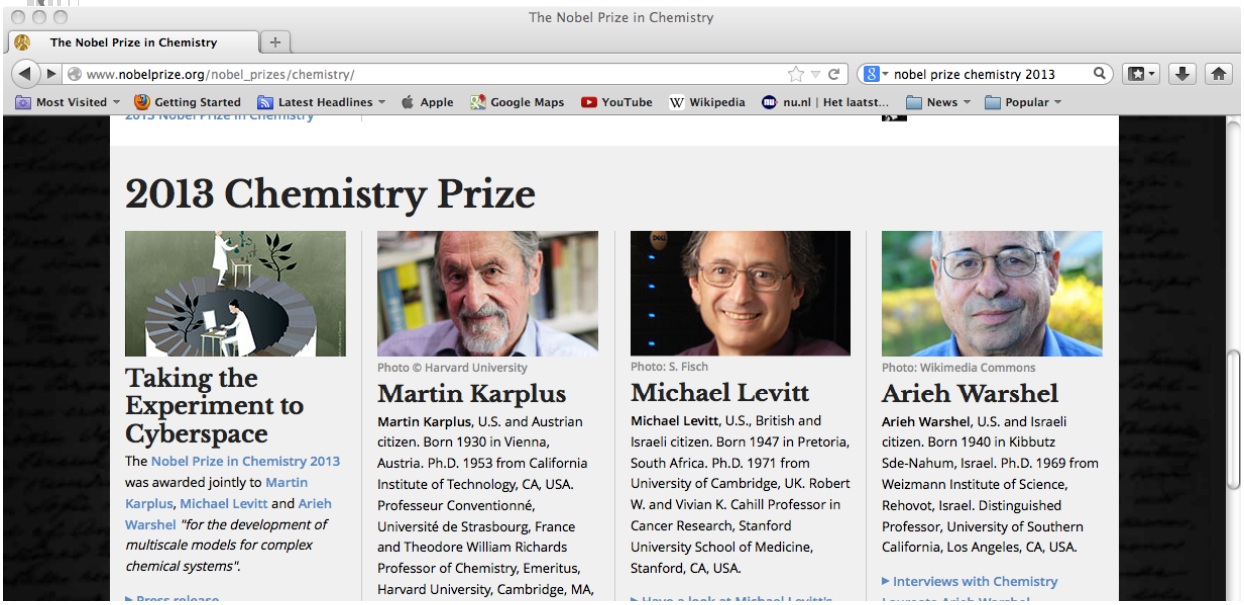
Molecular dynamics simulations

Outline

- Modeling and simulation of complex biomolecular systems
- Calculation of free energies



When is computational modeling useful ?

Simulation can replace or complement the experiment:

1. Experiment is impossible	*Inside of stars* *Weather forecast*
2. Experiment is too dangerous	*Flight simulation* *Explosion simulation*
3. Experiment is expensive	*High pressure simulation* *Windchannel simulation* *Trial and error drug design*
4. Experiment is blind	*Some properties cannot be observed on very short time-scales and very small space-scales*

Molecular simulation and experiment

	experiment	*simulation*
	 *(restricted)*	 *(unrestricted)*
Resolution*		
size :	10^{23} molecules	1 molecule
time :	1 second	10^{-15} seconds

*: Single molecules / 10^{-15} seconds possible
(but not both in the liquid phase)

Typical space / time scales

size :	10^{-3} meter	10^{-9} meter
time :	10^{3} seconds	10^{-6} seconds

Simulation and experiment are complementing methods to study different aspects of nature

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Visualize, rationalize, predict

- Inner life of the cell

A model for molecular computations

Three main questions to define a molecular model

1. What questions do we want to answer?
   - *Explain how a drug works*
   - *Design a new drug*
2. How much input do we have?
   - *Do we know of existing drugs?*
   - *Do we know the protein(s) involved?*
3. What are we able to do?
   - *What methodology is available?*
   - *What can we afford?*

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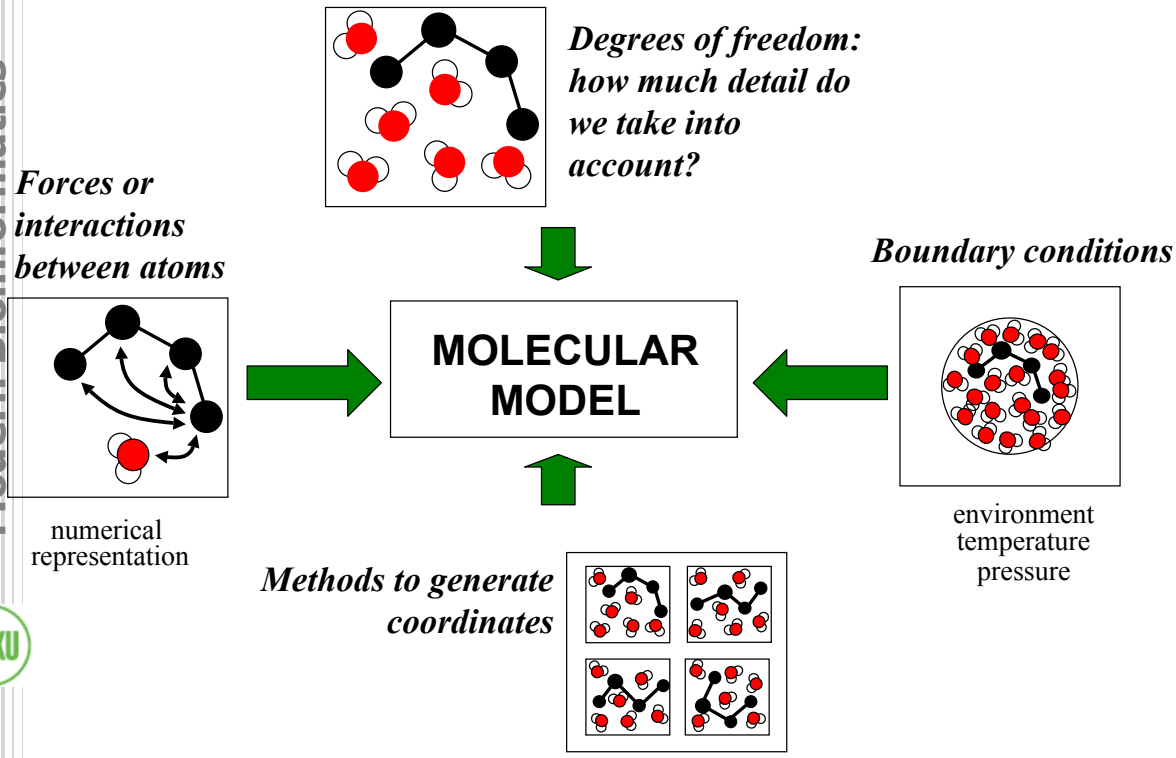
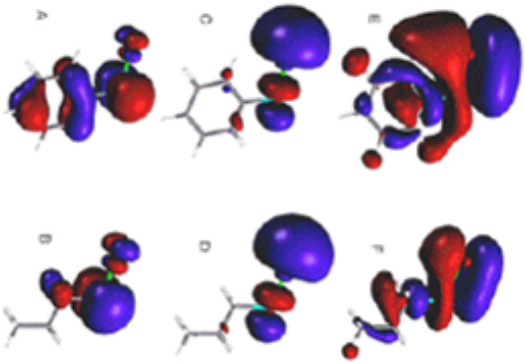
A model for molecular computations



A molecule has a certain energy

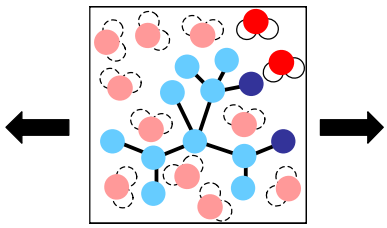
- Point charges with an electron cloud around it
  - Quantum mechanics, ab initio or semi-empirical



$$\hat{H}\psi(\mathbf{r}) = E\psi(\mathbf{r})$$

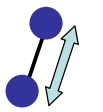
- Collection of balls and springs:
  - Molecular mechanics, force field representation

Molecular mechanical interactions

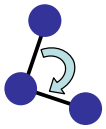


bonded interactions

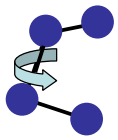
non-bonded interactions



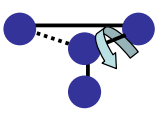
Bond stretching



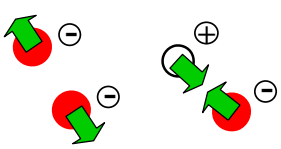
Angle bending



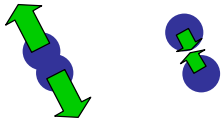
Rotation around bond



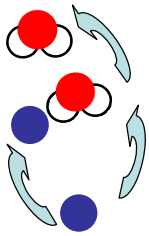
Planar atomgroups



Electrostatic interactions

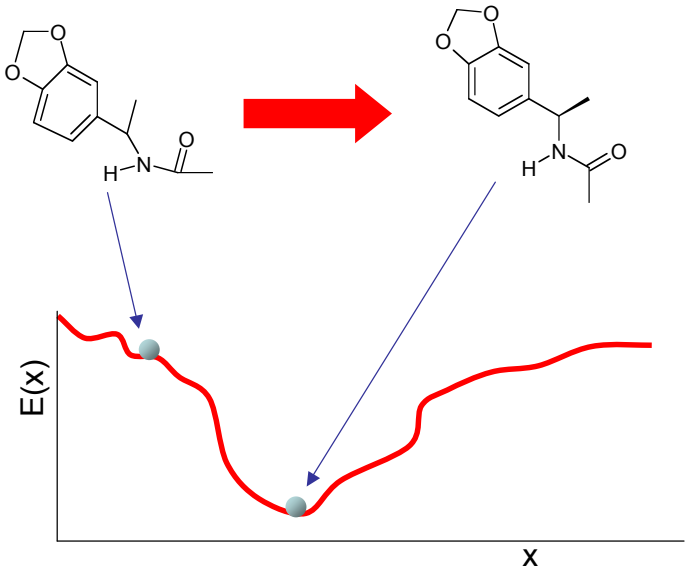


van der Waals interactions



Energy minimisation

- Find the lowest-energy conformation of a molecule



- Compare to a marble rolling down a slope

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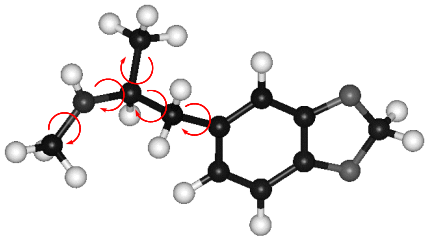
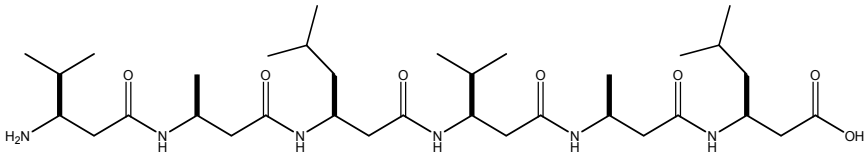
Different conformations

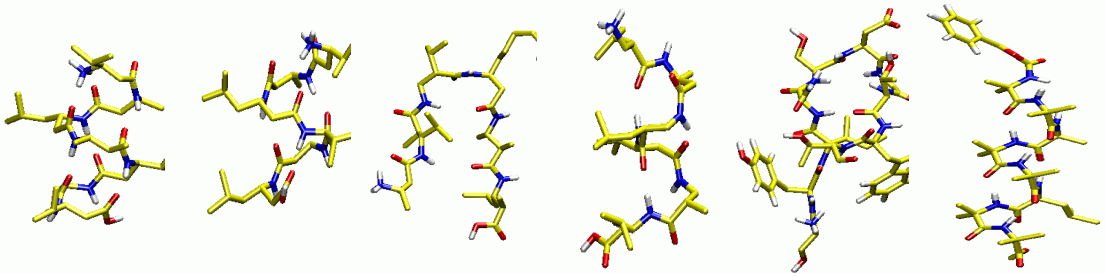
- Rotate around bonds



- One compound



- Many different conformations



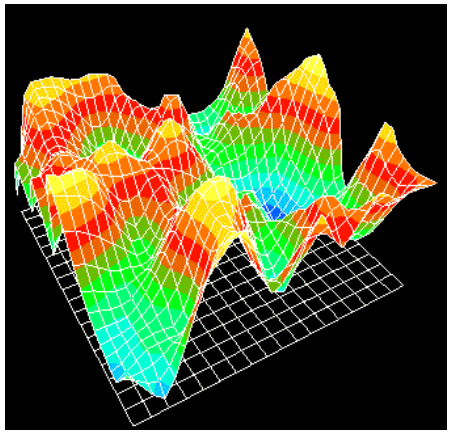
Different conformations

- Every conformation is associated with an energy, as a function of the positions of all particles, $\mathbf{q} = (x_1,y_1,z_1,x_2,y_2,z_2,\ldots)$

  $E = f(\mathbf{q}) = f(x_1,y_1,z_1,x_2,y_2,z_2,\ldots)$

- Compare **q** to a point on a multi-dimensional energy surface (3N-6)-dimensional

- Minima are favourable conformations
- Saddel points are transition states



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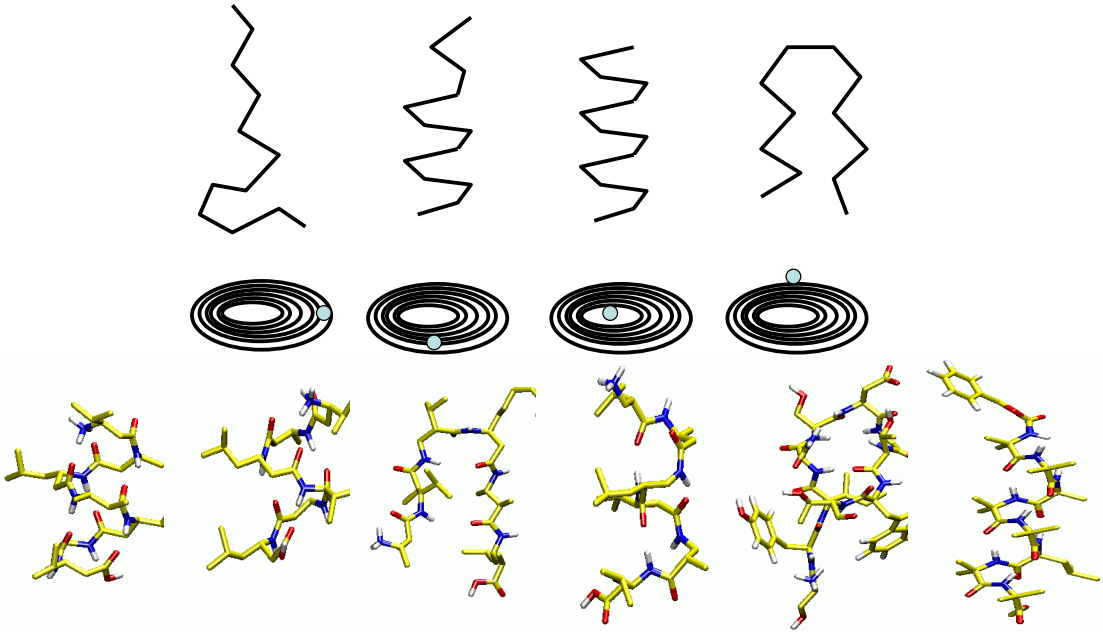
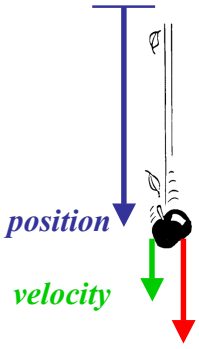
Conformations on the surface

- Every conformation is represented by a specific point on the 3N-6 dimensional surface



Classical laws of motion



Sir Isaac Newton
1642 -1727

Situation at time t

Force** is determined by relative **positions

$$\textit{acceleration} = \textit{force} / \textit{mass}$$

$$\Delta\textit{velocity} = \textit{acceleration} \times \Delta t$$

$$\Delta\textit{position} = \textit{velocity} \times \Delta t$$

Situation at time t+Δt

Determinism …

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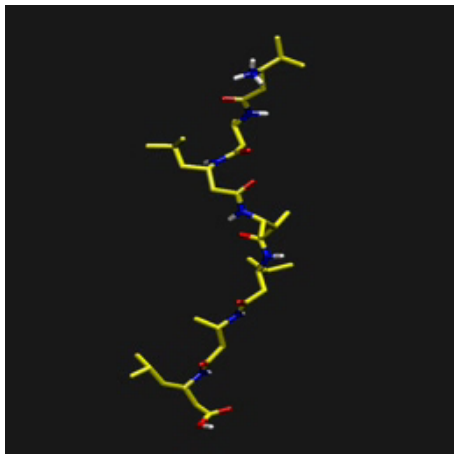
Folding simulation

- Proteins are too large systems to simulate the slow folding process.
- Smaller model compounds can be correctly folded on the computer.

⇨ Information about folding mechanisms and the unfolded state



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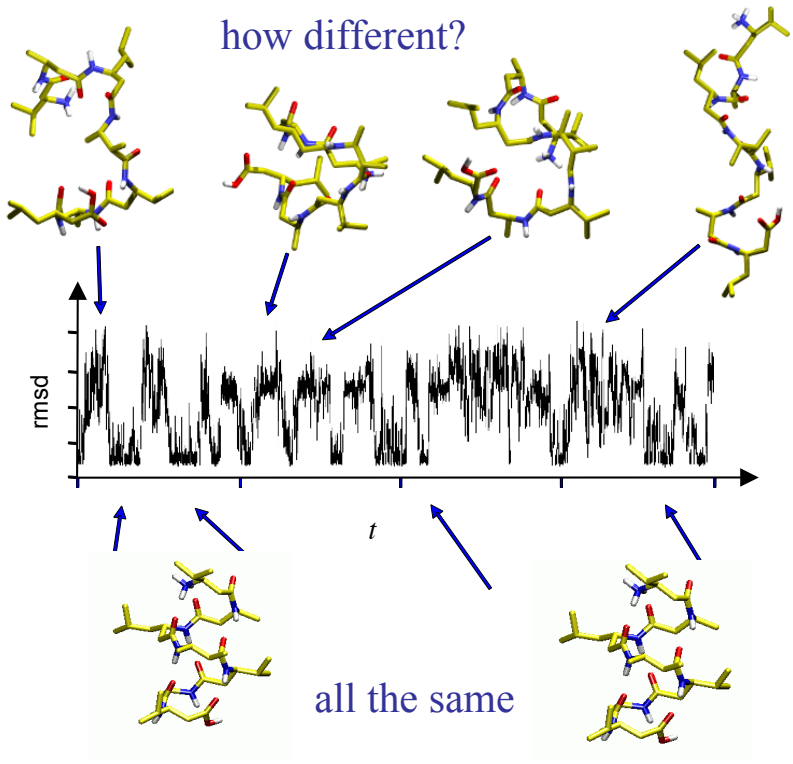
Conformational change over time

$3^{21} \approx 10^{10}$ possibilities!!

all different?
how different?

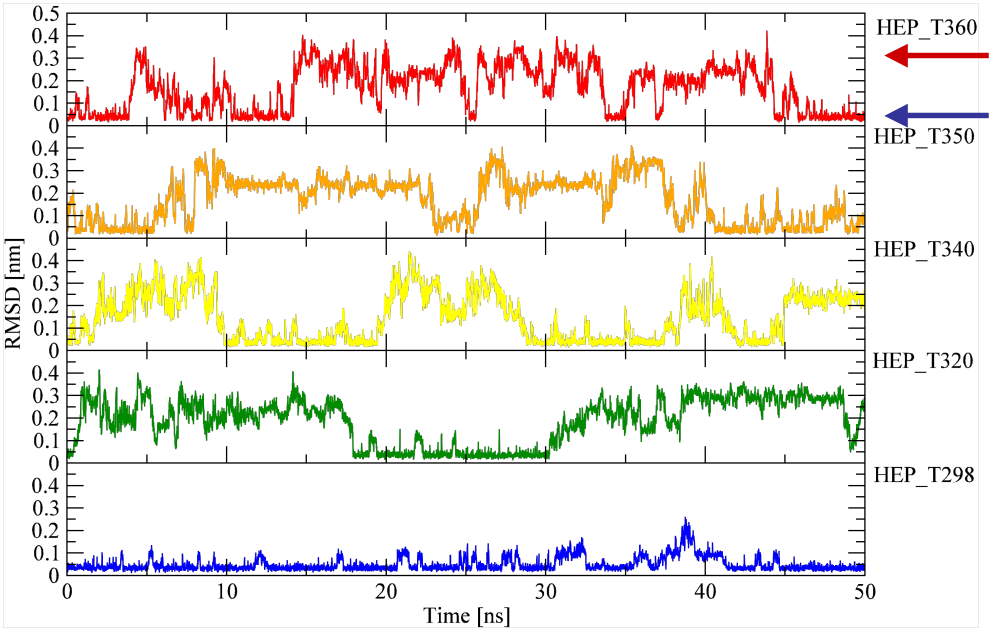


Unfolded structures

all the same

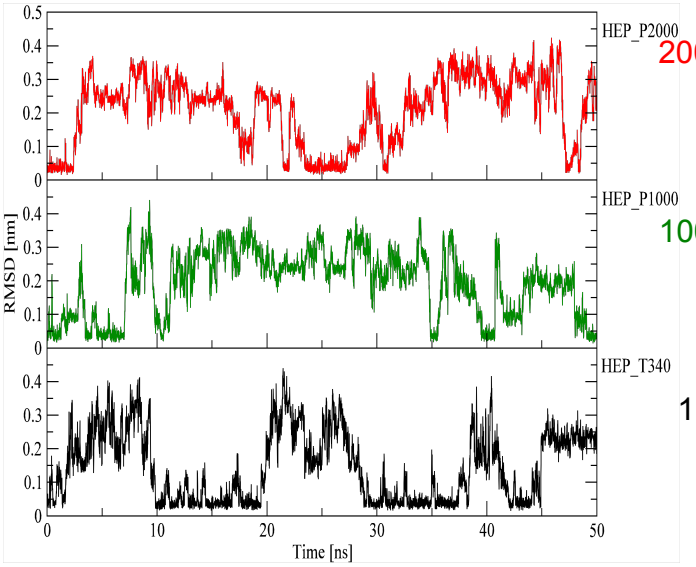
Folded structures

Temperature dependency



folding equilibrium depends on temperature

Pressure dependency



2000 atm

1000 atm

1 atm

folding equilibrium depends on pressure

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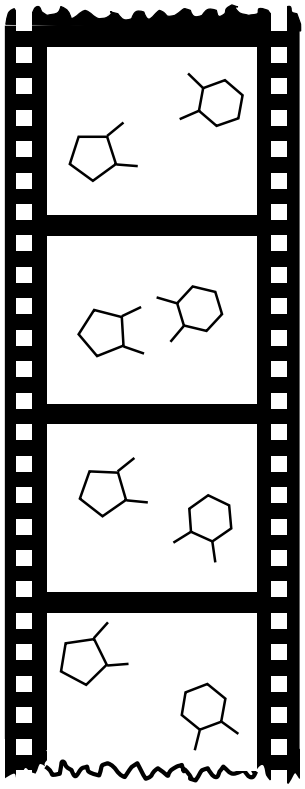
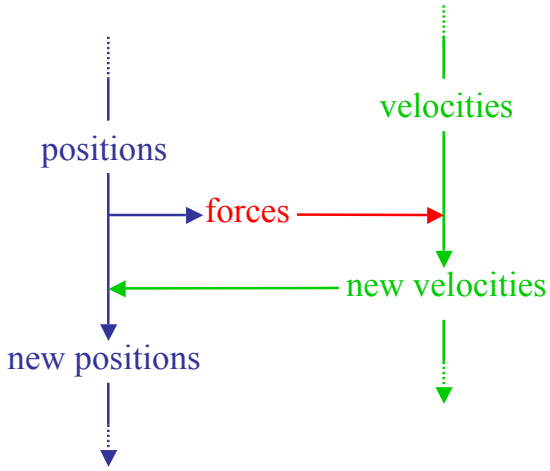
Molecular dynamics simulation



... comparable to shooting a movie of a molecular system...

Time t

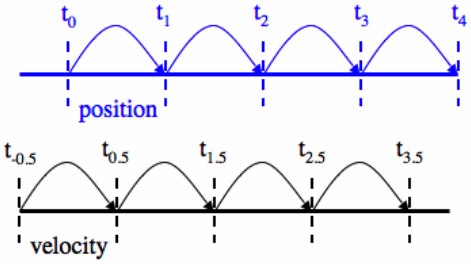
Time (t+Δt)

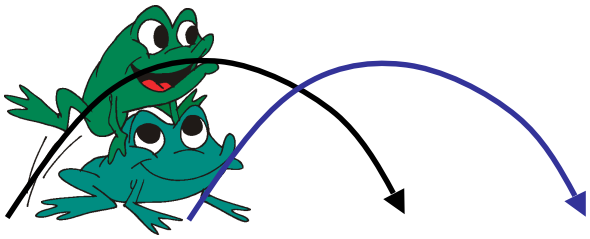


$\Delta t \approx 10^{-15}$ seconds

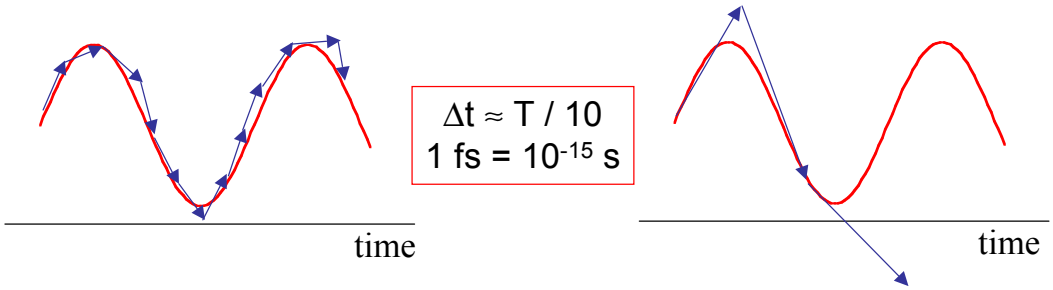
Leap-frog algorithm

- There are many integration algorithms
  - Verlet, Beeman en Leap-frog give identical coordinate trajectories





- The integration time step Δt should be sufficiently small, such that the fastest motion is correctly described



Molecular dynamics

1. Start at a certain conformation with initial velocities
2. Calculate the energy and the force on every atom i:

$$\vec{F}_i = -\vec{\nabla}_i E^{pot}(\vec{r}_1, \vec{r}_2, \ldots, \vec{r}_N)$$

3. From the force (acceleration) update velocity for every atom
4. From the velocity update the position
5. Propagate through time

- Total energy $E^{tot} = E^{pot} + E^{kin}$ is conserved (class. mech.)
- Kinetic energy allows us to go over barriers
- If we simulate infinitely long, we get the NVE ensemble

Forces from a force field

- The force on an atom is given by the derivative of the potential energy, with respect to its coordinates.

$$F_{x,1} = -\frac{\partial U(\mathbf{r})}{\partial x_1}$$

$$F_{y,1} = -\frac{\partial U(\mathbf{r})}{\partial y_1}$$

$$F_{z,1} = -\frac{\partial U(\mathbf{r})}{\partial z_1}$$

$$\mathbf{a}_1 = \frac{\partial^2 \mathbf{x}_1}{\partial t^2} = \frac{\mathbf{F}_1}{m_1}$$

U is given by a force field, with relatively simple functions.
The derivatives can be calculated analytically

For simple systems (harmonic oscillator) the equations of motion may be solved exactly: in all other cases we need to solve them numerically

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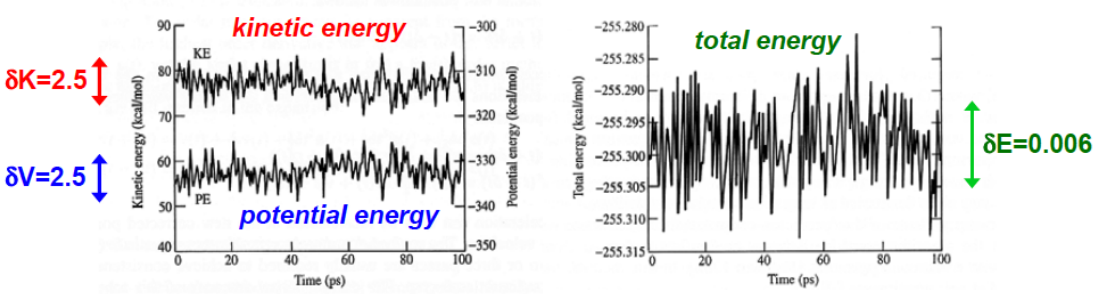
Energy conservation

- Simulation of liquid argon (256 atoms)



- The kinetic and potential energy fluctuates considerably
- The total energy is conserved
  - Remaining noise comes from the integration accuracy (Δt)

History

Year	molecular system: type, size	length of the simulation in seconds
1957	first molecular dynamics simulation (hard discs, two dimensions)	
1964	atomic liquid (argon)	10^{-11}
1971	molecular liquid (water)	$5 \cdot 10^{-12}$
1976	protein (no solvent)	$2 \cdot 10^{-11}$
1983	protein in water	$2 \cdot 10^{-11}$
1989	protein-DNA complex in water	10^{-10}
1997	polypeptide folding in solvent	10^{-7}
2001	micelle formation	10^{-7}
2010	folding of a small protein	10^{-6}

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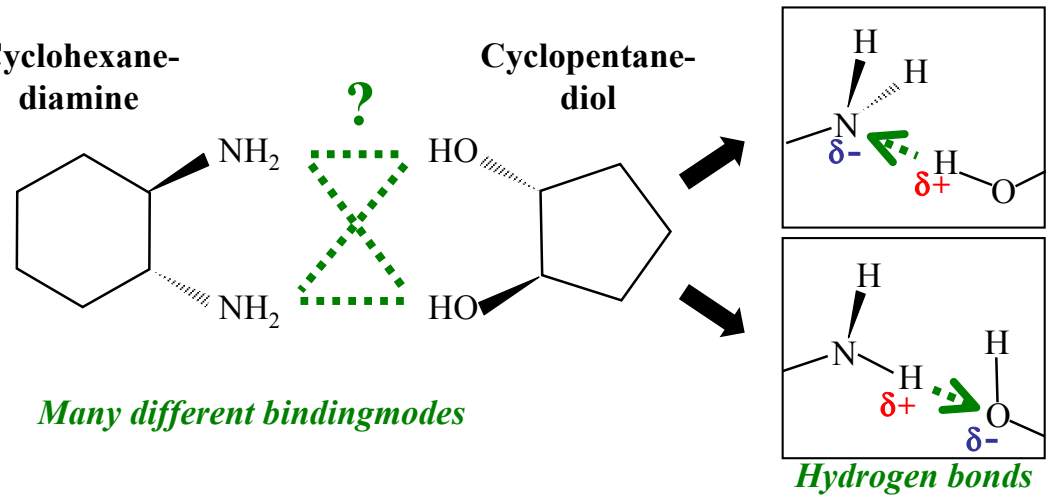
Binding equilibrium of two small molecules

Complex :

Cyclohexane-diamine

Cyclopentane-diol

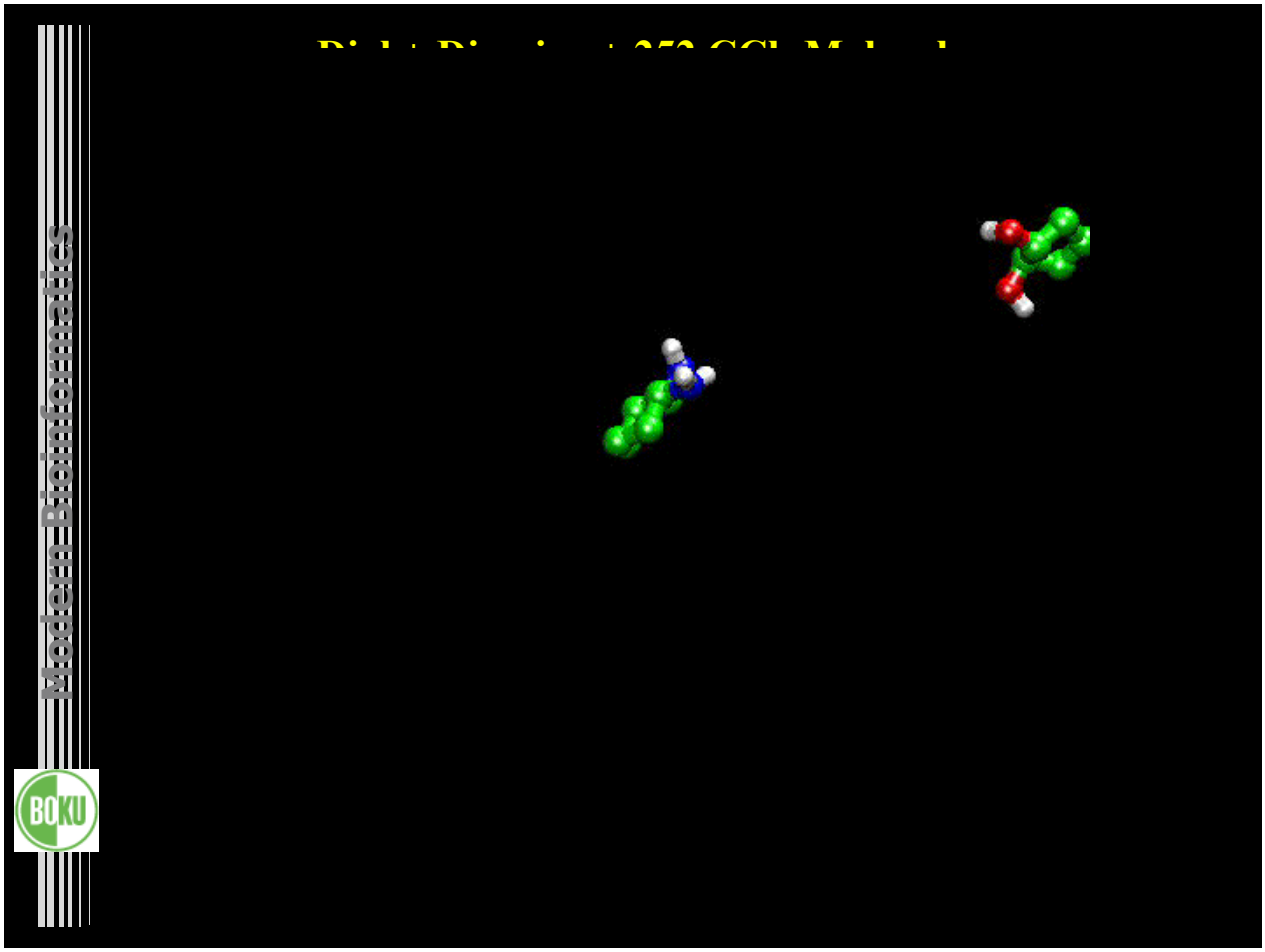


Many different bindingmodes

Hydrogen bonds

Average binding strength (free enthalpy) :

	Experimental		*MD simulation*
	Benzene	*CCl_4*	
ΔG_b [kJ/mol]	**-9.3**	**-11.5**	**-10.4**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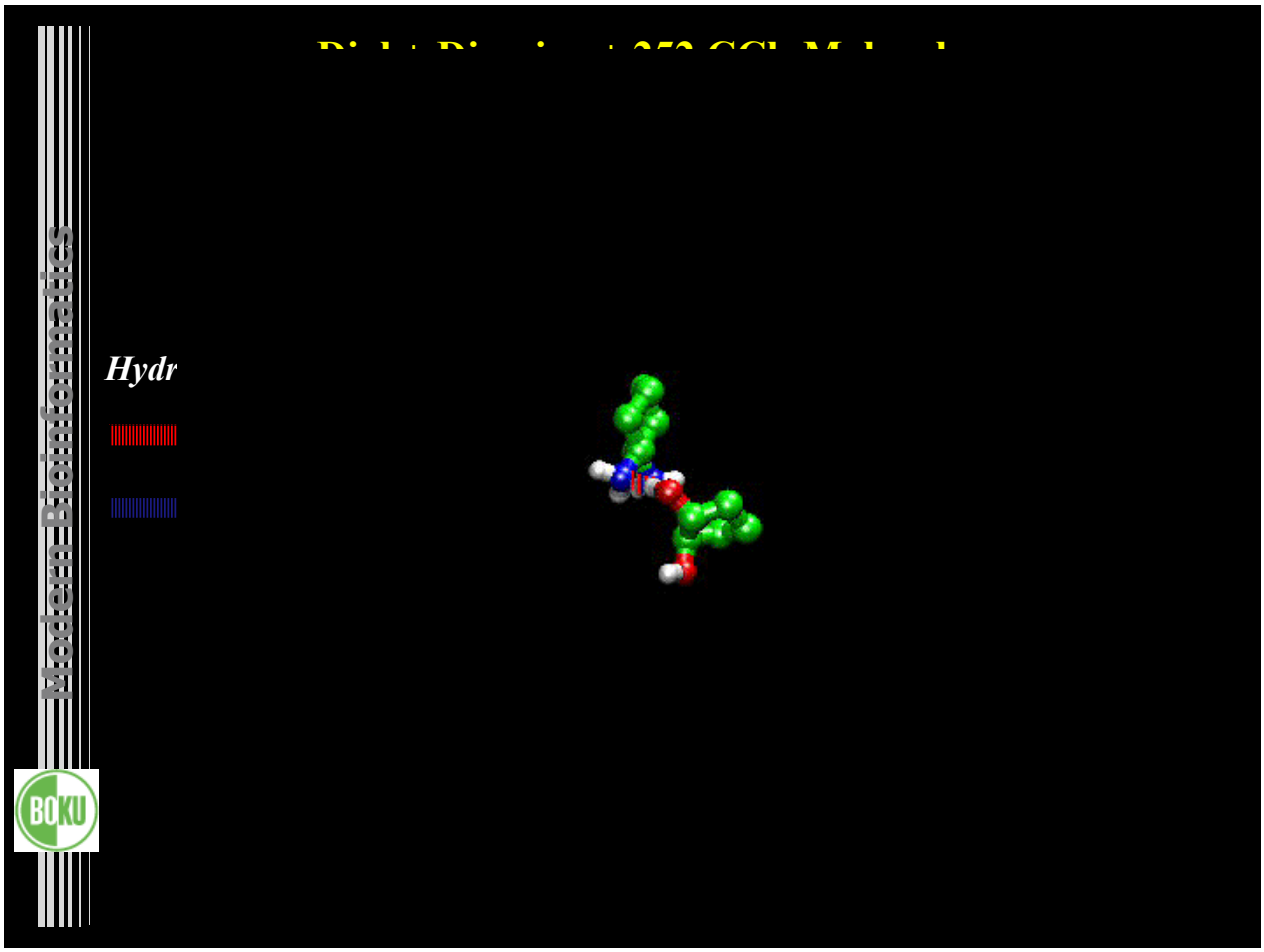
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Hydr
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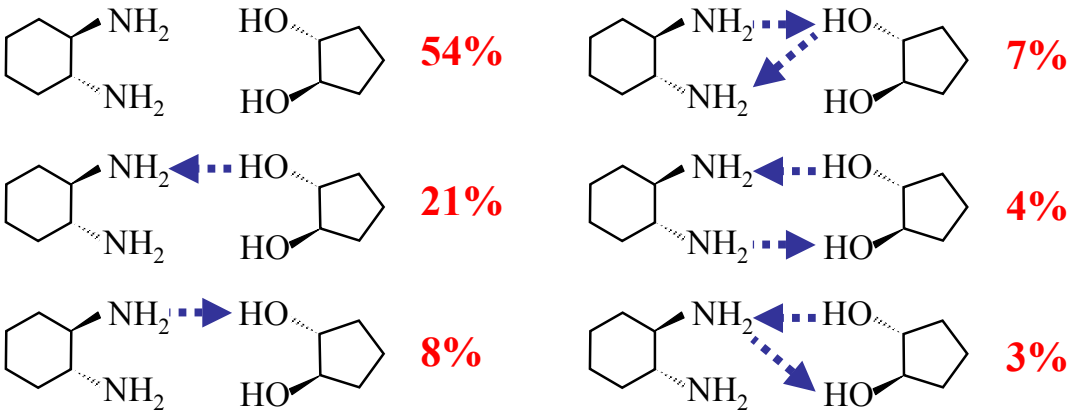
Results of the simulation

⇒ Experimentally hardly (or not) possible !

Occurrence of different binding modes :



Life time :

- Average life time of the complex: $2 \cdot 10^{-10}$ sec (max. $3 \cdot 10^{-9}$ sec)
- Average life time of a hydrogen bond: $5 \cdot 10^{-12}$ sec

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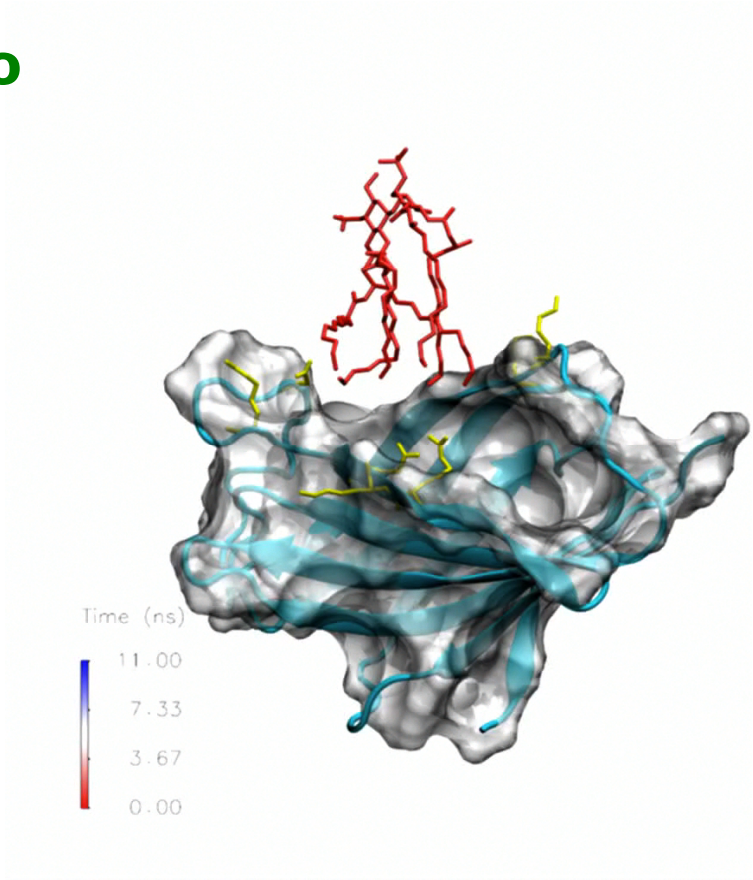
Lipid A binding to MD-2

- Complex molecules like proteins
- Dynamics becomes more difficult to describe
- Timescales are different



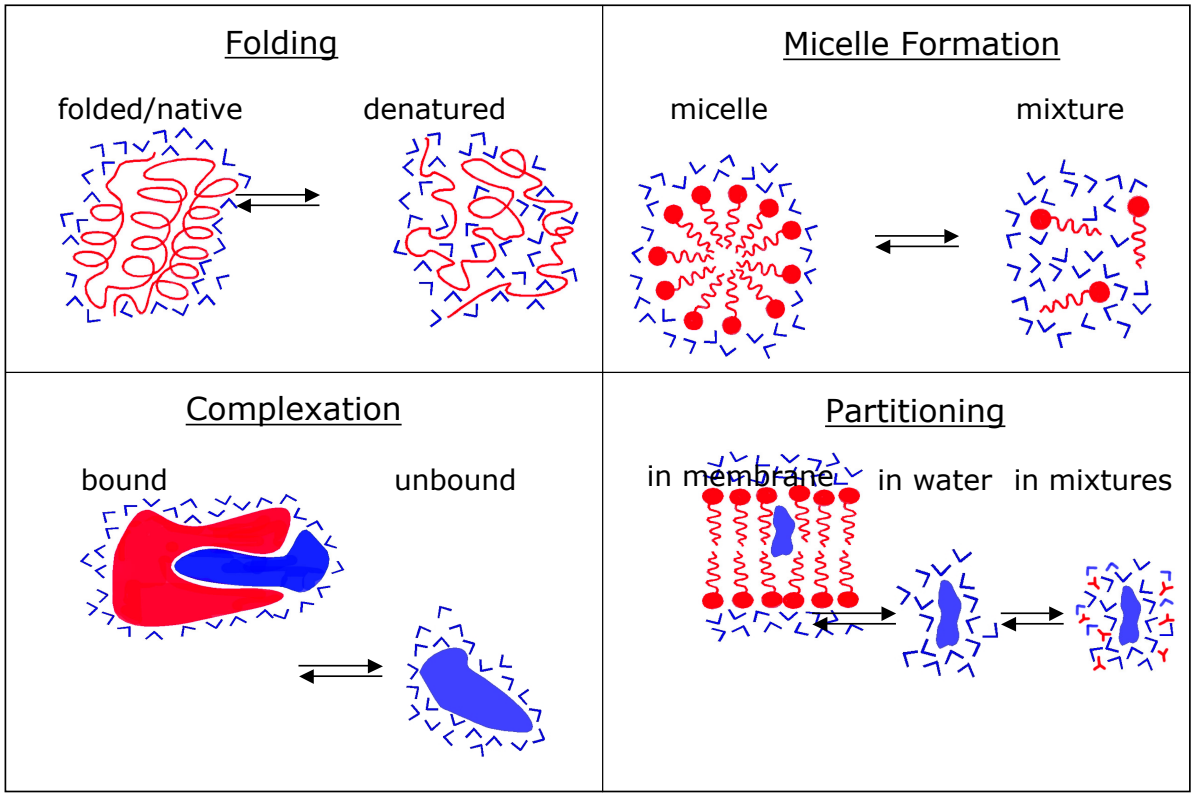
Molecular dynamics simulations

Inner life of the cell

Outline

- Modeling and simulation
- Calculation of free energies

Processes: Thermodynamic Equilibria



Definitions

free energy

The driving force for all physical processes

Free energy ΔA; Free enthalpy ΔG ($=\Delta A + p\Delta V$)

energy

The internal energy of the systems

Energy $\Delta E/\Delta U$; Enthalpy ΔH ($=\Delta E + p\Delta V$)

entropy

"The number of realization possibilities"

Entropy ΔS

Helmholtz / Gibbs equations

$\Delta A = \Delta E - T\Delta S$; $\Delta G = \Delta H - T\Delta S$

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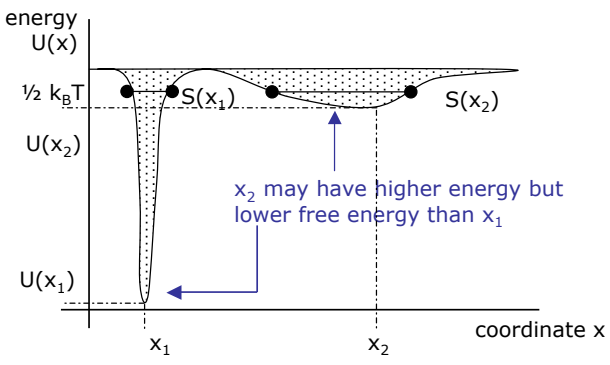
Statistical mechanics



mechanics: a state is characterised by ***one* minimum energy structure** (global minimum)

statistical mechanics: a state is characterised by ***an ensemble* of structures** or configurations or conformations

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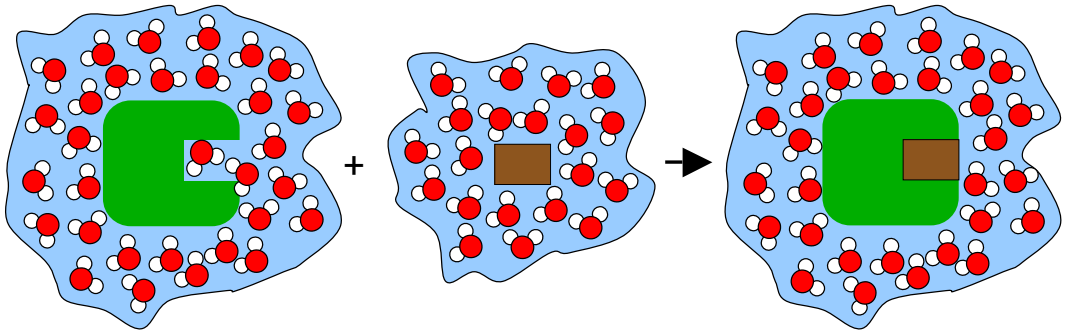
The system



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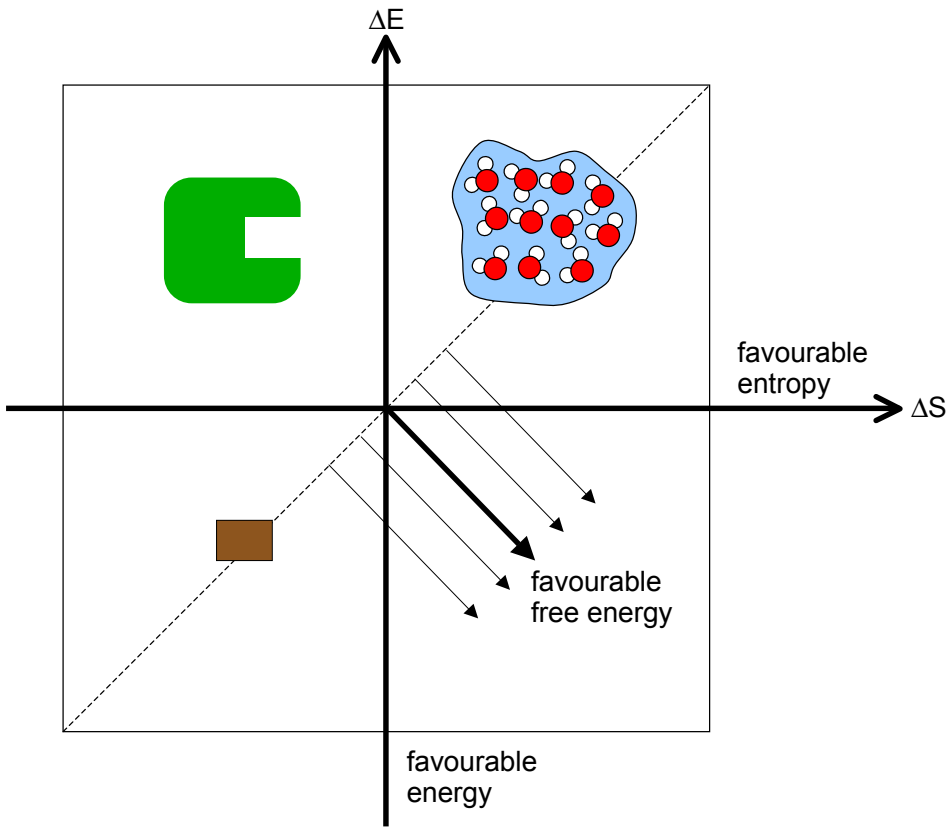

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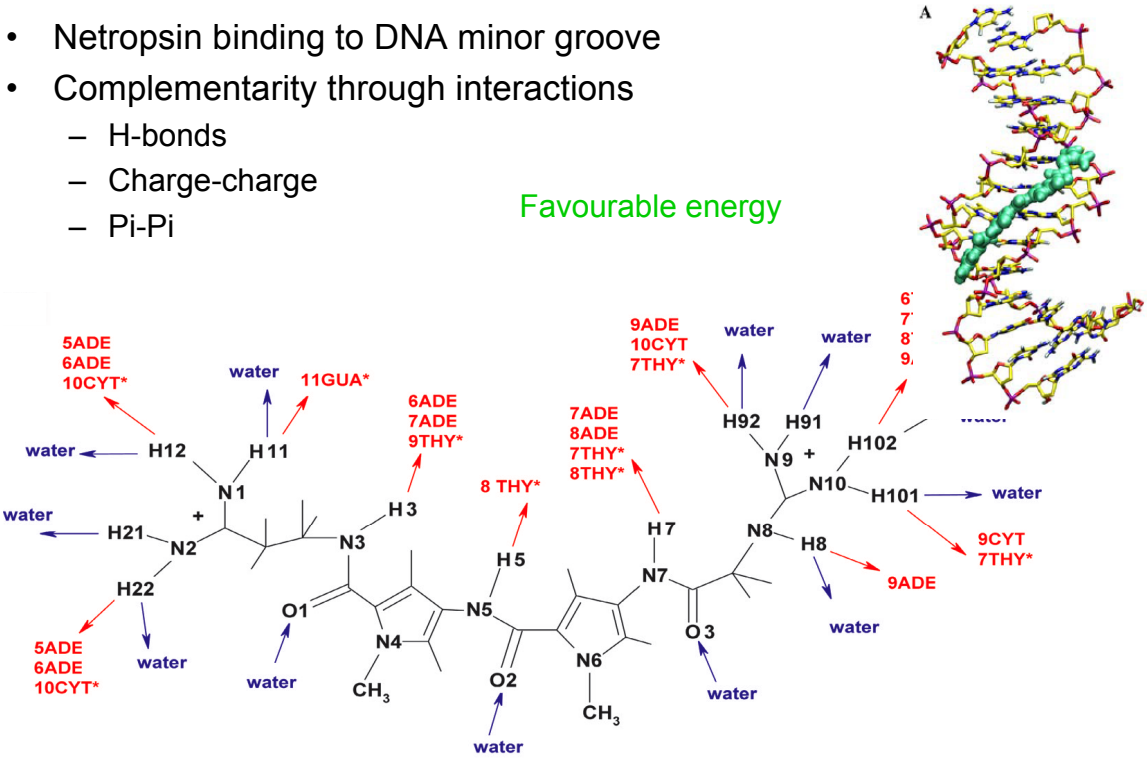
Guest-host complementarity

- Netropsin binding to DNA minor groove
- Complementarity through interactions
  - H-bonds
  - Charge-charge
  - Pi-Pi

Favourable energy



J. Dolenc, C. Oostenbrink, J. Koller, W.F. van Gunsteren, *Nucl. Acid Res.* (2005) 33:725

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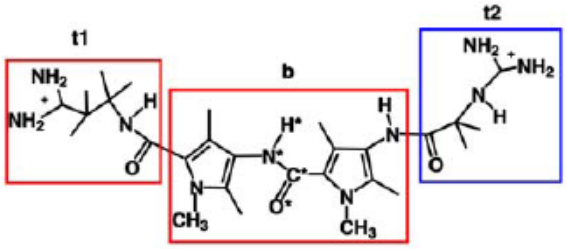
Entropy loss upon binding

- Conformational entropy calculated using Schlitter's formula
  - Netropsin and Distamycin A
  - In solution and when bound to DNA

	S_{free}	S_{bound}	ΔS_{bind}
Netropsin	862	735	-127
Distamycin	902	798	-104

in J/K/mol

Unfavourable entropy



Entropy loss mostly in the tails of the molecules

J. Dolenc, R. Baron, C. Oostenbrink, J. Koller and W.F. van Gunsteren, *Biophys J.* (2006) 91:1460

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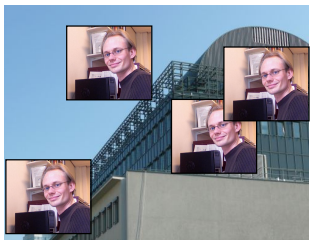
My affinity for BOKU

- Affinity is a combination of energy and entropy
  - Energy:
    Interactions with people at BOKU are mostly favourable
    (and I am even getting paid for it!)
  - Entropy:
    How much freedom do I have? At BOKU and elsewhere?



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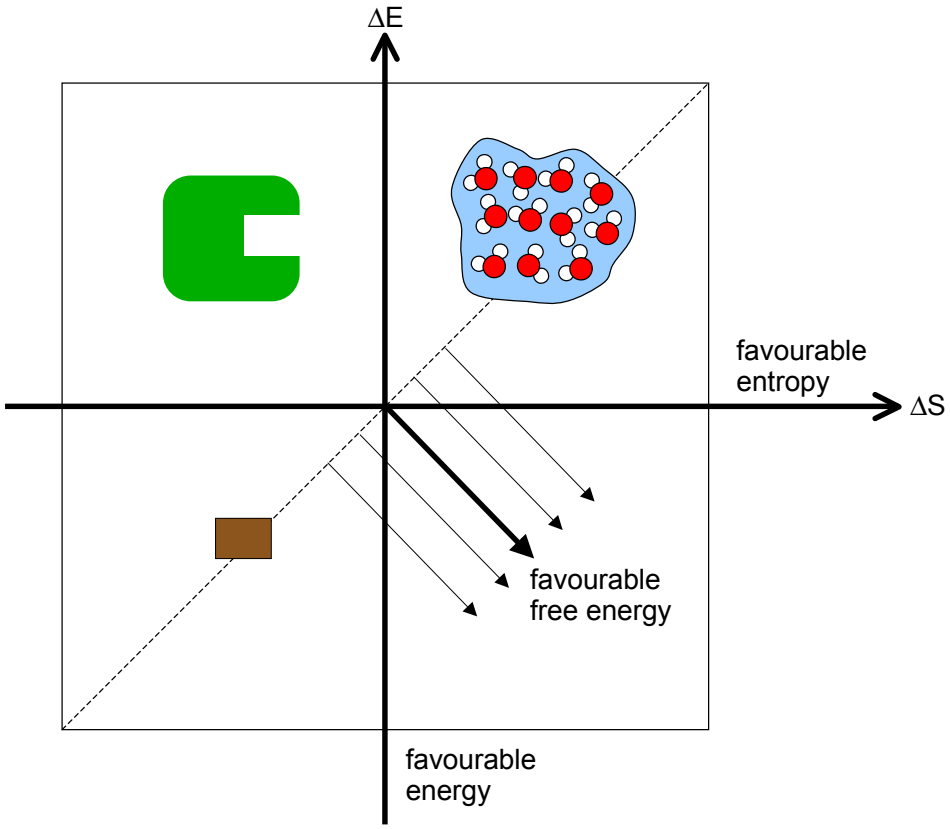
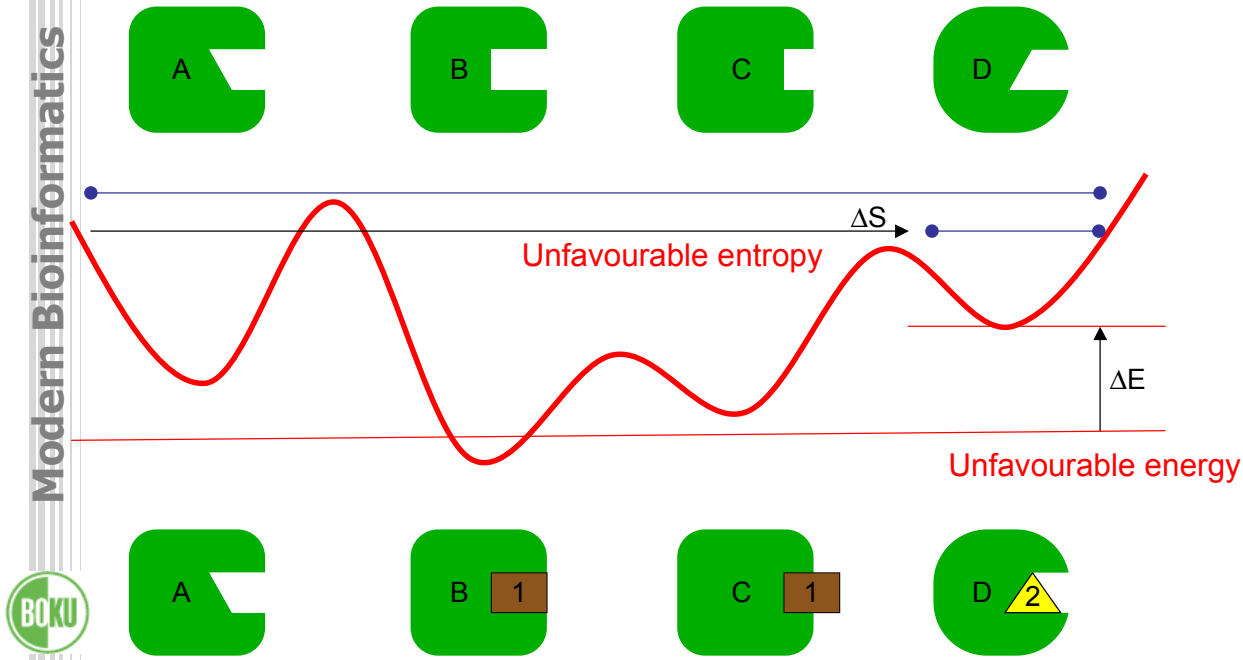


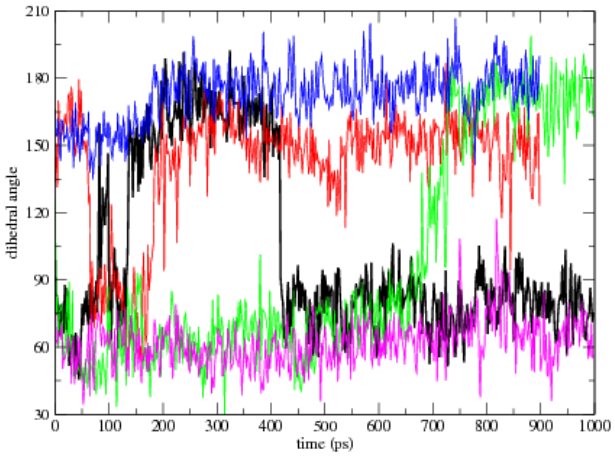
Conformational selection

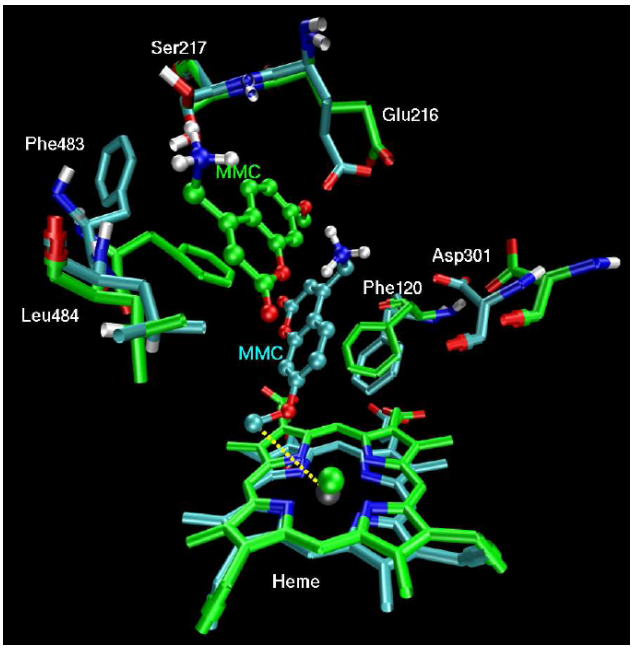


Binding to multiple structures

- Docking of 65 substrates in 2500 protein CYP2D6 structures
- Side-chain of Phe483 occupies multiple conformational states

- Different substrates have different preference





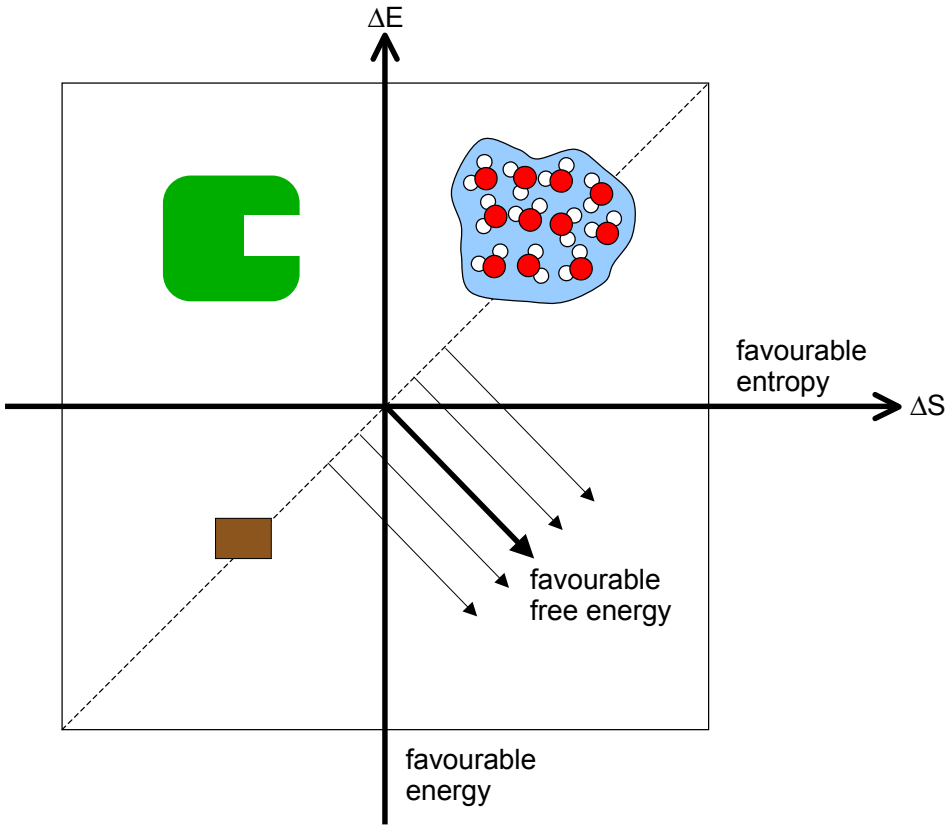
J. Med. Chem. 51 (2008) 7469 - 7477

The shape of the host

- Before we could come, people had to move and squeeze together
- Furniture was moved around

Unfavourable energy
Unfavourable entropy

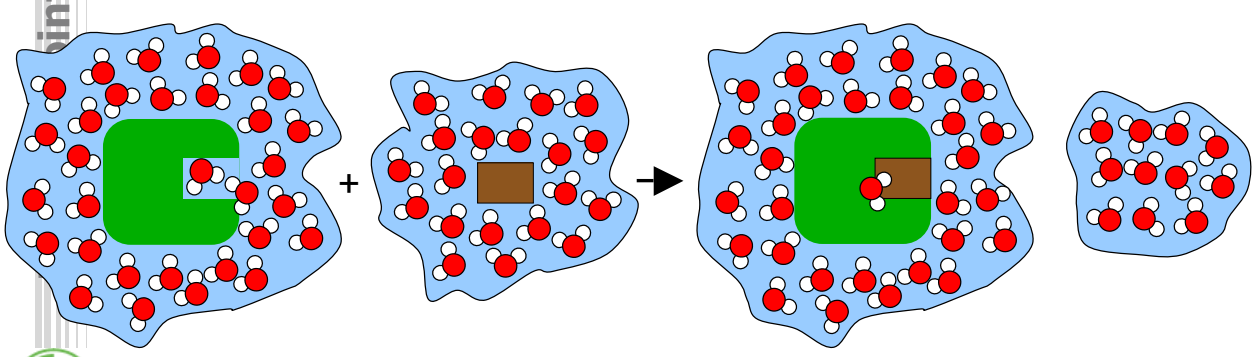
Modern Bioinformatics



Desolvation of protein and ligand

- Loss of solute-solvent hydrogen bonds
- Release of ordered water to bulk
  - Hydrophobic effect
- Structural water molecules in the active site

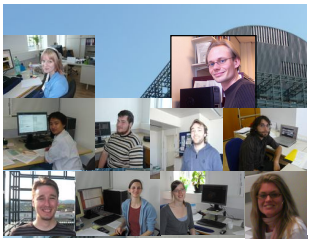
Unfavourable energy
Favourable entropy



Modern Bioinformatics

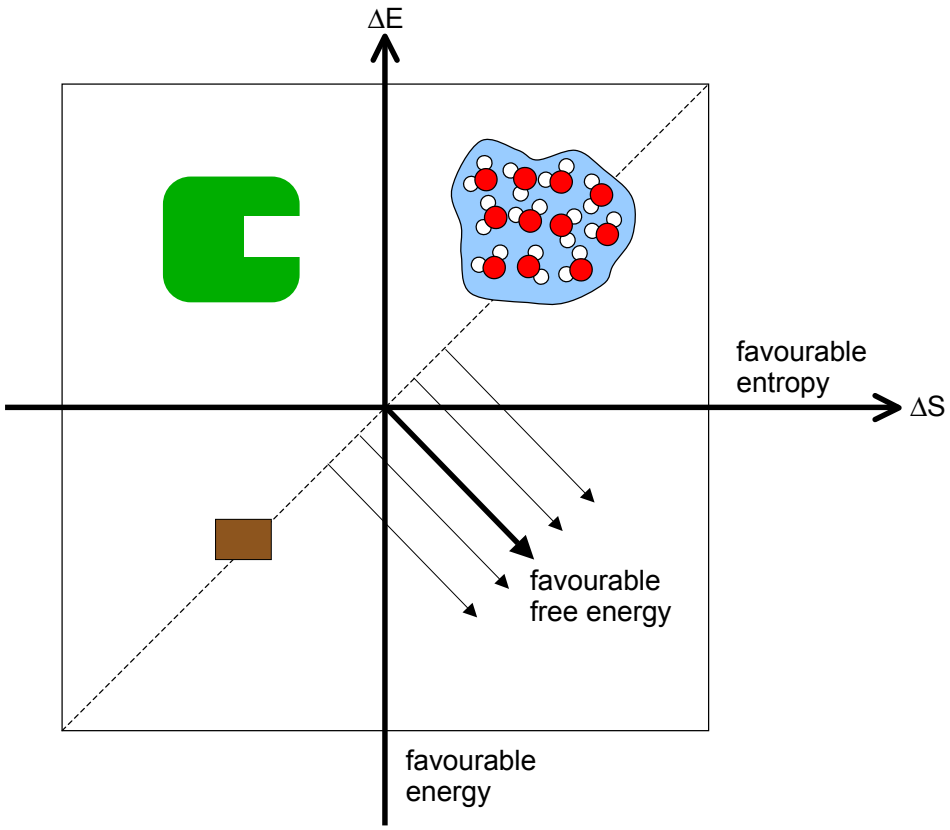
People and knowledge

students and co-workers…



strengthen the interaction

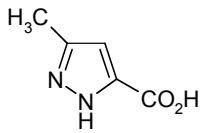
even if it hurts to say goodbye…
they have lot's of possibilities



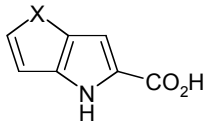
Modern Bioinformatics

Example: DAAO inhibitors

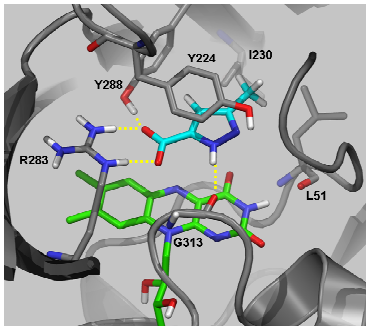
- Three inhibitors of the enzyme D-amino acid oxidase were studied

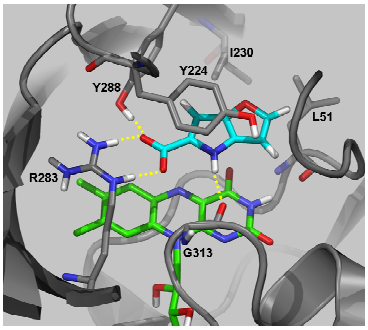


1



3 X = O
4 X = S





- Molecular dynamics simulations of the ligands in solution and bound to the protein, using GROMOS (parameter set 45A4)

Analysis of the simulations

Number of hydrogen bonds

	1	3	4
Free in solution	9.98	8.47	7.60
In complex:			
Tyr228 OH	1.00	1.00	1.00
Arg283 HE	1.03	0.99	1.00
Arg283 HH	1.28	1.35	1.34
Gly313	0.91	0.50	0.50
H_2O	2.15	1.87	0.61
Loss of H-bond	3.61	2.76	3.15

water is being released

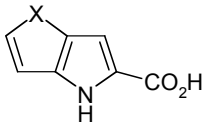
water still plays a role

Entropy (kJ/mol)

	calculated conformational entropy ligand	protein	full entropy experiment
$-T\Delta S$ (3)	15.3	17.0	-5.0
$-T\Delta S$ (4)	15.1	3.3	-15.9
$-T\Delta\Delta S$	0.2	13.7	10.9

the ligands are equally rigid

protein loss of entropy explains experiment?

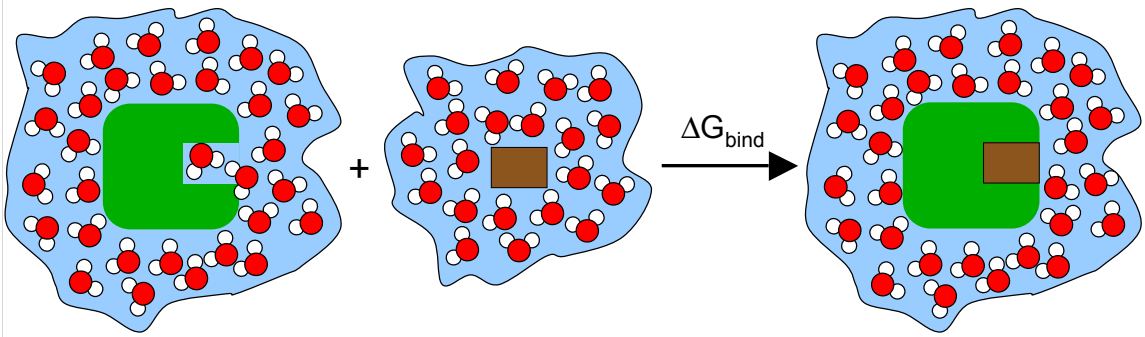


3 X = O
4 X = S

Modern Bioinformatics

Free energies of binding

- We hope to calculate the free energy of ligand binding
  - And need to consider all energetic and entropic contributions



$$\Delta G_{bind} = G_{complex} - G_{ligand} - G_{protein}$$

Definitions

free energy

$$A(N,V,T) = -k_B T \ln\left[N! h^{3N}\right]^{-1} \iint \exp\left(-H(\vec{p},\vec{r})/k_B T\right) d\vec{p}d\vec{r}$$
$$= U - TS$$

energy

$$U(N,V,T) = \left(\frac{\partial A/T}{\partial 1/T}\right)_{N,V} = \left\langle H(\ ,\ )\right\rangle_{\vec{p},\vec{r}}$$

entropy

$$S(N,V,T) = -\left(\frac{\partial A}{\partial T}\right)_{N,V} = \frac{U-A}{T}$$

Partition function

$$Z(N,V,T) = \left[N! h^{3N}\right]^{-1} \iint \exp\left(-H(\vec{p},\vec{r})/k_B T\right) d\vec{p}d\vec{r}$$

Free energy, energy and entropy are defined from statistical thermodynamics

Modern Bioinformatics

Statistical mechanics

- Equation to calculate free energy is simple:

$$G = -k_B T \ln Z_{NpT}$$

- Where Z_{NpT} is the partition function of the system

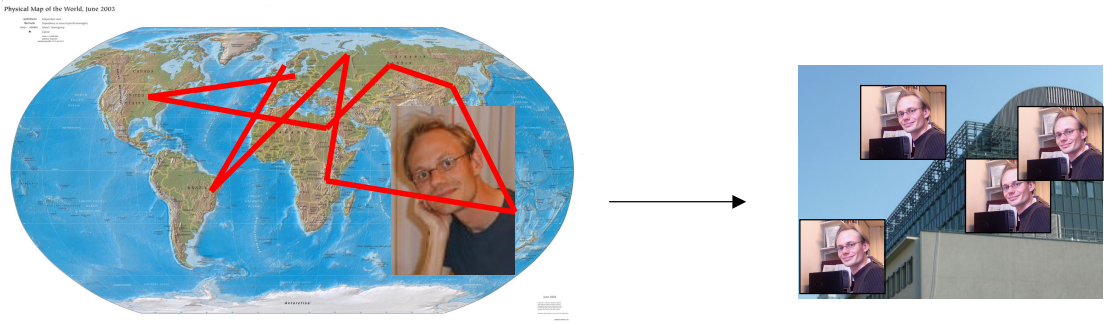
$$Z_{NpT} = \frac{1}{h^{3N} N!} \iiint e^{-(H(\mathbf{r},\mathbf{p})+pV)/k_B T}\, d\mathbf{p}\, d\mathbf{r}\, dV$$

Simulation samples over positions, momenta and volumes in physically relevant way

Integral over all possible positions, and all possible momenta of all particles in all different volumes

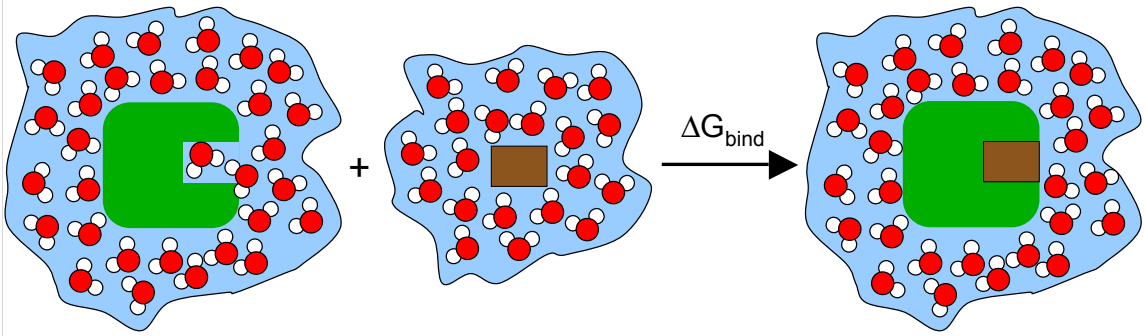
Sampling of all positions

- Sampling of all positions to calculate absolute free energy is practically impossible
- For molecules and for me



Do we really want that? (I)

- We only really care about the difference, ΔG



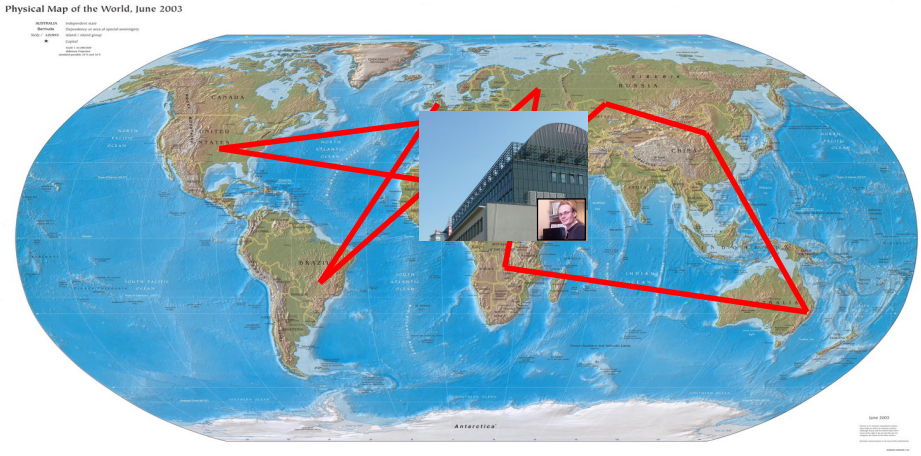
- The perturbation formula calculates free energy differences directly

$$\Delta G = -k_B T \ln \left\langle e^{-(H_{complex} - H_{ligand} - H_{protein})/k_B T} \right\rangle$$

Works if the positions, momenta and volumes of both sides are not too different

Directly consider the difference

- Consider my presence at BOKU as one of many possibilities



- Is still very difficult and unlikely to work

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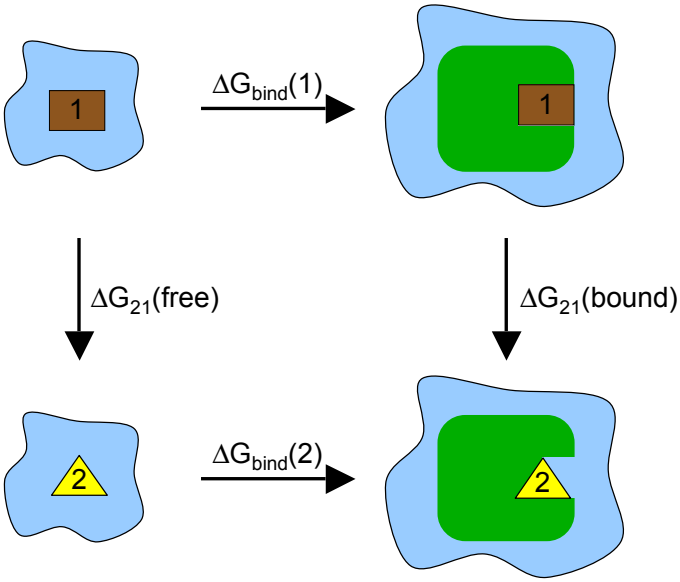
Do we really want that? (II)

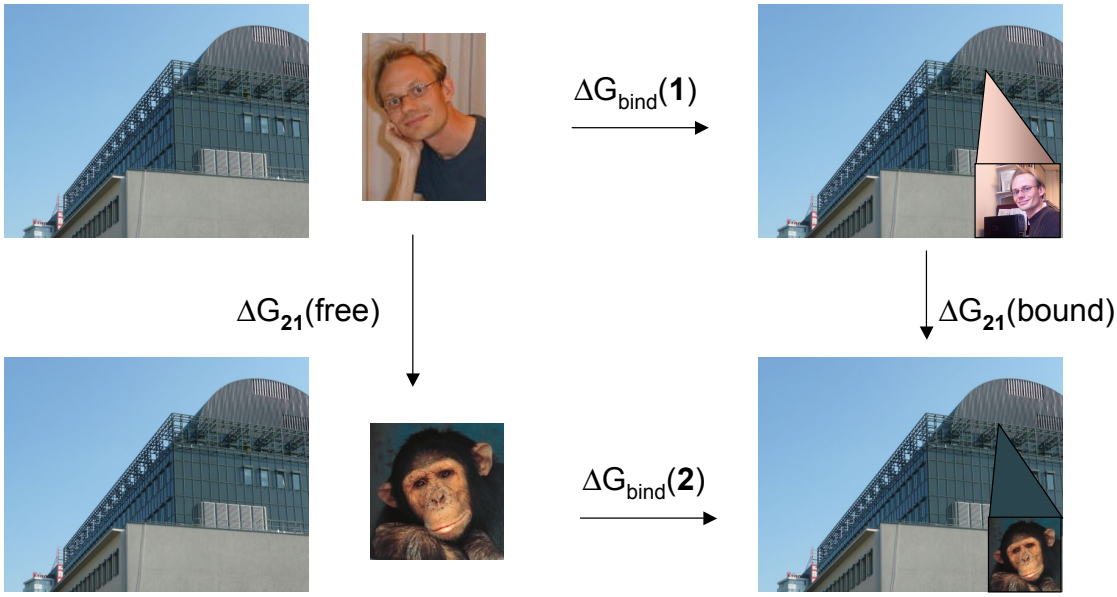
- Free energy is independent of the path (state function)
- Thermodynamic cycle
- Relative free energies
- Computational alchemy



$\Delta\Delta G_{bind} = \Delta G_{bind}(2) - \Delta G_{bind}(1)$
$= \Delta G_{21}(bound) - \Delta G_{21}(free)$

Who fits better at BOKU?

- Are there others that are more suitable?



- Compare two employees when they are free and at BOKU

Free energy difference

Free energy perturbation

$$\Delta A_{BA} = A_B - A_A = -k_B T \ln \frac{Z_B(N,V,T)}{Z_A(N,V,T)}$$

$$= -k_B T \ln \frac{\iint e^{-H_B(\mathbf{r},\mathbf{p})/k_B T} d\mathbf{p}d\mathbf{r}}{\iint e^{-H_A(\mathbf{r},\mathbf{p})/k_B T} d\mathbf{p}d\mathbf{r}}$$

$$= -k_B T \ln \frac{\iint e^{-(H_B(\mathbf{r},\mathbf{p})-H_A(\mathbf{r},\mathbf{p}))/k_B T} e^{-H_A(\mathbf{r},\mathbf{p})/k_B T} d\mathbf{p}d\mathbf{r}}{\iint e^{-H_A(\mathbf{r},\mathbf{p})/k_B T} d\mathbf{p}d\mathbf{r}}$$

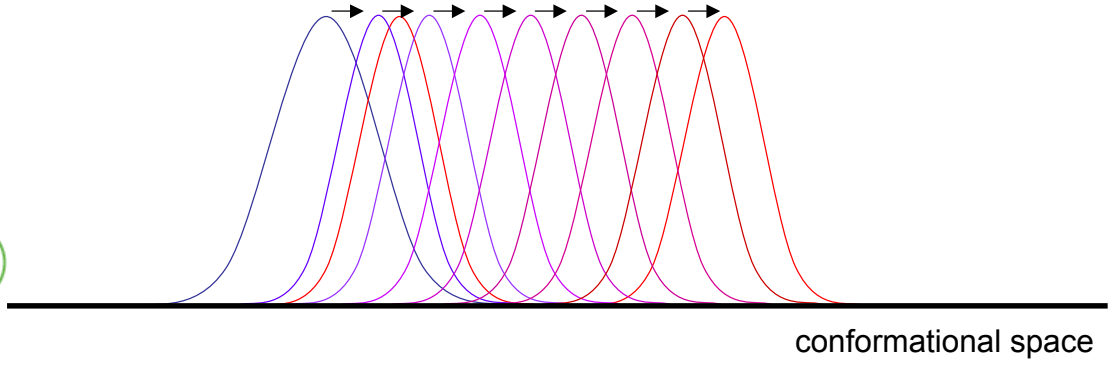
$$= -k_B T \ln \left\langle e^{-(H_B - H_A)/k_B T} \right\rangle_A$$

Zwanzig, J. Chem. Phys (1954) 22:1420

Free energy calculation

$$\Delta G_{AB} = -k_B T \ln \left\langle e^{-(E_A - E_B)/k_B T} \right\rangle_B$$

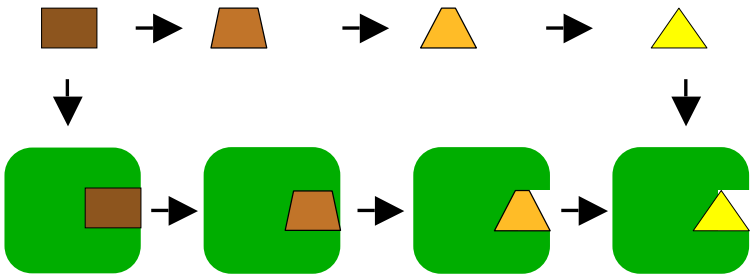
$$\Delta G_{AB} = \sum_{\lambda} \Delta G_{\lambda,\lambda+\delta\lambda}$$



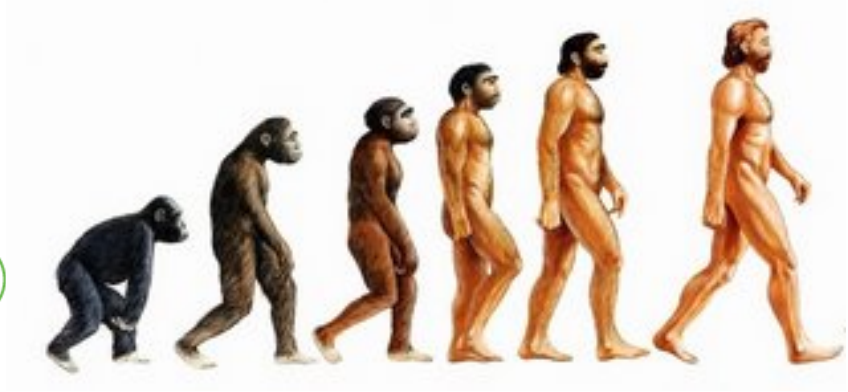
conformational space

Modern Bioinformatics

Gradually change one in the other



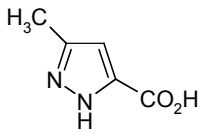
Change ligand 1 into ligand 2, in solution and when bound to the protein



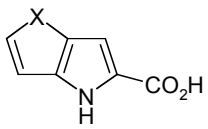
As long as the end-states are defined, the intermediates do not have to be physically possible

Example: DAAO inhibitors

- Three inhibitors of the enzyme D-amino acid oxidase were studied



1



3 X = O
4 X = S

	3->1	**3->4**	**4->1**
Calculated values:			
ΔG_{free}	106.3 ±1.5	86.1 ±0.8	20.4 ±1.1
$\Delta G_{complex}$	113.8 ± 2.2	87.3 ±3.5	36.7 ±2.0
$\Delta\Delta G_{bind}$	7.5 ± 3.7	1.2 ± 4.3	16.3 ± 3.1
Experimental $\Delta\Delta G_{bind}$ *based on:*			
IC_{50}[a]	8.2	-0.9	9.1
IC_{50}[b]	4.6	0.1	4.6
ITC	9.4	0.8	8.6
SPR[c]	14.1	1.6	12.4

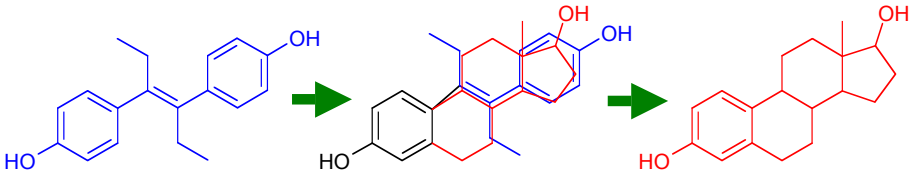
Overall, the relative binding free energies are very well reproduced

J.H.M. Lange, J. Venhorst, M.J.P. van Dongen, J. Frankena, F. Bassissi, N.M.W.J. de Bruin, C. den Besten, S.B.A. de Beer, C. Oostenbrink, N. Markova and C.G. Kruse, *Eur. J. Med. Chem.* (2011) **46**, 4808 - 4819

Modern Bioinformatics

Computational alchemy

- Modify one compound into another one in small steps



$$E(\mathbf{q},\mathbf{p},\lambda) = (1-\lambda)E_A(\mathbf{q},\mathbf{p}) + \lambda E_B(\mathbf{q},\mathbf{p})$$

- In a formula:

$$\lambda = 0 \Rightarrow E = E_A \qquad \lambda = 1 \Rightarrow E = E_B$$

Along the way? The protein ‘sees’ a mixture of A and B

$$\Delta G_{AB} = \sum_{\lambda=0}^{1} -k_B T \ln \left\langle e^{-\Delta E(\lambda \to \lambda + d\lambda)/k_B T} \right\rangle$$

Example: ER

- Relative free energy of three compounds
- In three different media (vacuum, solution, protein)
- In 11 discrete steps, forward and backward TI

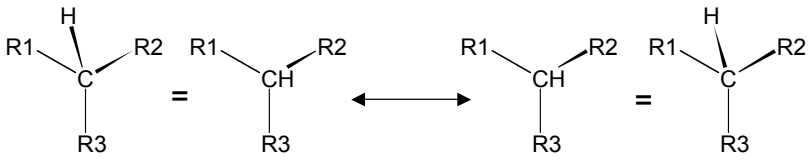
Table 4. TI Results (kJ mol^{-1})[a]

	DES ↔ E2			DES ↔ GEN		
TI	for-ward	back-ward	hysteresis	for-ward	back-ward	hysteresis
vacuum	76.3	76.1	0.2	187.1	186.9	0.2
solvent	79.0	81.6	−2.6	151.5	157.3	−5.8
protein	80.4	78.2	2.2	173.1	165.3	7.8
$\Delta\Delta G_{solv}$	2.8	5.5	−2.7	−35.6	−29.5	−6.0
$\Delta\Delta G_{bind}$	1.4	−3.4	4.8	21.6	8.0	13.6
$\Delta\Delta G_{bind}$	3.8[b]			11.3[b]		
(expt)	0.79[c]			21.69[c]		

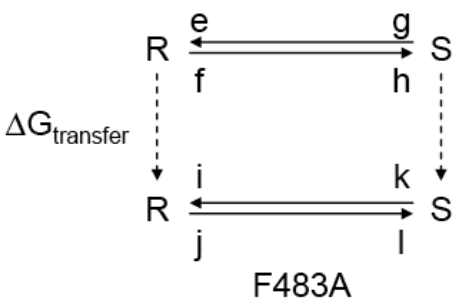
Stereospecific propranolol binding

- R- and S-Propranolol have similar affinity for CYP450 2D6
- 20 fold decrease of affinity of R-Propranolol to F483A mutant

- Free energy calculation to convert R-propranolol into S-propranolol



WT



F483A

$\Delta\Delta G_{bind}$ = ΔG_{bind}(R) - ΔG_{bind}(S)
= -7.7 kJ/mol (experimental)

= ΔG_{inv}(F483A) - ΔG_{inv}(WT)
= -7.5 kJ/mol (simulation)

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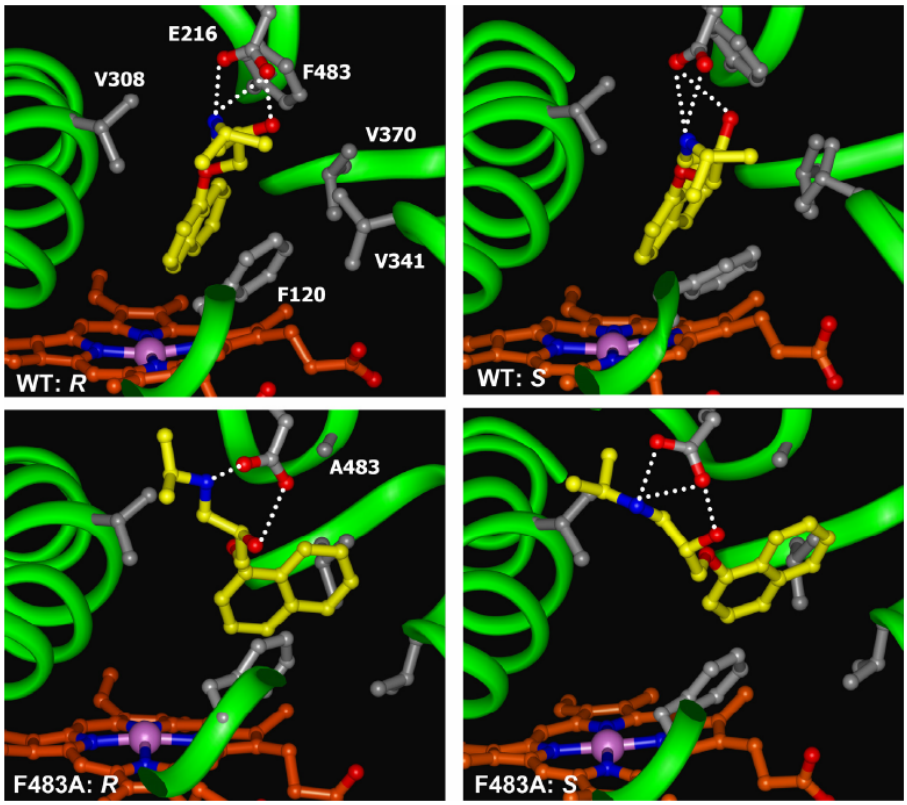
Molecular picture

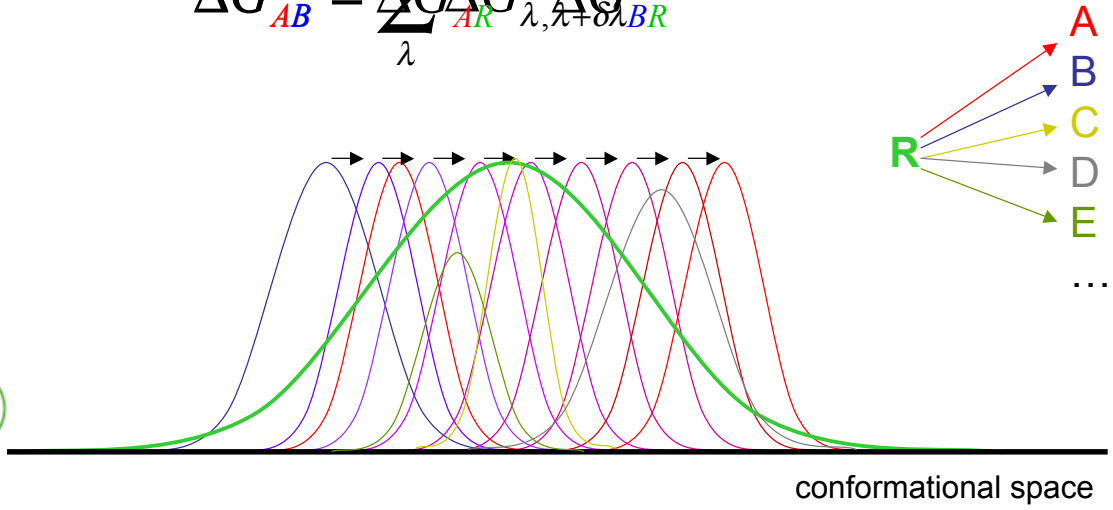


Modern Bioinformatics

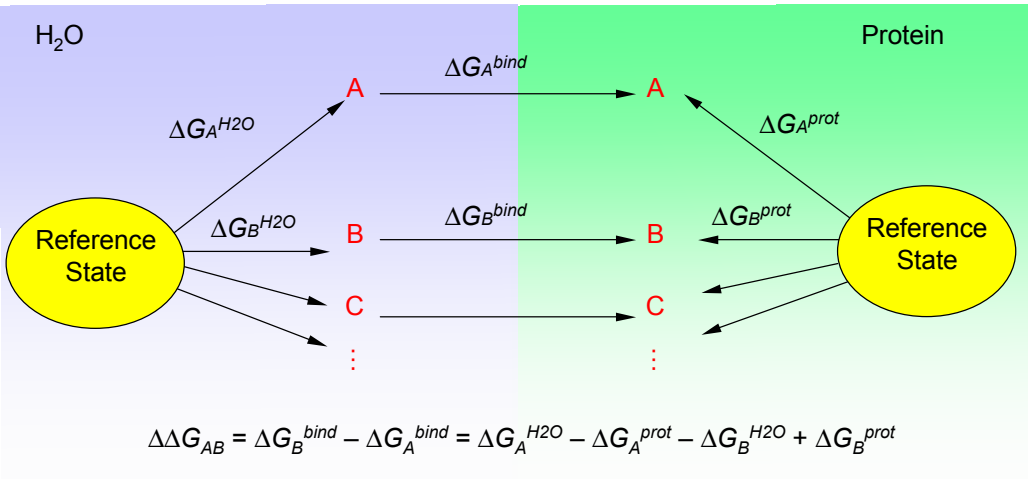
One step perturbation Enveloping Distribution Sampling

$$\Delta G_{AB} = -k_B T \ln \left\langle e^{-(E_A - E_B)/k_B T} \right\rangle_B$$

$$\Delta G_{AB} = \sum_{\lambda} \Delta G_{AR}^{\lambda,\lambda+\delta\lambda} - \Delta G_{BR}$$



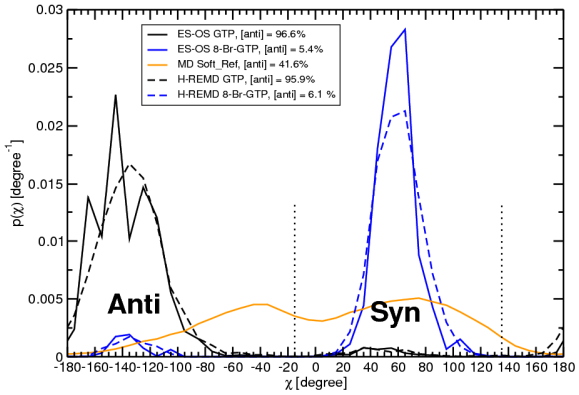
Thermodynamic cycle

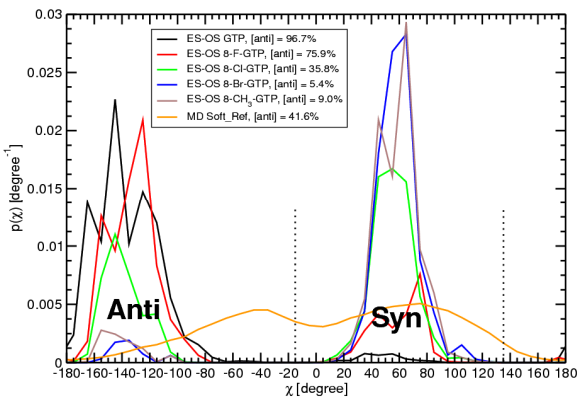


$$\Delta\Delta G_{AB} = \Delta G_B^{bind} - \Delta G_A^{bind} = \Delta G_A^{H2O} - \Delta G_A^{prot} - \Delta G_B^{H2O} + \Delta G_B^{prot}$$

Using the one step perturbation

- Simulate a reference molecule
  - Sugar-base interaction soft
  - 8-substituent soft with everything else
  - Apply perturbation formula to project back to real molecules
- Reproduce
  - experimental preference
  - ^{3}J-value for GTP (Exp:2.5/2.6/2.6/3.5 Hz Calc:2.42 Hz)
- OSP for 5 compounds
- In different media
  - Relative free energy of solvation
  - Relative LogP values





Free energies of solvation

- Both conformations contribute significantly
- The overall value does not follow intuitively from the values in one conformation

	$\Delta\Delta G_{AB}^{anti}(solv)$	$\Delta\Delta G_{AB}^{syn}(solv)$	$\Delta\Delta G_{AB}(solv)$
GTP	0	0	0
8-F-GTP	4.0	4.5	5.8
8-Cl-GTP	4.3	4.4	7.3
8-Br-GTP	1.4	0.4	4.1
8-CH_3-GTP	4.0	4.6	8.2

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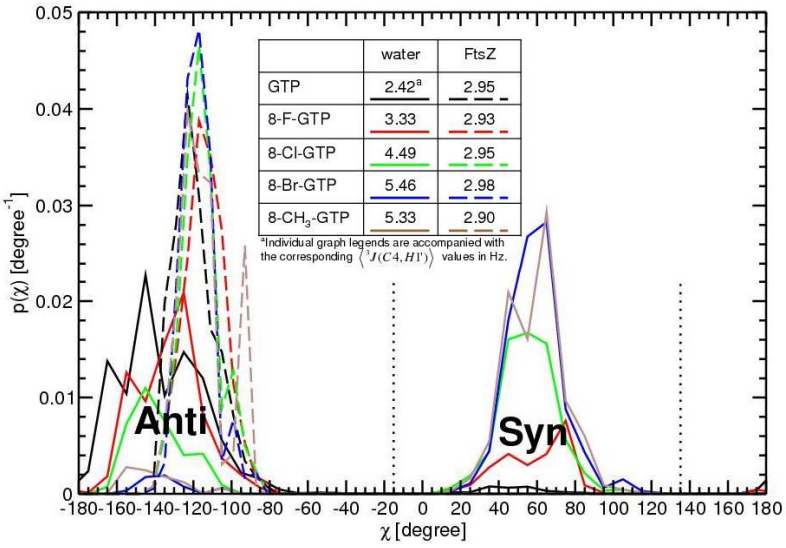
And now in a protein

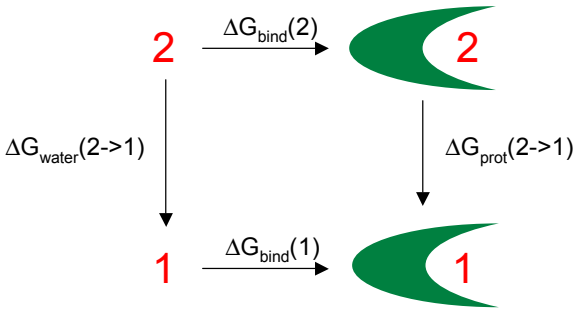
- Compounds are inhibitors of bacterial FtsZ protein
  - Different conformations of the compounds in the protein
- Recent X-ray structure shows only one conformation
  - Also with large substituents
- OSP in which only substituent is soft
  - No need for conformational enhancement

Conformational restriction

- In protein the conformational freedom is much restricted



Thermodynamic cycle



$$\Delta\Delta G_{bind} = \Delta G_{bind}(\mathbf{2}) - \Delta G_{bind}(\mathbf{1})$$
$$= \Delta G_{21}(prot) - \Delta G_{21}(water)$$

	$\Delta G^{OS}_{GTP,R}(FtsZ)$	$\Delta G^{ES-OS}_{GTP,R}(aq)$	$\Delta\Delta G^{calc}_{GTP,R}(bind)$	$\Delta\Delta G^{\exp}_{GTP,R}(bind)$
GTP	0	0	0	0
8-F-GTP	14.6	15.0	-0.5	--
8-Cl-GTP	16.5	12.8	3.7	8.0
8-Br-GTP	16.0	5.9	10.1	9.2
8-CH_3-GTP	23.0	12.6	10.5	~8.7

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Conformational restriction

- Free energy of restricting the analogs in water to the conformations observed in the protein

	$\left[\langle -140^\circ, -90^\circ \rangle\right]^R_{(aq)}$	$\Delta G^{rest}_R(aq)$	$\Delta\Delta G^{rest}_{GTP,R}(aq)$	$\Delta\Delta G^{calc}_{GTP,R}(bind)$
GTP	50%	1.7	0.0	0.0
8-F-GTP	48%	1.8	0.1	-0.5
8-Cl-GTP	18%	4.2	2.5	3.7
8-Br-GTP	3.3%	8.4	6.7	10.1
8-CH_3-GTP	4.0%	8.0	6.2	10.5

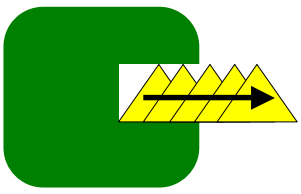
- The free energy of restricting the free energy explains 65% of the differences in binding free energies between the compounds
- Conformational selection of the ligand

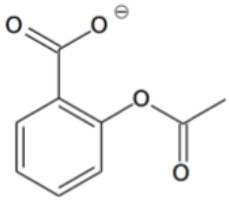
J. Hritz, T. Läppchen, C. Oostenbrink. *Eur. Biophys. J. (2010)* 29:1573

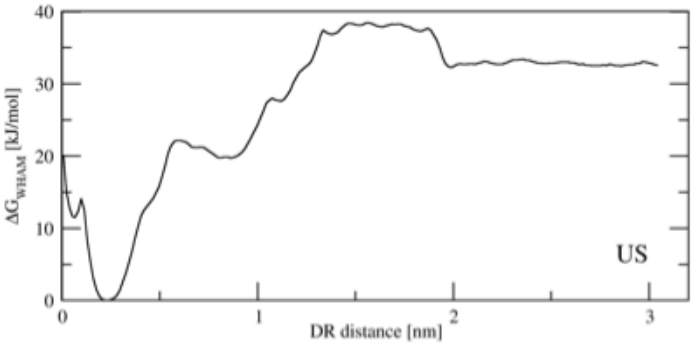
Modern Bioinformatics

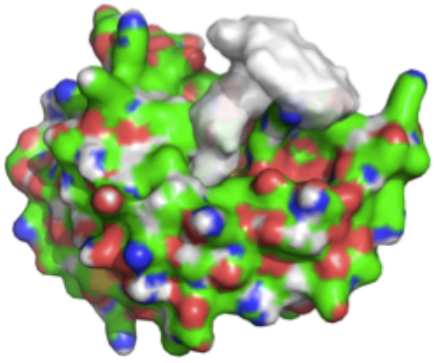
Binding free energies by pulling

- Aspirin binding to cytosolic Phospholipase 2
- Umbrella sampling with distance restraints from the active site
  GROMOS11, 54A7 parameter, 31 x 10 ns
- Weighted histogram analysis (WHAM)
- Barriers along the way



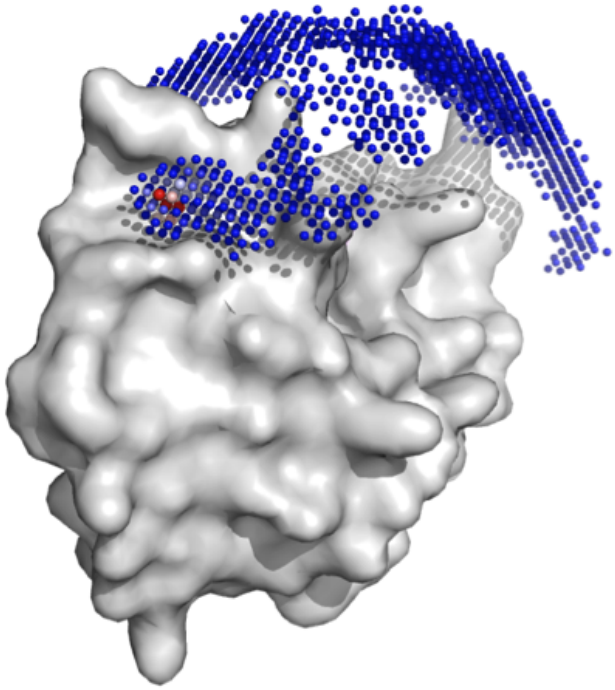






Barriers: real or artifact

- Centre of mass of aspirin
- Seems to get stuck behind a part of the protein
- Resolve
  - Single path
  - Reversible binding
- REMD with different replicas at different distance restraints



Modern Bioinformatics

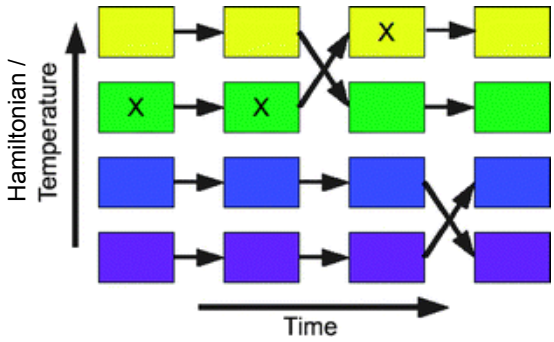
Replica exchange

- Run simulations at different conditions
- Mix them using the Metropolis criterion (MC)
- For each of the simulations you get a correct ensemble
- Replicas differ in temperature or in (λ-dependent) Hamiltonian

$$H(\mathbf{p},\mathbf{r},\lambda) = K(\mathbf{p}) + V^{phys}(\mathbf{r}) + V^{rest}(\mathbf{r},\lambda)$$

$$V^{rest}(\mathbf{r},\lambda) = \tfrac{1}{2} K\left[(1-\lambda) r_0^A + \lambda r_0^B - r_{ij}\right]^2$$

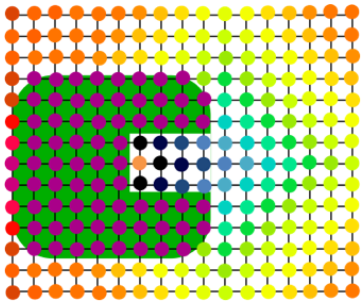
- At large distances, the ligand diffuses
- Returns via a different pathway
- Broad ensemble at every λ
- Reversible binding



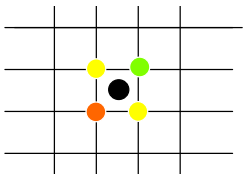
Sugita, Y.; Kitao, A.; Okamoto, Y. *J. Chem. Phys.* **113**, 6042–6051 (2000)
Figure: A. Patriksson, D. van der Spoel, Phys. Chem. Chem. Phys., **10**, 2073 (2008)

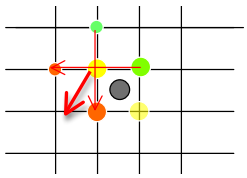
Distancefield distances

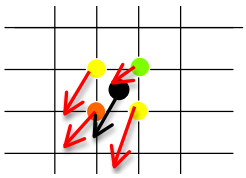
- Grid based distances to *avoid* the protein
- Curved routes
- Dijkstra's algorithm



$$V(\mathbf{r}) = \tfrac{1}{2} K \left[l(\mathbf{r}) - l_0 \right]^2 \qquad \mathbf{f}_i(\mathbf{r}) = -\frac{\partial V(\mathbf{r})}{\partial \mathbf{r}_i} = -K \left[l(\mathbf{r}) - l_0 \right] \frac{\partial l(\mathbf{r})}{\partial \mathbf{r}_i}$$





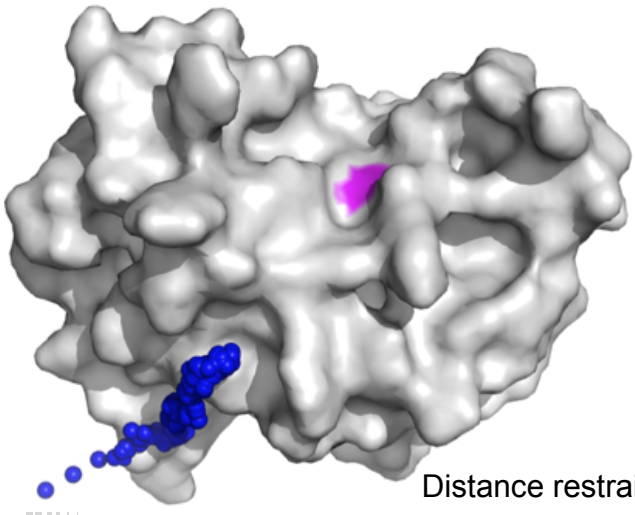


$l(r)$ by interpolation

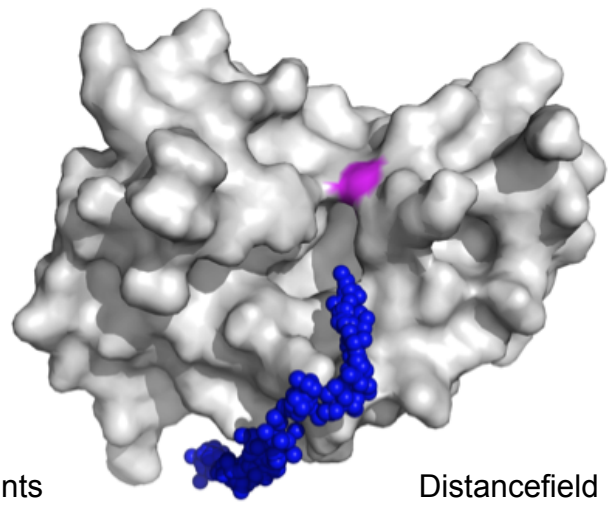
$\frac{\partial l(\mathbf{r})}{\partial \mathbf{r}_i}$ from finite differences, followed by interpolation

Distancefield distances

- Distance restraints push into the protein and distort structure
- Distancefield restraints curve around the protein



Distance restraints

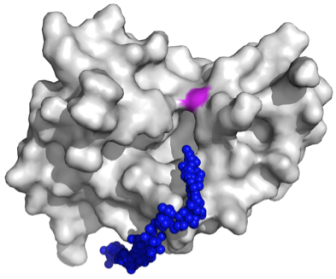


Distancefield

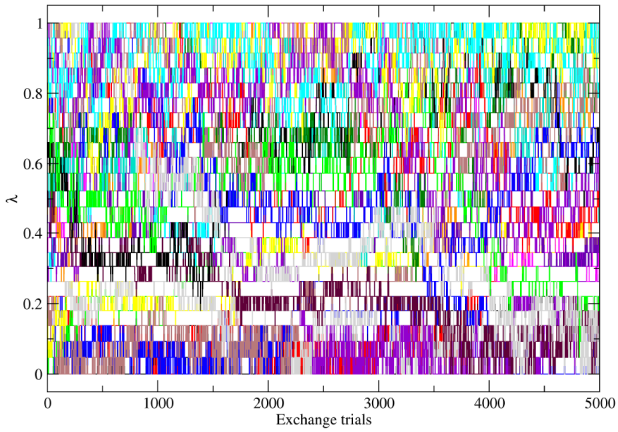
Application of distancefield

Distancefield coordinate allows for reversible binding / unbinding

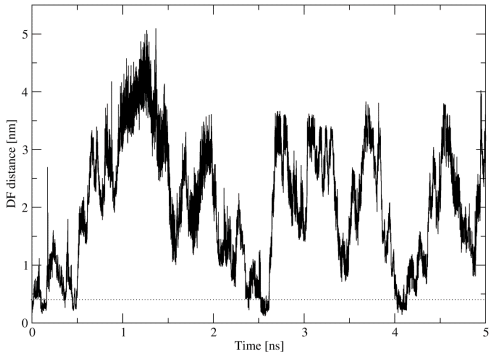
Various applications implemented in GROMOS



Slow / fast growth free energy calculations

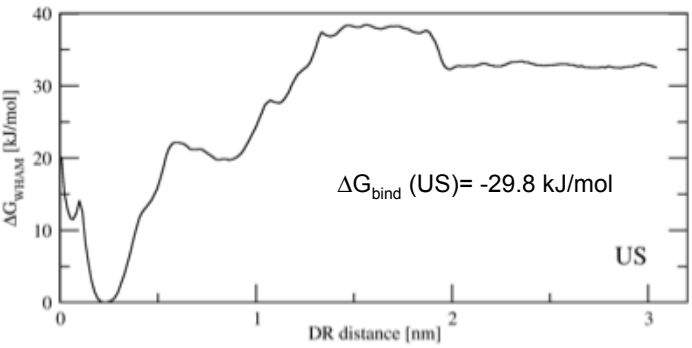


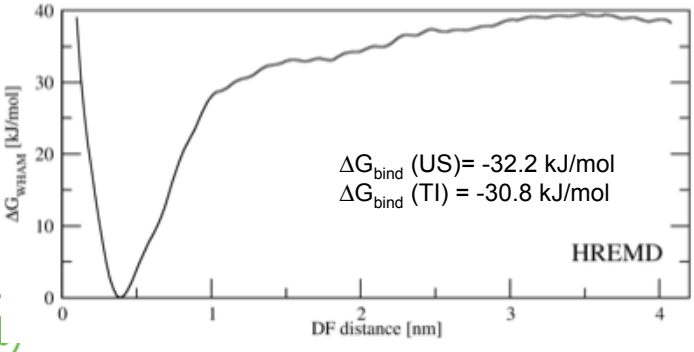
Hamiltonian replica exchange

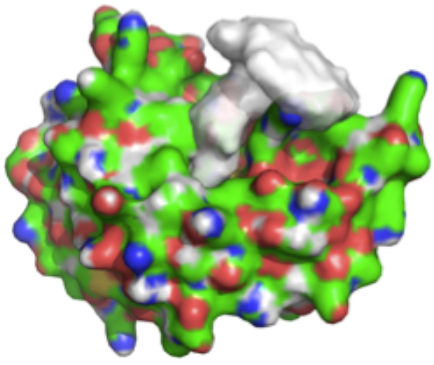


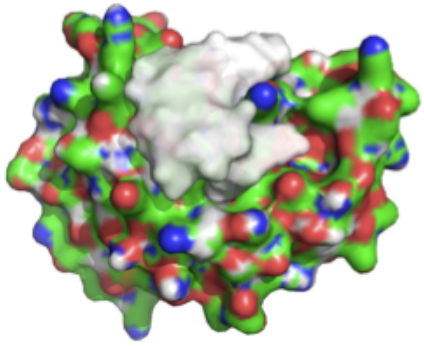
Local elevation on the distance field coordinate

Resulting PMF and routes









ΔG_{bind} (exp)= -29.6 kJ/moll

De Ruiter and Oostenbrink, *J. Chem. Theory Comp.* (2013) **9**:883

Modern Bioinformatics

BOKL

n Bioinformatics

Papers I

- J. Am. Chem. Soc. 2011, 133, 7016
- A nice demonstration on how molecular dynamics simulations may be used to understand the effect of experiments that are difficult to control.

JACS
JOURNAL OF THE AMERICAN CHEMICAL SOCIETY
ARTICLE
pubs.acs.org/JACS

Microscopic Analysis of Protein Oxidative Damage: Effect of Carbonylation on Structure, Dynamics, and Aggregability of Villin Headpiece

Drazen Petrov and Bojan Zagrovic*

n Bioinformatics

Papers II

- J. Chem. Theory Comput. 2012, 8, 3686
- A paper by ourselves, in which a more efficient method to calculate free energies is evaluated.
- (quite challenging…)

JCTC Journal of Chemical Theory and Computation
Article
pubs.acs.org/JCTC

Efficient and Accurate Free Energy Calculations on Trypsin Inhibitors

Anita de Ruiter and Chris Oostenbrink*

Institute for Molecular Modeling and Simulation, University of Natural Resources and Life Sciences (BOKU), Vienna, Austria

Conclusions

- Molecular dynamics simulations form a powerful tool to study biomolecules
  - Insight into structure, dynamics and function at an atomic level
  - Complementary to experiment
- Free energy calculations for e.g. drug design / lead optimisation
  - Enthalpic and entropic effects should be included
  - Statistical mechanics and thermodynamic cycles
    - Efficient calculations
    - Unphysical intermediates
- Protein flexibility and multiple binding conformations contribute