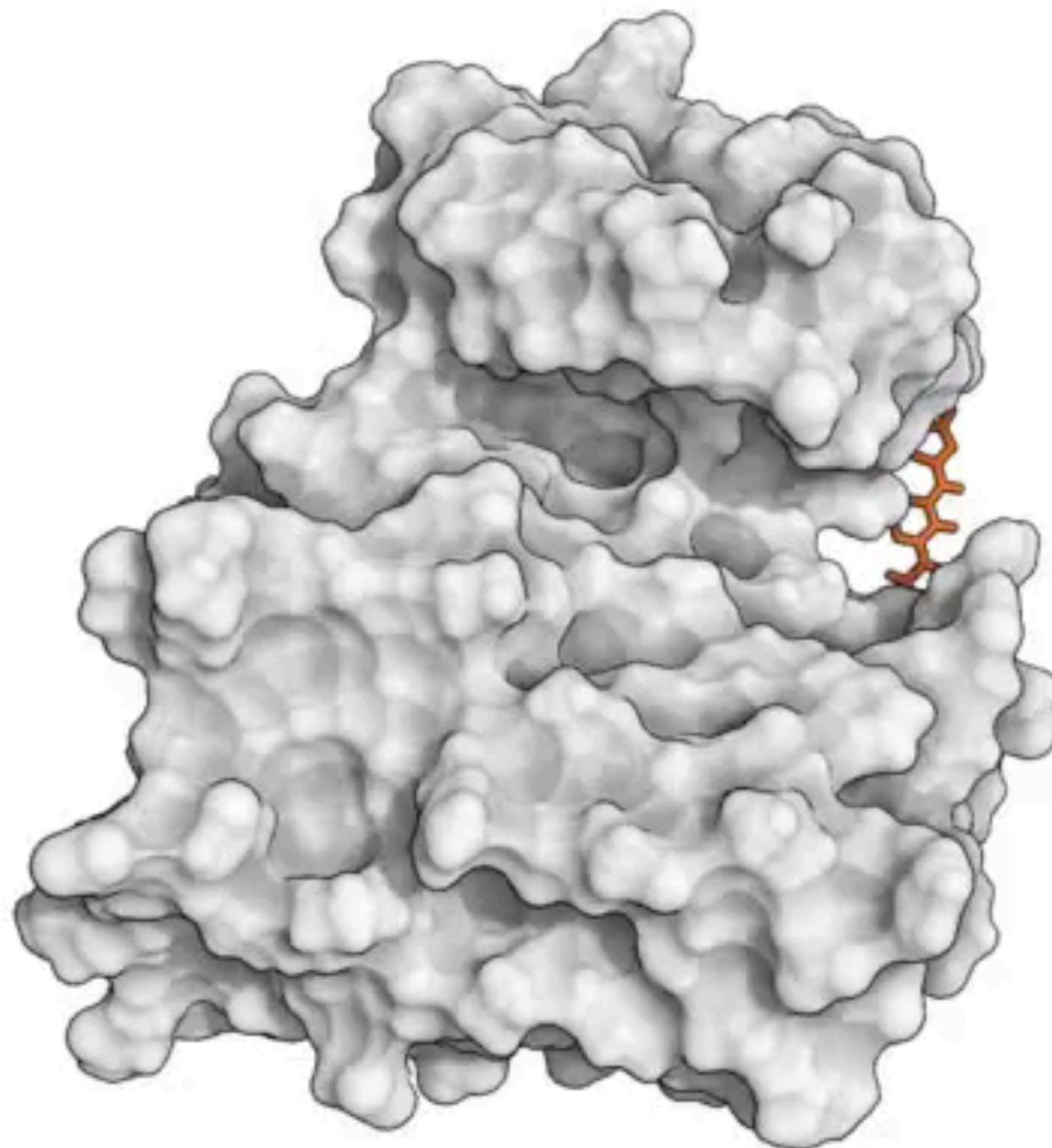
Compounds with similar properties are near each other



Apply multivariate statistics and machine learning for descriptor-selection. (e.g. partial least squares, PCA, support vector machines, random forest, deep learning etc.)

# Key Challenge: Proteins & Ligand are Flexible



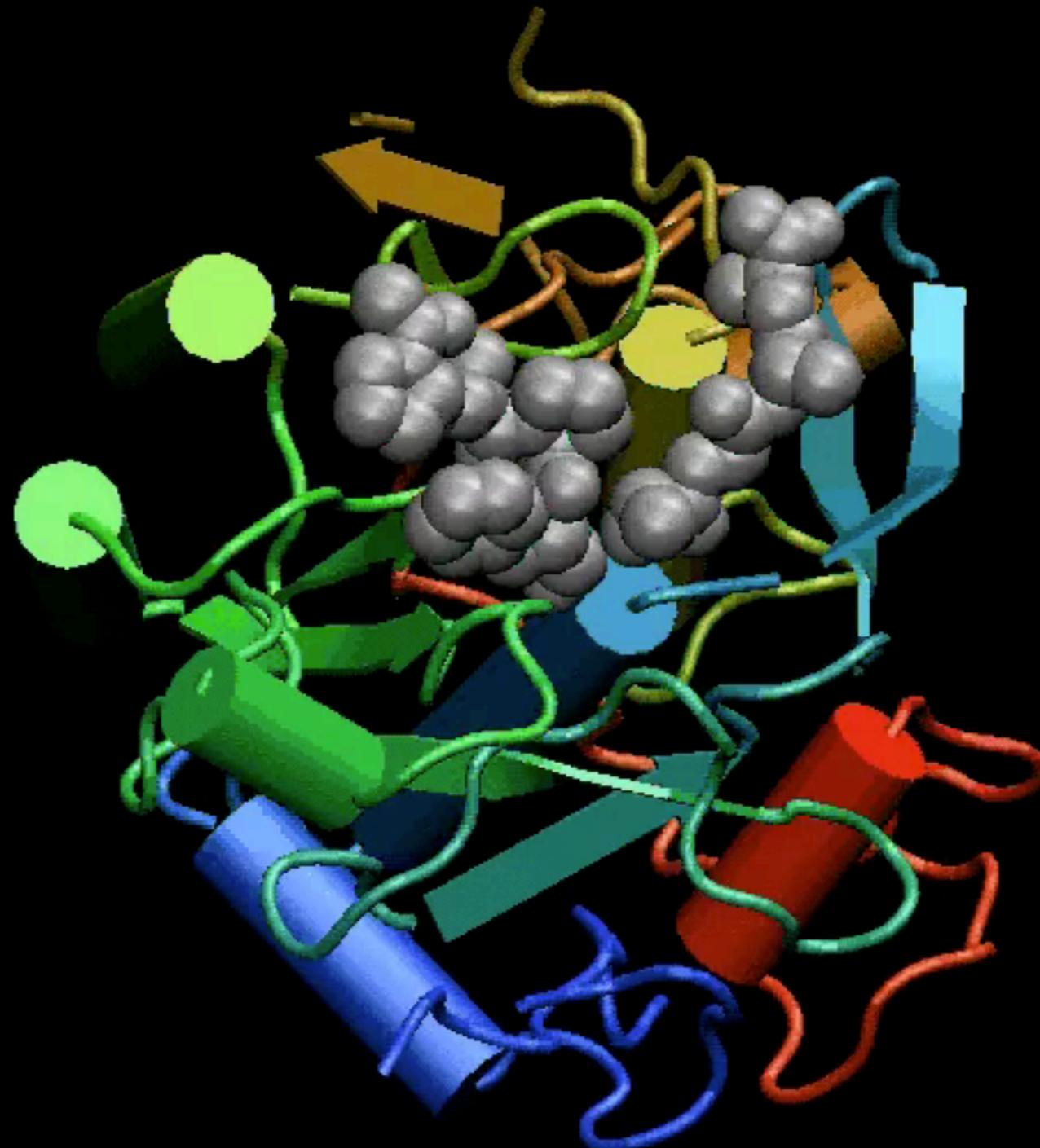


More on this later...

Proteins are flexible, which is a limitation in current rigid docking approaches... but when combined with **molecular dynamics** bioinformatics can be a powerful tool!

Do it Yourself!

**NMA** (Normal Mode Analysis) is a bioinformatics method to predict the intrinsic dynamics of biomolecules



[https://bioboot.github.io/bggn213\\_F19/lectures/#12](https://bioboot.github.io/bggn213_F19/lectures/#12)

# NMA in Bio3D

- Normal Mode Analysis (NMA) is a bioinformatics method that can predict the major motions of biomolecules.

```
pdb <- read.pdb("1hel")
modes <- nma( pdb )
m7 <- mktrj(modes, mode=7, file="mode_7.pdb")
```

Then you can open the resulting **mode\_7.pdb** file in **VMD**  
- Use "TUBE" representation and hit the play button...

Or use the **bio3d.view view()** function

```
library("bio3d.view")
view(m7, col=vec2color(rmsf(m7)))
```

# SUMMARY

- Structural bioinformatics is computer aided structural biology
- Described major motivations, goals and challenges of structural bioinformatics
- Reviewed the fundamentals of protein structure
- Explored how to use R to perform structural bioinformatics analysis!
- Introduced both physics and knowledge based modeling approaches for describing the structure, energetics and dynamics of proteins computationally
- Introduced both structure and ligand based bioinformatics approaches for drug discovery and design

# Reference Slides

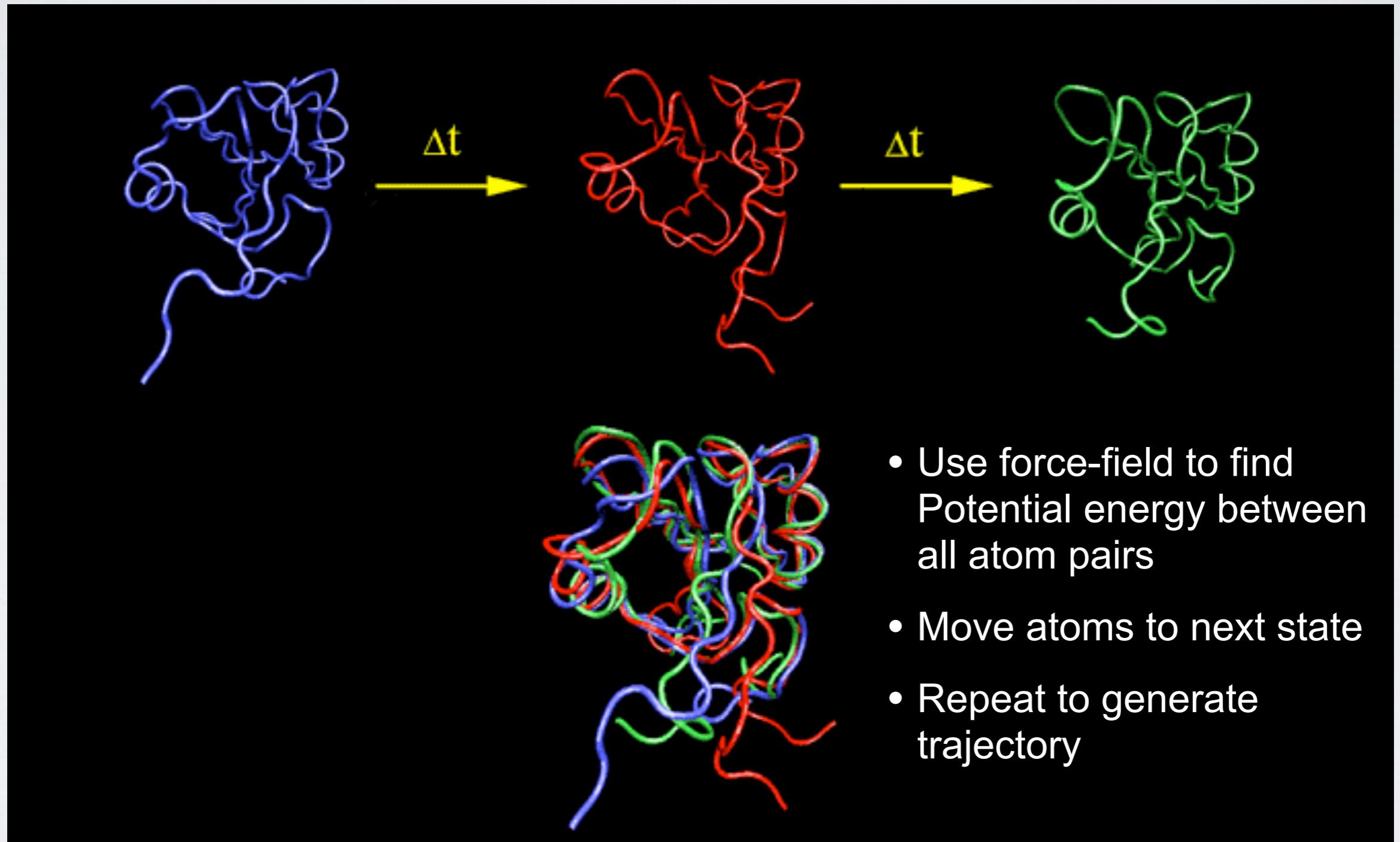
Molecular Dynamics (MD) and Normal Mode Analysis  
(NMA) Background and Cautionary Notes

[ Muddy Point Assessment ]

# PREDICTING FUNCTIONAL DYNAMICS

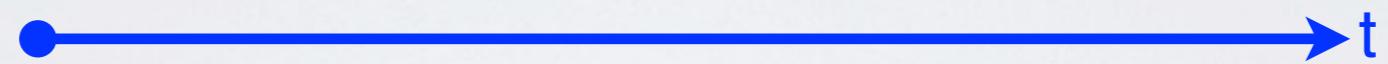
- Proteins are intrinsically flexible molecules with internal motions that are often intimately coupled to their biochemical function
  - E.g. ligand and substrate binding, conformational activation, allosteric regulation, etc.
- Thus knowledge of dynamics can provide a deeper understanding of the mapping of structure to function
  - Molecular dynamics (MD) and normal mode analysis (NMA) are two major methods for predicting and characterizing molecular motions and their properties

# MOLECULAR DYNAMICS SIMULATION



McCammon, Gelin & Karplus, *Nature* (1977)  
[ See: <https://www.youtube.com/watch?v=ui1ZysMFcKk> ]

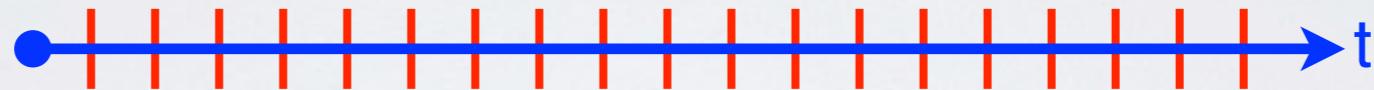
- ▶ Divide **time** into discrete ( $\sim 1\text{fs}$ ) **time steps** ( $\Delta t$ )  
(for integrating equations of motion, see below)



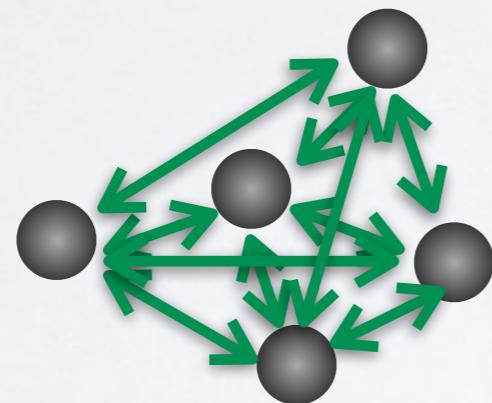
- ▶ Divide **time** into discrete ( $\sim 1\text{fs}$ ) **time steps** ( $\Delta t$ )  
(for integrating equations of motion, see below)



- ▶ Divide **time** into discrete ( $\sim 1\text{fs}$ ) **time steps** ( $\Delta t$ )  
(for integrating equations of motion, see below)



- ▶ At each time step calculate pair-wise atomic **forces** ( $F(t)$ )  
(by evaluating force-field gradient)



*Nucleic motion described classically*

$$m_i \frac{d^2}{dt^2} \vec{R}_i = -\vec{\nabla}_i E(\vec{R})$$

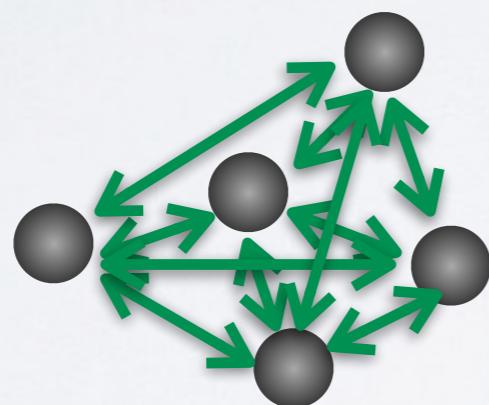
*Empirical force field*

$$E(\vec{R}) = \sum_{\text{bonded}} E_i(\vec{R}) + \sum_{\text{non-bonded}} E_i(\vec{R})$$

- ▶ Divide time into discrete ( $\sim 1\text{fs}$ ) time steps ( $\Delta t$ )  
(for integrating equations of motion, see below)



- ▶ At each time step calculate pair-wise atomic forces ( $F(t)$ )  
(by evaluating force-field gradient)



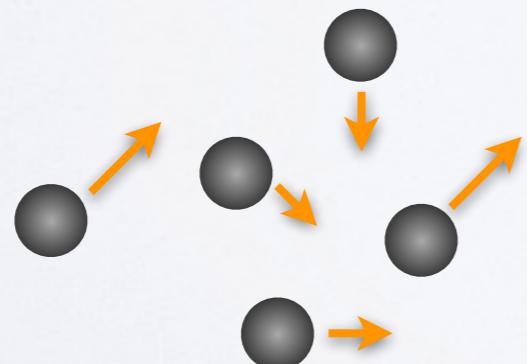
*Nucleic motion described classically*

$$m_i \frac{d^2}{dt^2} \vec{R}_i = -\vec{\nabla}_i E(\vec{R})$$

*Empirical force field*

$$E(\vec{R}) = \sum_{\text{bonded}} E_i(\vec{R}) + \sum_{\text{non-bonded}} E_i(\vec{R})$$

- ▶ Use the forces to calculate velocities and move atoms to new positions  
(by integrating numerically via the “leapfrog” scheme)

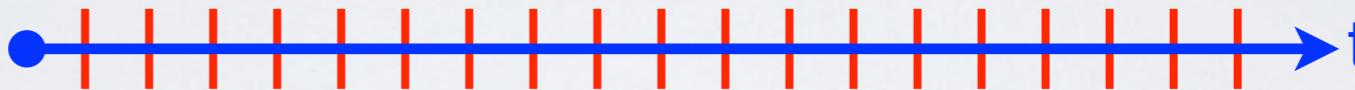


$$\boxed{v(t + \frac{\Delta t}{2})} = v(t - \frac{\Delta t}{2}) + \frac{F(t)}{m} \Delta t$$

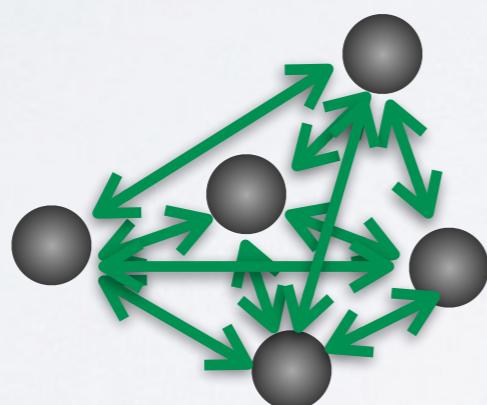
$$r(t + \Delta t) = r(t) + \boxed{v(t + \frac{\Delta t}{2})} \Delta t$$

# BASIC ANATOMY OF A MD SIMULATION

- ▶ Divide **time** into discrete ( $\sim 1\text{fs}$ ) **time steps** ( $\Delta t$ )  
(for integrating equations of motion, see below)



- ▶ At each time step calculate pair-wise atomic **forces** ( $F(t)$ )  
(by evaluating force-field gradient)



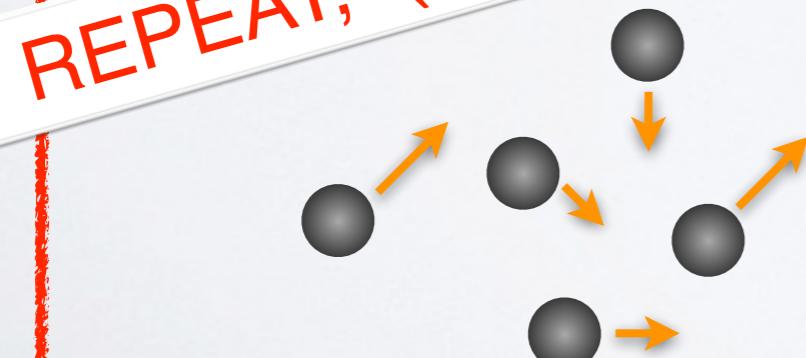
*Nucleic motion described classically*

$$m_i \frac{d^2}{dt^2} \vec{R}_i = -\vec{\nabla}_i E(\vec{R})$$

*Empirical force function*

$$E(\vec{R}) = \sum_{\text{bonded}} E_b(\vec{R}) + \sum_{\text{non-bonded}} E_n(\vec{R})$$

- ▶ Use the forces to calculate velocities and move atoms to new positions  
(numerically via the “leapfrog” scheme)

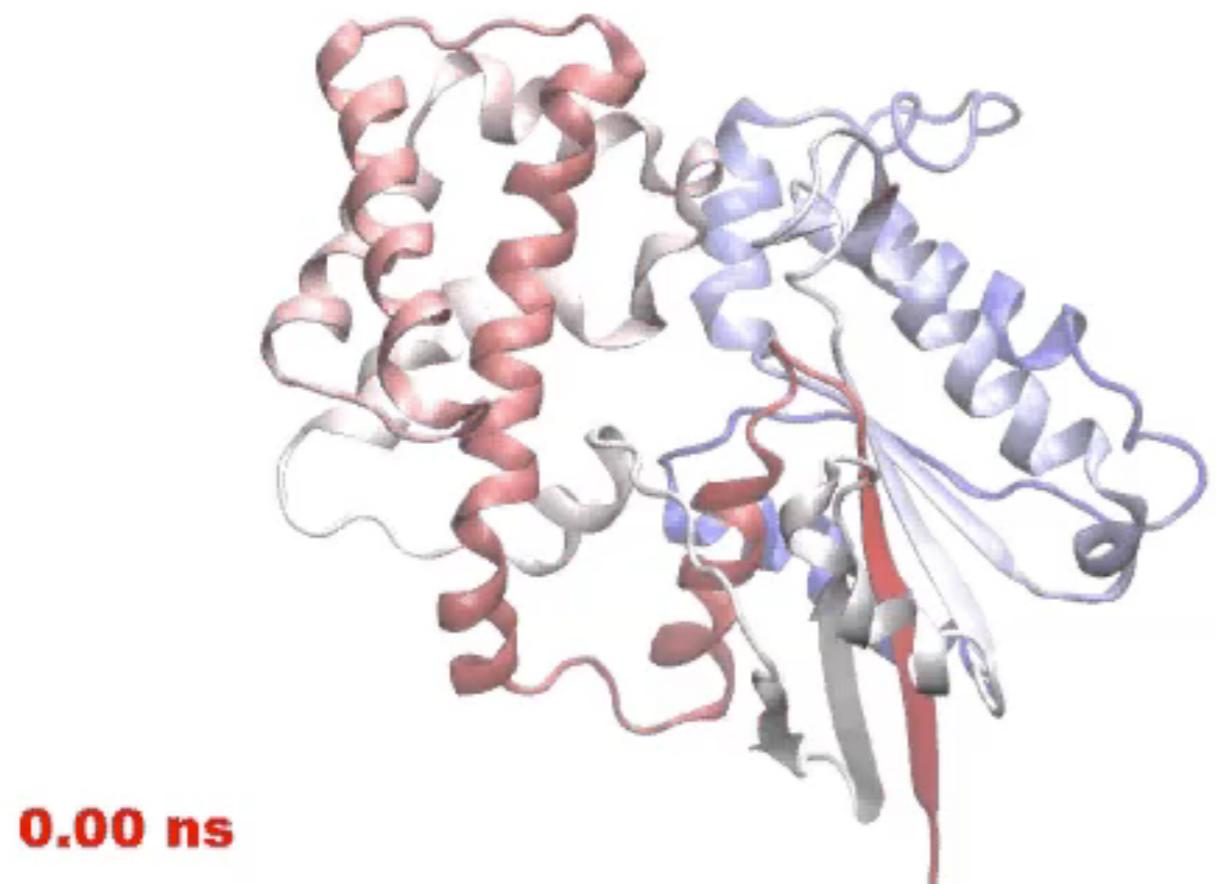


$$\begin{aligned} \mathbf{v}(t + \frac{\Delta t}{2}) &= \mathbf{v}(t - \frac{\Delta t}{2}) + \frac{\mathbf{F}(t)}{m} \Delta t \\ \mathbf{r}(t + \Delta t) &= \mathbf{r}(t) + \mathbf{v}(t + \frac{\Delta t}{2}) \Delta t \end{aligned}$$

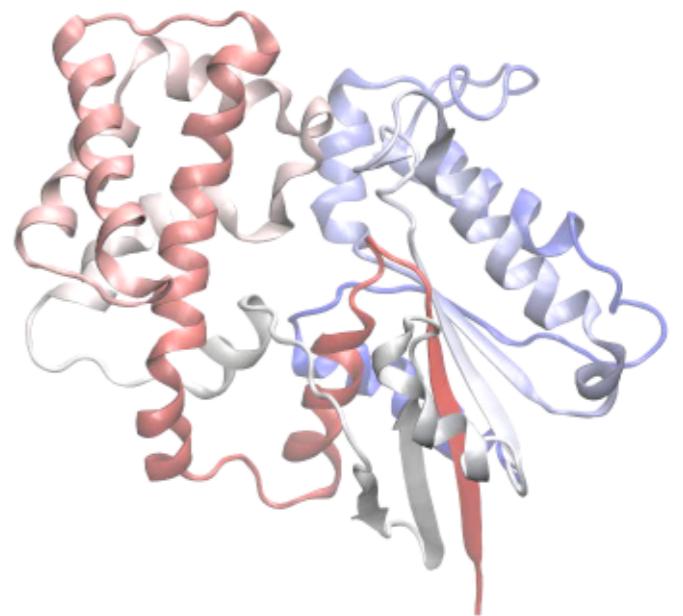
**REPEAT, (iterate many, many times... 1ms =  $10^{12}$  time steps)**

# MD Prediction of Functional Motions

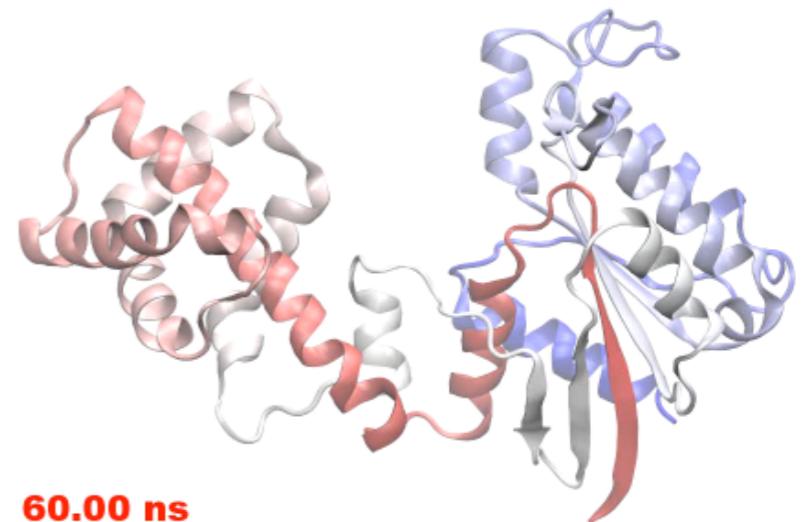
Accelerated MD simulation of  
nucleotide-free transducin alpha subunit



“close”

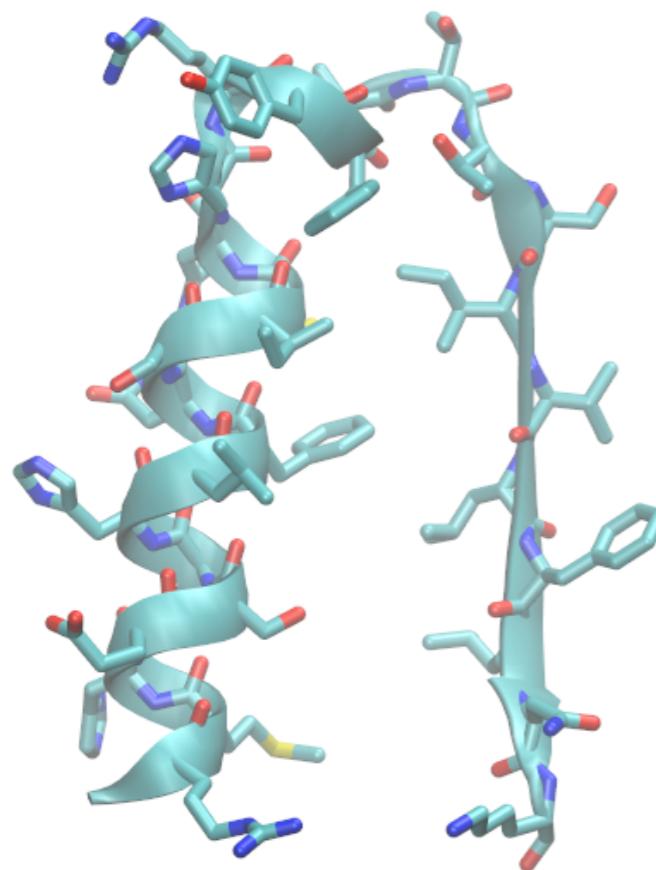


“open”

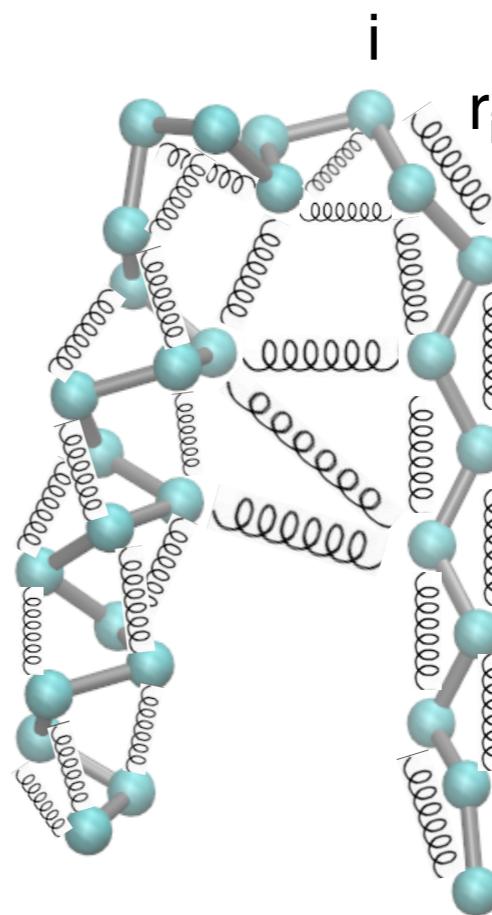
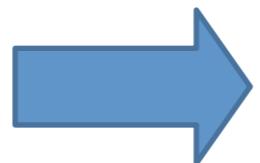


# COARSE GRAINING: **NORMAL MODE ANALYSIS** (NMA)

- MD is still time-consuming for large systems
- Elastic network model NMA (ENM-NMA) is an example of a lower resolution approach that finishes in seconds even for large systems.



C. G.

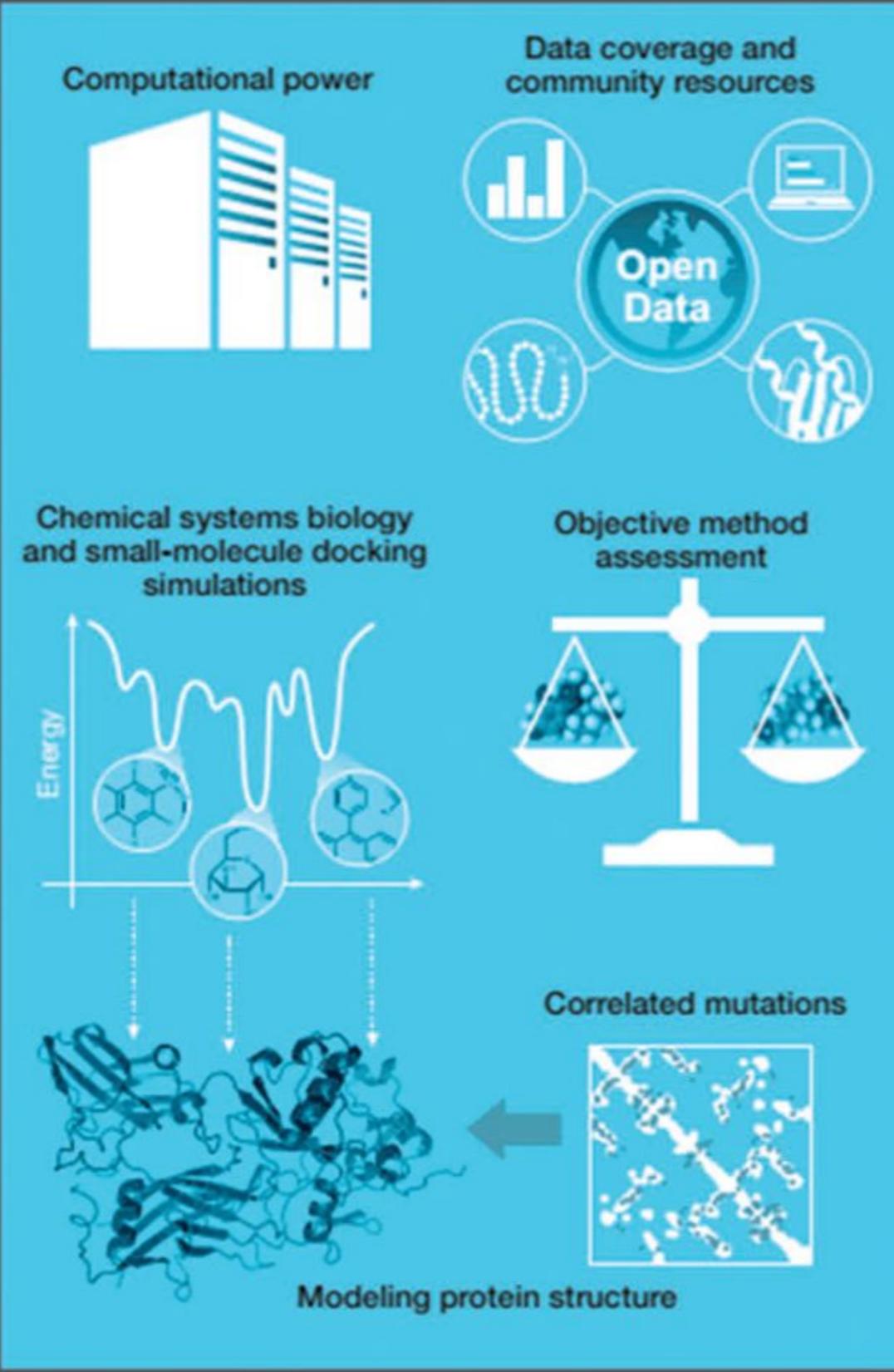


Atomistic

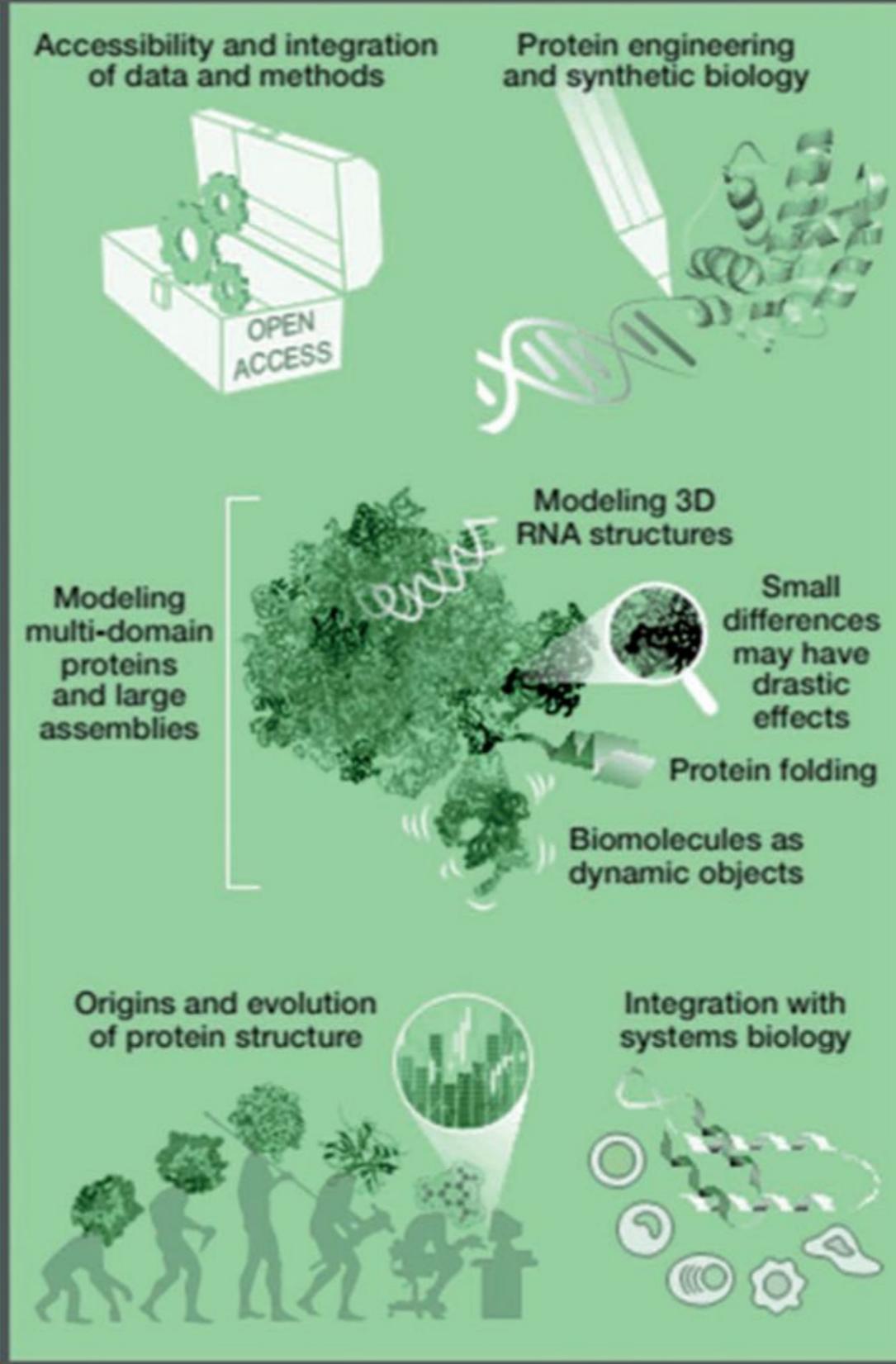
Coarse Grained

- 1 bead / 1 amino acid
- Connected by springs

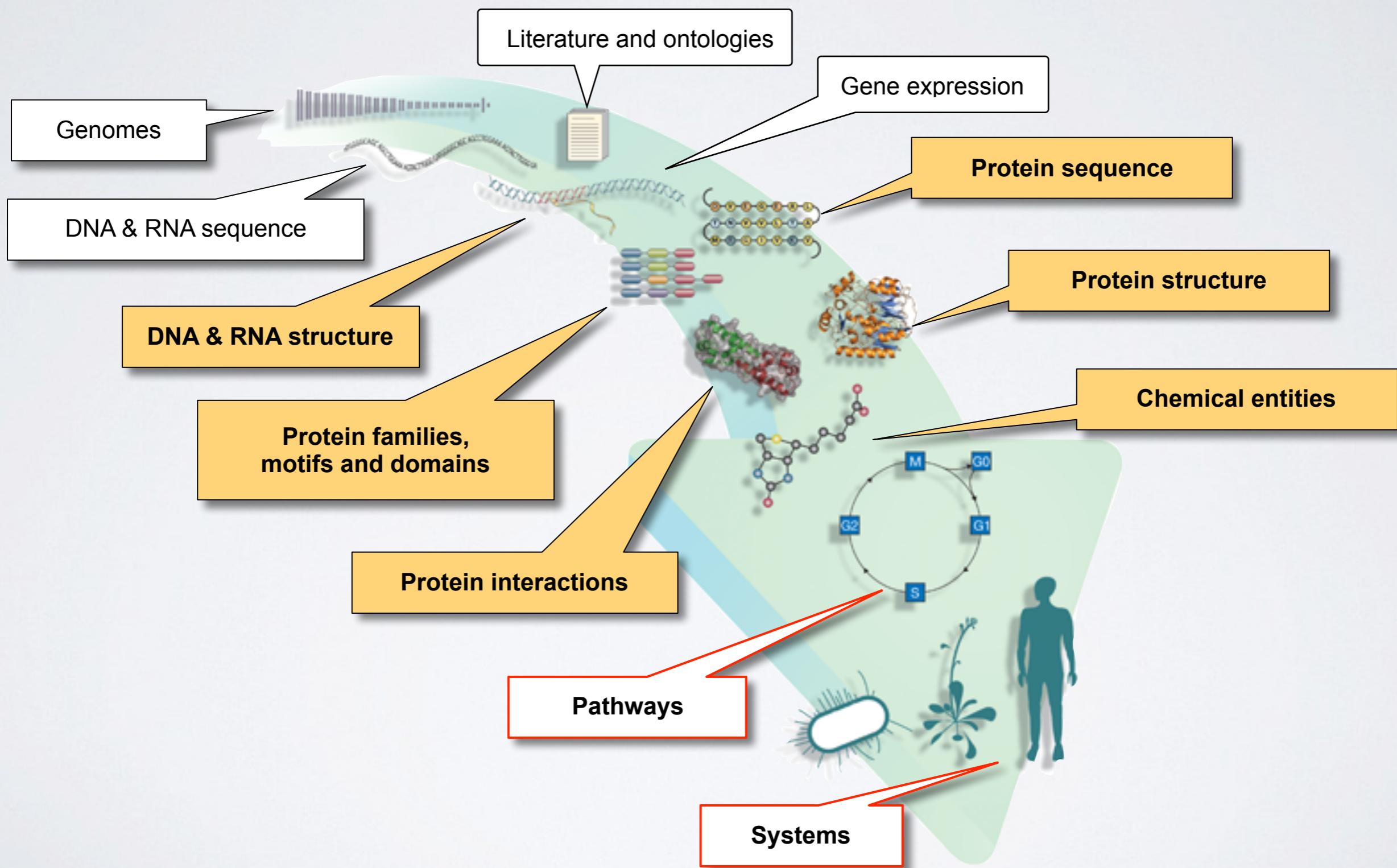
## ACHIEVEMENTS



## CHALLENGES



# INFORMING SYSTEMS BIOLOGY?



# CAUTIONARY NOTES

- A model is never perfect

A model that is not quantitatively accurate in every respect does not preclude one from establishing results relevant to our understanding of biomolecules as long as the biophysics of the model are properly understood and explored.

- Calibration of parameters is an ongoing imperfect process

Questions and hypotheses should always be designed such that they do not depend crucially on the precise numbers used for the various parameters.

- A computational model is rarely universally right or wrong

A model may be accurate in some regards, inaccurate in others. These subtleties can only be uncovered by comparing to all available experimental data.