Protein-Ligand Interactions and Energy Evaluation Methods*

*with a revealing look at roles of water

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Where Do We Start?

Thermodynamics:

ΔH, ΔS, ΔG are the **language** of energy and interactions, and it *seems* simple, but parsing atomic-scale observations into these terms is not often easy.

Chemistry (and Physics):

Everything that happens at the atomic scale is chemistry and/or physics; we must interpret interactions from this perspective *before* attempting energetic evaluations.
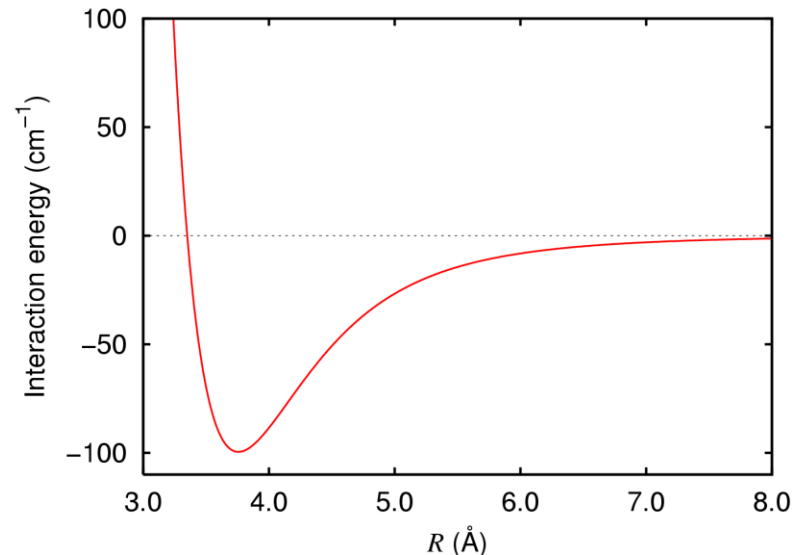
Interactions

Physics:

Coulombic (electrostatic)

$$E_{Coulombic} = C \sum_{i=1} \sum_{j>1}^{\text{All nonbonded}} \frac{Q_i \, Q_j}{\varepsilon_{ij} \, r_{ij}}$$

Q_i and Q_j are *partial* atomic charges, e.g., from electronegativity differences between atoms i and j.
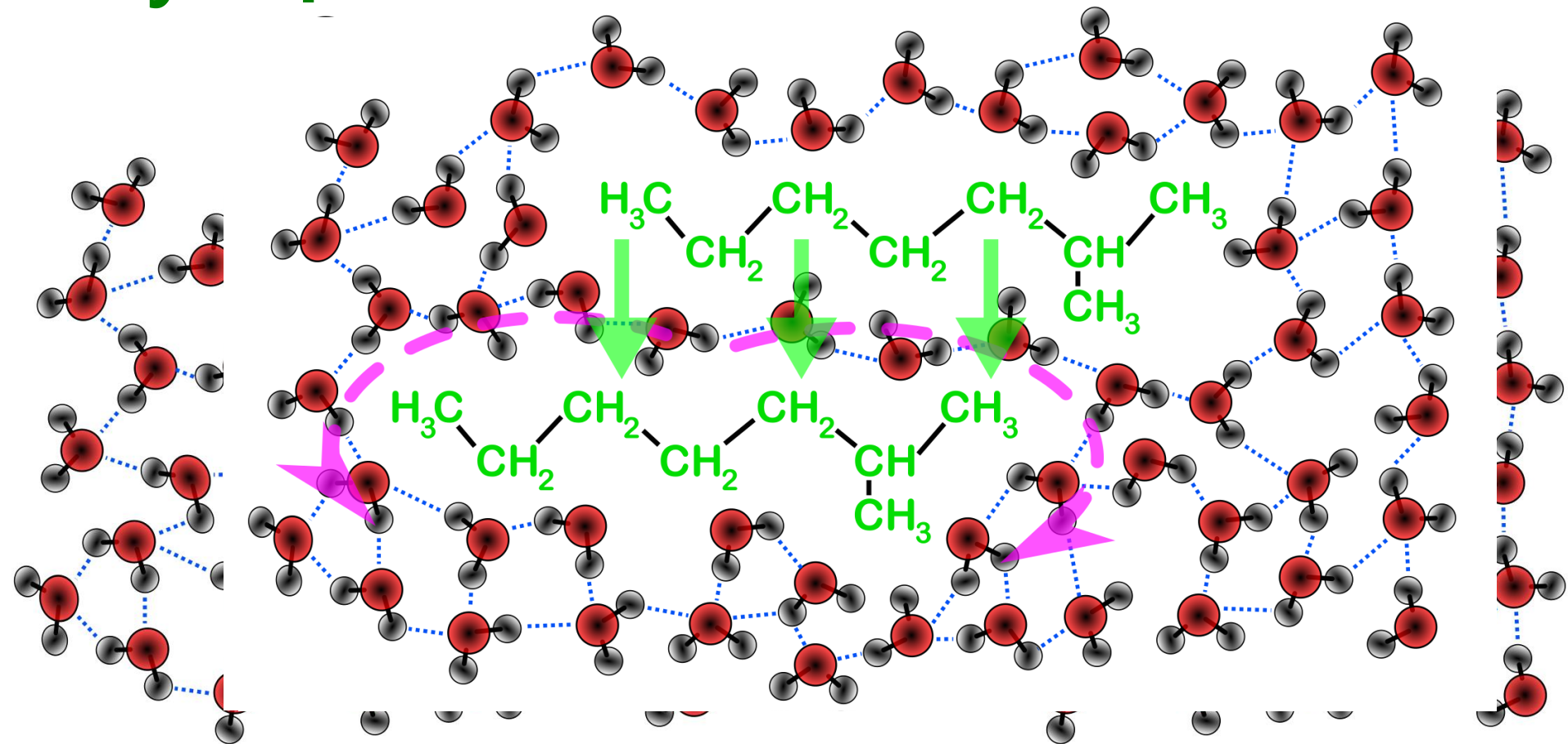
van der Waals (London forces)

$$E_{vdW} = \sum_{i=1} \sum_{j>1}^{\text{All nonbonded}} \varphi_{ij} \left[\frac{1.0}{r_{ij}^{12}} - \frac{2.0}{r_{ij}^{6}} \right]$$

Interactions

Chemistry:

Hydrophobic
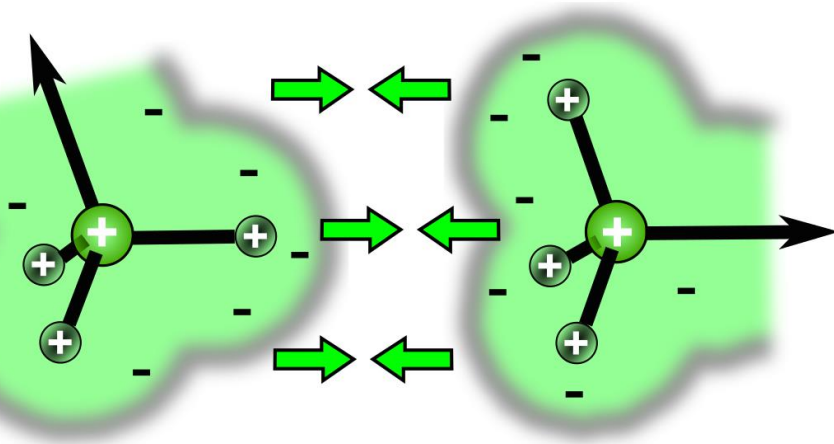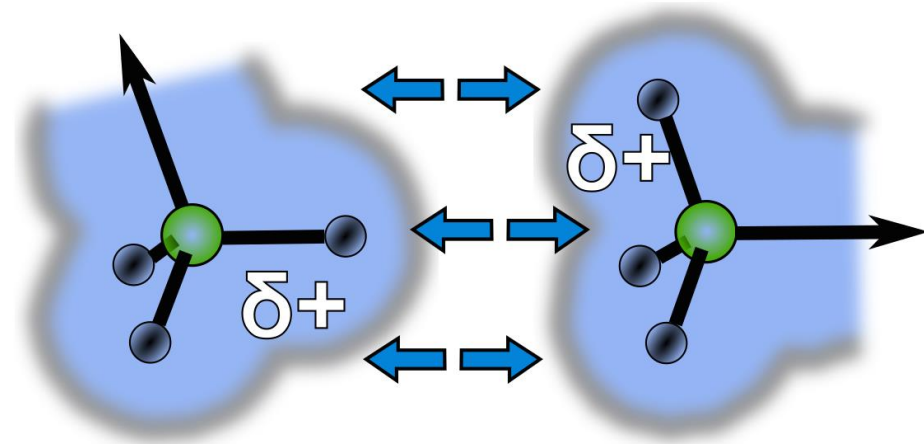
Interactions

Chemistry:

Are hydrophobic interactions the same as vdW?

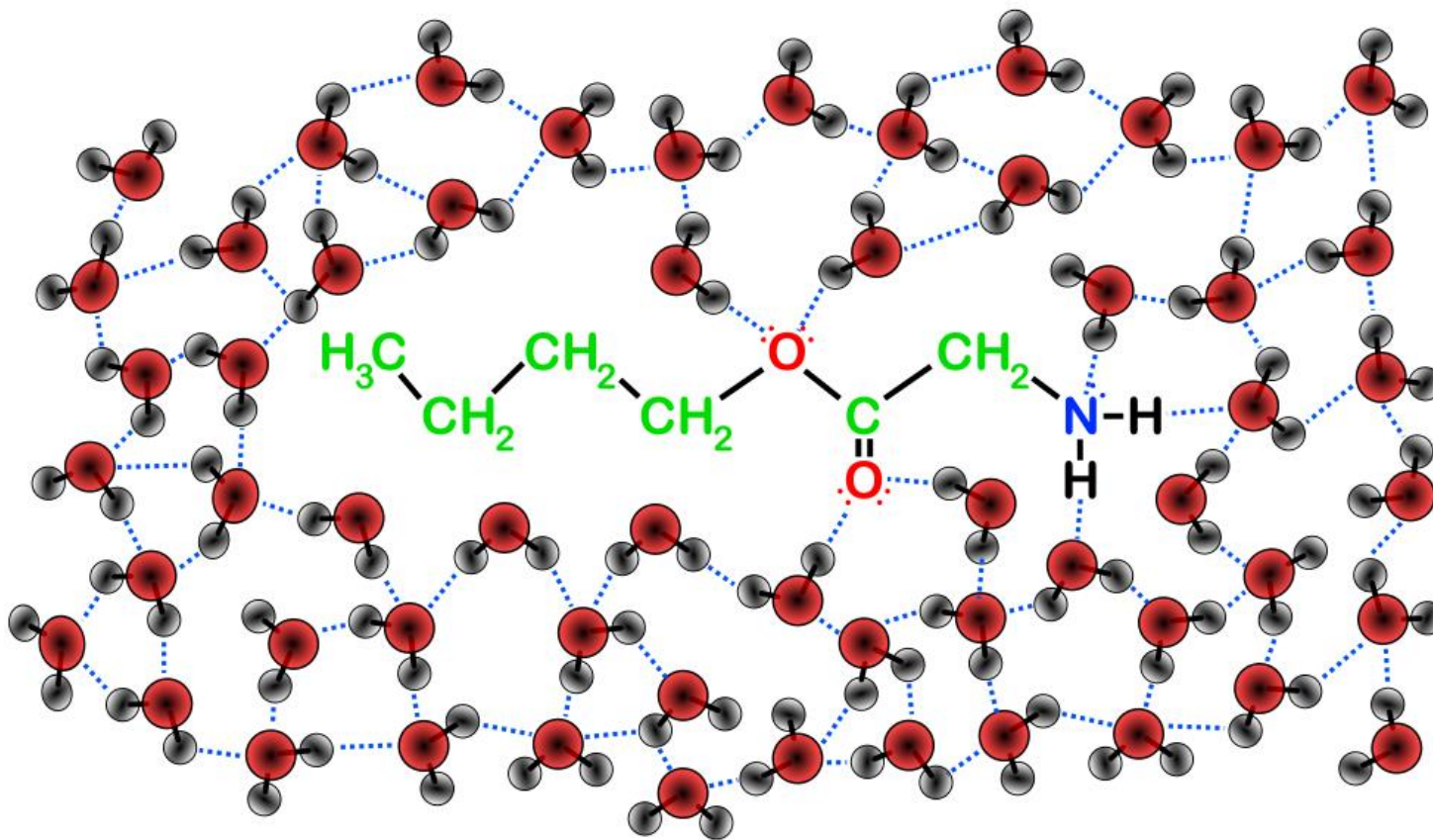van der Waals (London)   Electrostatic (Coulombic)

While pairwise van der Waals supports the hydrophobic effect, pairwise Coulombic forces contraindicate attraction between hydrophobic groups or atoms. The hydrophobic effect must be considered an **emergent property** of the entire system.
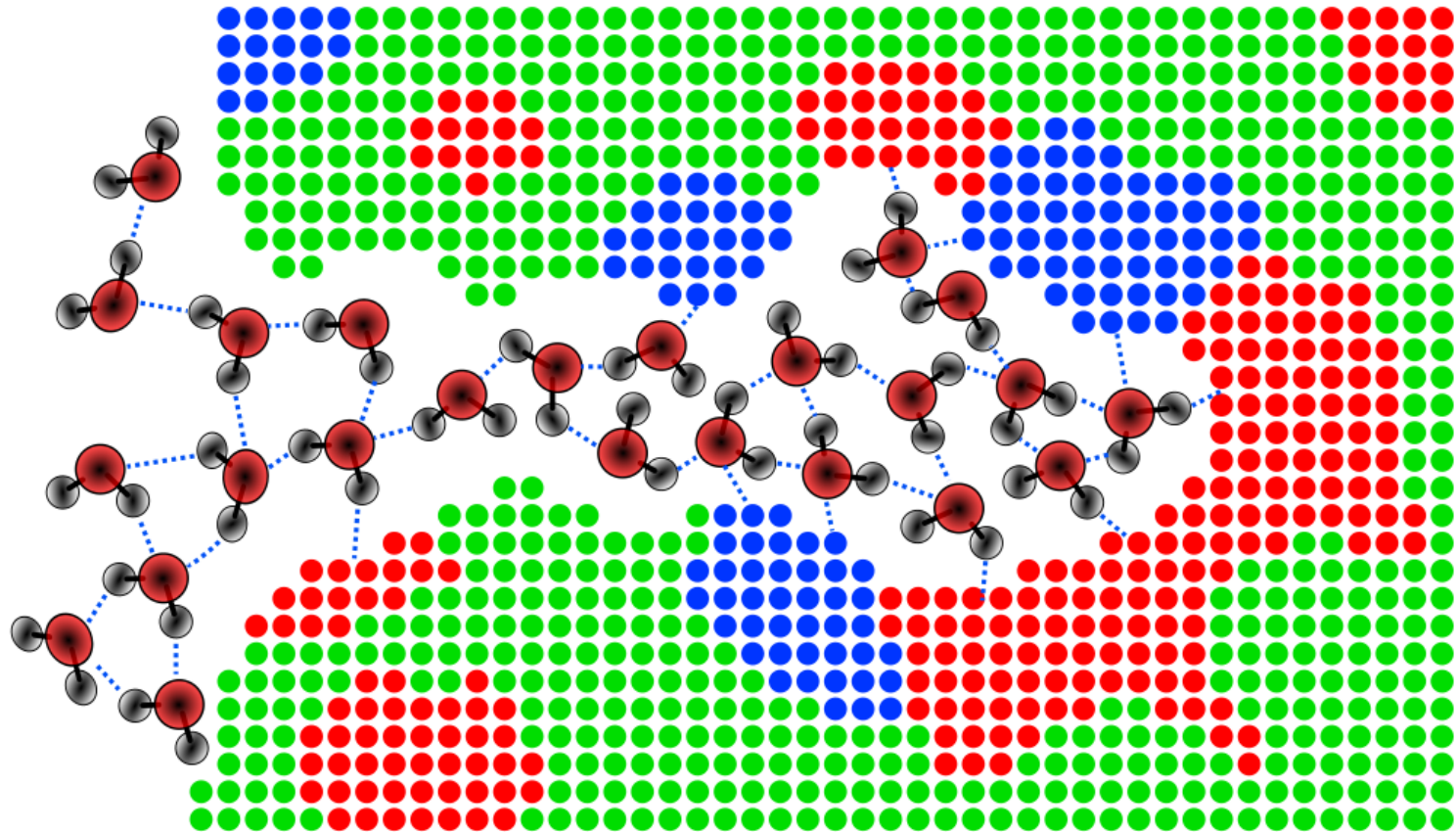
Interactions

Chemistry:

Desolvation of Ligand

Interactions

Chemistry:

Site-Ligand Complex

Thermodynamics

Enthalpy (ΔH):

- ΔH during a chemical reaction is the heat absorbed or released in the breaking or formation of bonds.

- This applies to ligand binding as well, even if no *covalent* bonds are changed:

  - *van der Waals interactions*

  - *hydrogen bonding*

  - *charge (Coulombic) interactions*

Thermodynamics

Entropy (ΔS):

- ΔS measures order (disorder) in the system. For a reaction to be spontaneous, the entropy of the universe, ΔS_{total}, must increase.

- In ligand binding, this manifests as:

  - *changes in solvent or counterion arrangement*

  - *rotational and translational changes*

  - *reflecting the degrees of freedom*

Thermodynamics

Gibb's Free Energy (ΔG):

$$\Delta S_{total} = \Delta S_{system} + \Delta S_{surroundings}$$

$$(\Delta S_{surroundings} = -\Delta H_{system} / T)$$

$$\Delta S_{total} = \Delta S_{system} - \Delta H_{system} / T$$

$$-T \Delta S_{total} = \Delta H_{system} - T \Delta S_{system}$$

$$\Delta G = \Delta H_{system} - T \Delta S_{system}$$

Also, since **for spontaneous reactions**,

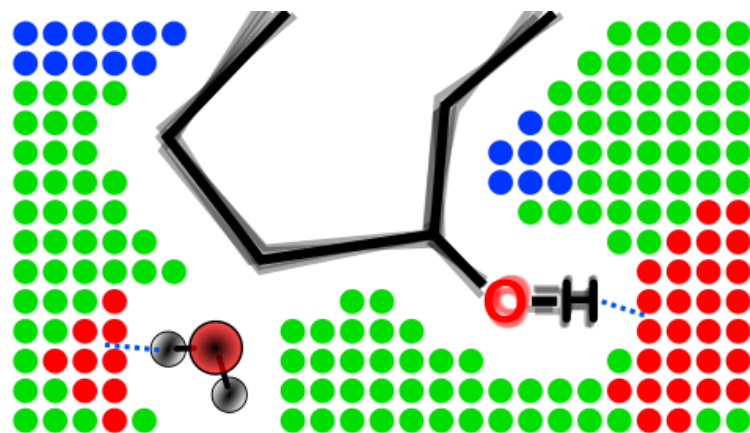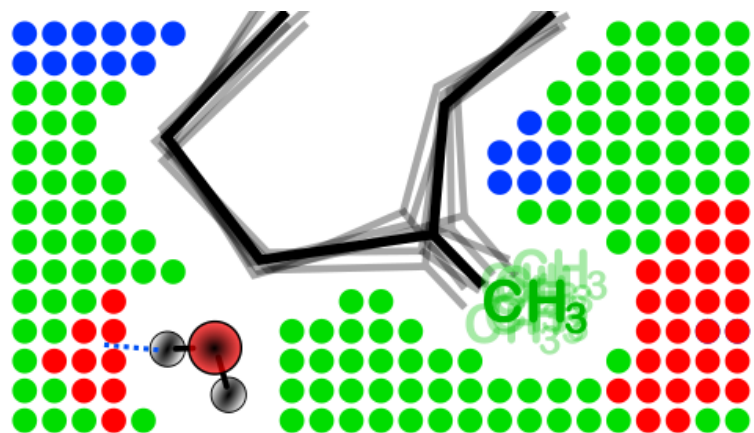$\Delta S_{system} > \Delta H_{system} / T$, then:

$\Delta G = \Delta H_{system} - T \Delta S_{system} < 0$;

i.e., the **Gibbs free energy** *must* be **negative**.

Thermodynamics

Enthalpy/Entropy Compensation (H/SC):
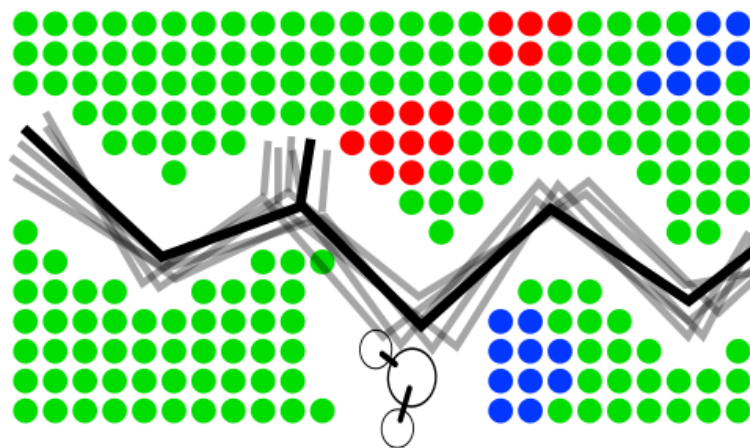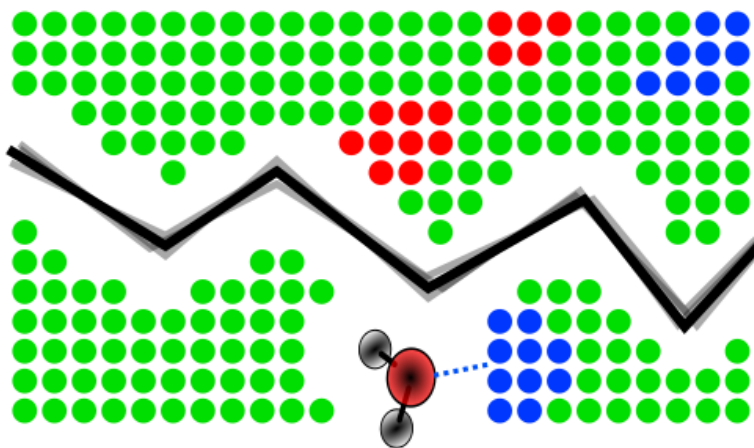
Thermodynamics

The Hydrophobic Effect:

- **Enthalpic or Entropic?**

  - It *seems* to be a lot like a van der Waals interaction, which is most certainly enthalpic.

  - But, things are not always what they seem!

  - Recall that the **origin** of the hydrophobic effect is related to water rearrangements (and possibly motion), i.e., as the water seeks to form better hydrogen bonds with other water molecules and polar species.

  - **The hydrophobic effect is thus *entropic!***

Thermodynamics

Displacing Water I:

- **Enthalpic or Entropic?**

  -
  -
  -
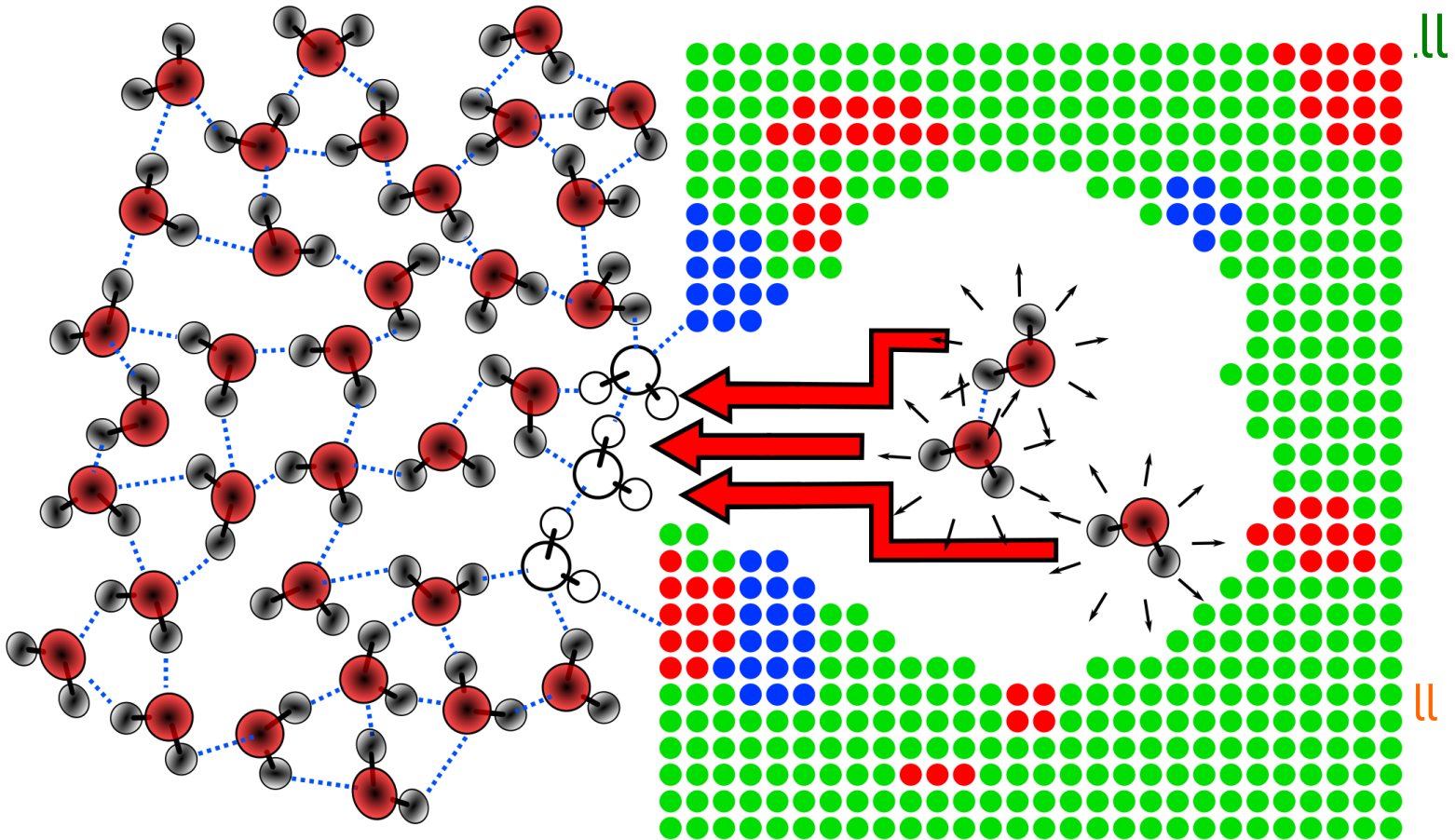
Thermodynamics

Displacing Water II:

- **Enthalpic or Entropic?**

  - If the w.., it will be difficul.......
    - (Thi.....Arg87............
  - Such a...................................... entropic gain ca.....
    - New...enthalpically favo.....
  - Howev.......................................olar site has a much.....
    - Both...er favorably or unfa.....
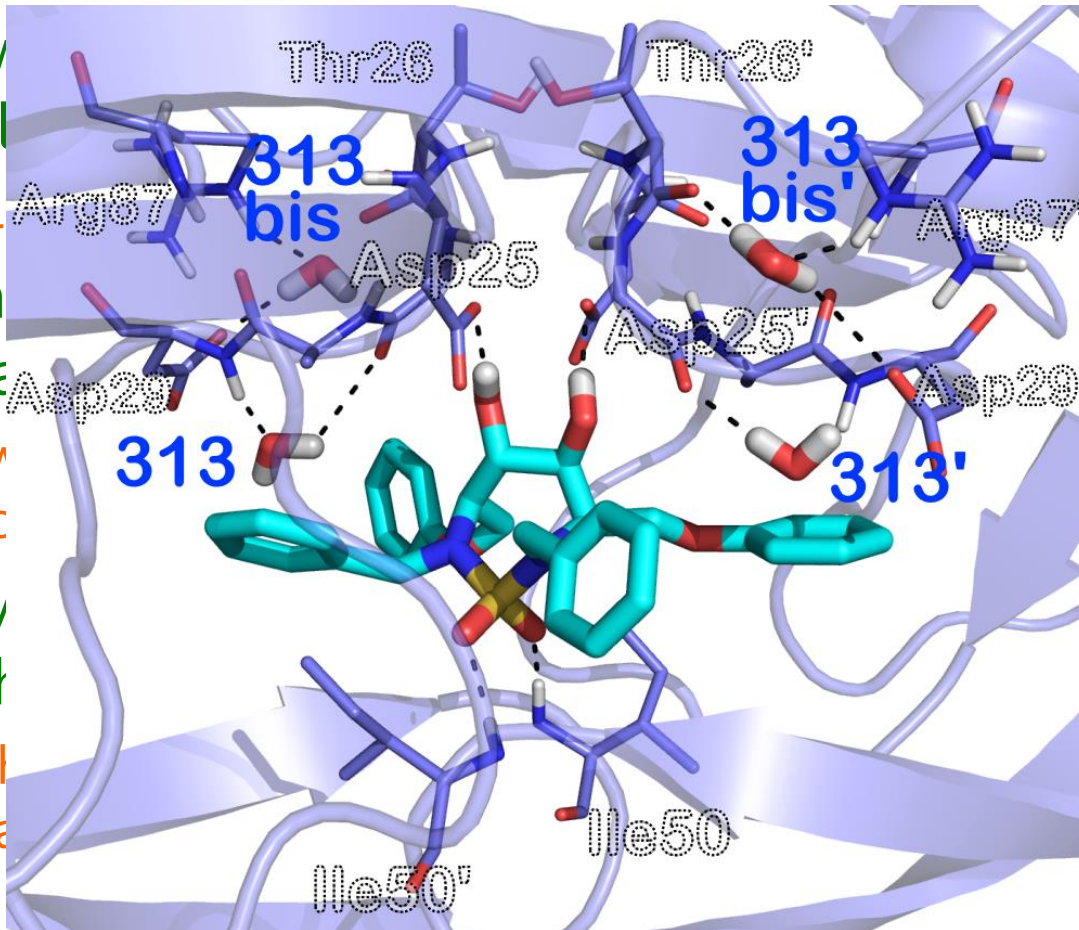
Energy Evaluation Methods

Free Energy:

- **It should be obvious that to get the complete picture of an interaction, *both* its enthalpic and entropic contributions must be considered.**

  - Estimating/calculating the hydrophobic effect is problematical – at best!

    - Few, if any, full-scale simulations have demonstrated this emergent property of the protein/ligand/solvent system.

    - Thus, "phenomenological" observations are usually used to estimate the hydrophobic contribution to an interaction

  - But, the schizophrenic roles that water can play in mediating interactions are very difficult to predict.

    - Each case may be *special*. Only by supplementing models with complete experimental structural and thermodynamic data can the case-specific contributions of water be understood.

Energy Evaluation Methods

Free Energy:

- **Empirical and Knowledge-based Methods**

  - Structural and other experimentally measured data form the basis of several types of energy evaluation methods.

  - Knowledge-based methods presume that there is nothing new: just identify a similar collection of atoms and the results can be extrapolated.

  - Empirical methods (HINT) use relevant data from other phenomena (like logP) to evaluate interactions.

  - Some programs:
    - HINT
    - PyWATER
    - WaterDock
    - PMF / wPMF

Energy Evaluation Methods

Free Energy:

- **Static Molecular Mechanics Methods**

  - Use forcefields to determine likely loci of water or other functional groups in a protein matrix.

  - Most docking algorithms apply molecular mechanics forcefields to energy-evaluate poses.

  - No real scope to simulate entropic terms.

  - Some programs:

    - GRID
    - MCSS
    - FLAP/WaterFLAP
    - AutoDock
    - WaterMap

Energy Evaluation Methods

Free Energy:

- **Statistical Mechanics Methods**
  - Based on well-validated theories of fluid motion/ mechanics and density functional theory.
  - Entropy is implicit in the models.
  - Some programs:
    - 3D-RISM
    - SPAM

Energy Evaluation Methods

Free Energy:

- **Molecular Dynamics / Monte Carlo Methods**
  - Attempt to "model" entropy with temperature- and/or time-dependent simulations.
  - Free Energy Perturbation methods have been widely and effectively used.
  - Explicit water simulations have often revealed roles and energetics of water in protein-ligand systems.
  - Some programs:
    - BiKi Hydra
    - JAWS
    - RETI
    - STOW
    - WaterMap

Energy Evaluation Methods

Free Energy:

- **Continuum Solvent Model Methods**
    - Two key models: Poisson-Boltzmann (PB) and Generalized Born (GB). PB has better physics.
    - These are *implicit* solvation models, i.e., most (or all) of the effects of solvent molecules are treated as fields.
    - Best results obtained when hybridized with other methods.
    - Some programs:
        - DelPhi
        - Zap
        - SZMAP

Further Reading

The Roles of Water in the Protein Matrix: A Largely Untapped Resource for Drug Discovery. F. Spyrakis, M.H. Ahmed, A.S. Bayden, P. Cozzini, A. Mozzarelli and G.E. Kellogg; *J. Med. Chem.* (in press)

Applying Thermodynamic Profiling in Lead Finding and Optimization. G. Klebe; *Nat. Rev. Drug Discovery* **2015**, *14*, 95-110.

A Medicinal Chemist's Guide to Molecular Interactions. C. Bissantz, B. Kuhn, M. Stahl; *J. Med. Chem.* **2010**, *53*, 5061-84.

Is It the Shape of the Cavity, or the Shape of the Water in the Cavity? P. Snyder, M. Lockett, D. Moustakas, G. Whitesides; *Eur. Phys. J. Spec. Top.* **2013**, *223*, 853-891.

Hydrophobicity-Shake Flasks, Protein Folding and Drug Discovery. A. Sarkar, G.E. Kellogg; *Curr. Top. Med. Chem.* **2010**, *10*, 67-83.

High End GPCR design: Crafted Ligand Design and Druggability Analysis Using Protein Structure, Lipophilic Hotspots and Explicit Water Networks. J.S. Mason, A. Bortolato, D.R. Weiss, F. Deflorian, B. Tehan, F.H. Marshall; *In Silico Pharmacol.* **2013**, *1*, 23.

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