## Lecture 7:  Protein Structure & Biochemical Energetics I

Assigned reading in Campbell: Chapter 4.4-4.6 (Chapter 1.10-1.12)

Key Terms:
> **Properties of folded states**
> **Enthalpy ( $\Delta H$ )**
> **Hydrogen bonds**
> **Van der Waals forces**
> **Electrostatic interactions**
> **Entropy ( $\Delta S$ )**
> **Conformational entropy (S=R lnW)**
> **Hydrophobic effect, water ordering**

### 7.1  Forces that drive protein folding:

Non-covalent interactions are 2 to 3 orders of magnitude weaker than covalent bonds; they act at short range and are exceedingly numerous.  *A key feature of protein structure is that the stability depends on the simultaneous presence of all the non-covalent interactions of the native state*.  Thus, the interactions described below cooperate to produce the native structure.

> **Hydrogen bonds:** due to partial electrostatic charges most commonly between amide (N-H) and carbonyl (C=O) groups in the main chain (~1-2 kJ/m per H-bond), as well as side chain-side chain and main chain-side chain (another ~0.5 kJ/m per residue).

> **Van der Waals contacts:** attractive force that occurs when transiently induced dipoles between any pair of molecules (polar or non-polar) come in close proximity (0.5-1 kJ/m per pair of atoms)

> **Electrostatic interactions:** attraction between oppositely charged ionic groups; generally do not impact protein folding

> **Hydrophobic effect:** due to the entropy of *water* in the system.

**Features of the folded State:**

| Amino Acid Distribution | Location | |
|---|---|---|
| | Inside | Surface |
| Charged | | |
| Polar | | |
| Non-polar | | |
| Amphipathic (e.g. Lysine) | | |

| Native State | Denatured or Unfolded State |
|---|---|
| Compact | |
| Single Conformation | |
| Extensive Secondary Structure | |
| Extensive Tertiary Structure | |

**Thermodynamic Factors that affect the Stability of the Native State:**

A. **Enthalpy ( $\Delta H$ )** is the amount of heat generated/consumed by the reaction and is related to changes in molecular interactions (i.e. hydrogen bonds, van der Waals contacts). In biochemistry, these interactions usually involve non-covalent changes.
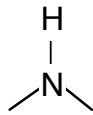
Contribution of ( $\Delta H$ ): Reactions that release heat (*decrease* in $\Delta H$ ) are *favorable*. Therefore, a *negative* value for $\Delta H$ indicates that the *products are favored.*

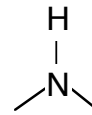In the case of the transition from the native, or folded, state to the unfolded state:

   $\Delta H$ associated with **hydrogen bonding** is *unfavorable* for *unfolding*. Hydrogen bonds are more stable in the native form of the protein.
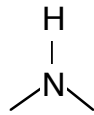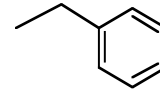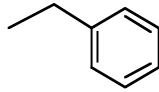
| H | H | H |
|---|---|---|
| | | |
| N | N | N |
| Native | Unfolded | No acceptor |

   $\Delta H$ associated with **van der Waals** forces is *unfavorable* for *unfolding*. Van der Waals interactions are more stable in the native form of the protein.

   **Electrostatic forces**: Although these forces contribute to $\Delta H$ changes in biochemical interactions, they are not important for protein folding. They do, however, play important roles in quaternary structure and protein-protein interactions.

B. **Entropy ( $\Delta S$ )** is related to the change in the number of possible configurations (W) of the system when the reaction occurs:
Disorder is always favored! Reactions that *increase* disorder *increase* $\Delta S$ .

**Two components to $\Delta S$ :**
**Configurational Entropy**: When a protein unfolds the entropy of the molecule increases dramatically due to a change in the conformational freedom of the $\Phi$ and $\Psi$ angles of the main-chain, as well as disordering of the side-chain.

The number of conformational states in a 50 residue *folded* protein is:

Giving an entropy of:

_____

The number of conformational states/residue in the *unfolded* protein is:

Giving an entropy of:

_____

The number of conformational states in a 50 residue *unfolded* protein is:

Giving an entropy of:

_____

**Entropy Changes of the Solvent:  The hydrophobic effect:**
The hydrophobic effect is due to the entropy of the *water* in the system.  When a non-polar side chain is exposed to water, it orders, or *decreases* the entropy of, the water molecules. However, when the non-polar residue becomes buried in the non-polar center of the protein it releases all of the water which coated it.  The released water can now freely diffuse in the solvent.  This results in an increase in entropy of the water.  Note that this is an effect, not really a force.  However, it is often referred to as the "driving force for protein folding".
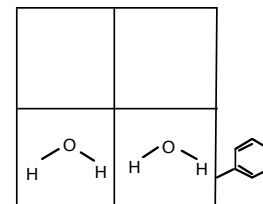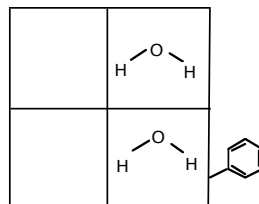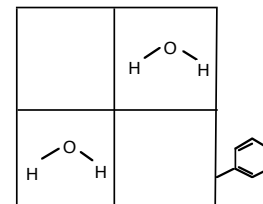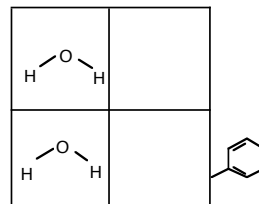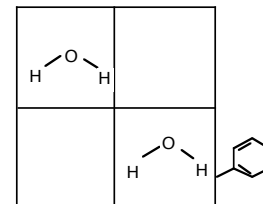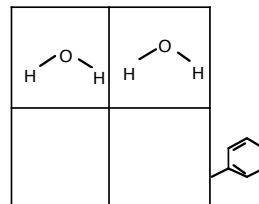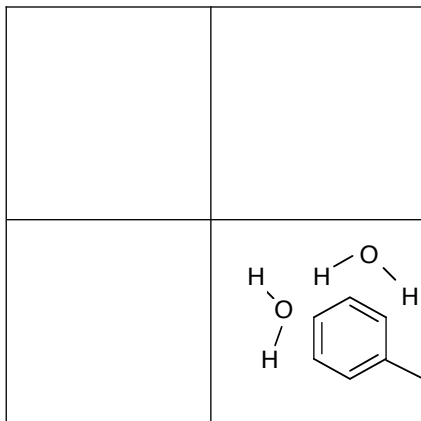
*Quantitative Description of the Hydrophobic Effect*: Since the coating of water molecules on a non-polar sidechain is loosely similar to the coating of ice on a surface you would expect the magnitude of the effect to depend on the surface area of the non-polar residue. The graph below shows the entropy change for 10 hydrophobic amino acids relative to glycine when the environment is changed from water to being buried in the protein.

The x-axis is the $H_2O$ accessible surface area of the side-chain.

Two conclusions can be drawn from this correlation:
1) For all non-polar side-chains $\Delta S$ is positive, indicating the release of ordered water when they are transferred to the organic phase.
2) The $\Delta S$ is proportional to the surface area. The line has a slope of 22 cal/mol/$A^{o2}$.