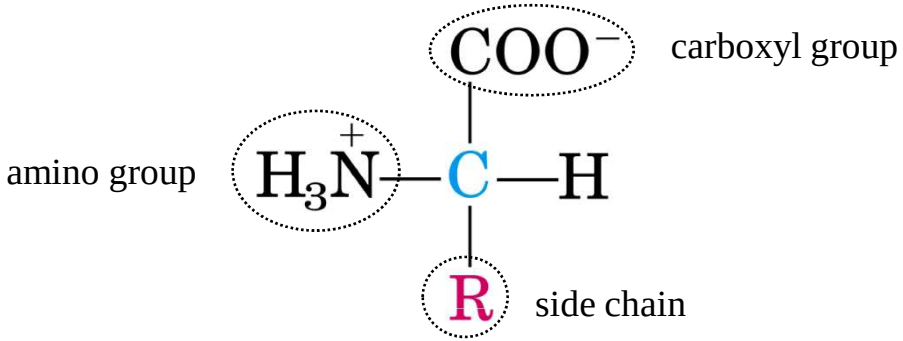
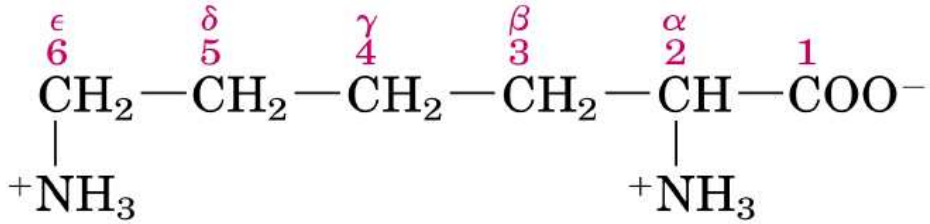
Stability of Protein Structures

http://www.biochem.med.umich.edu/bc550_secure/lecture%20materials/xu/lecture_1.ppt

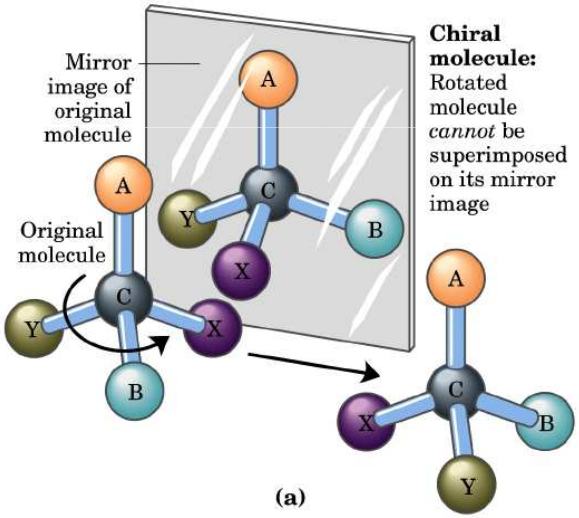
All proteins are composed of the 20 “standard” amino acids

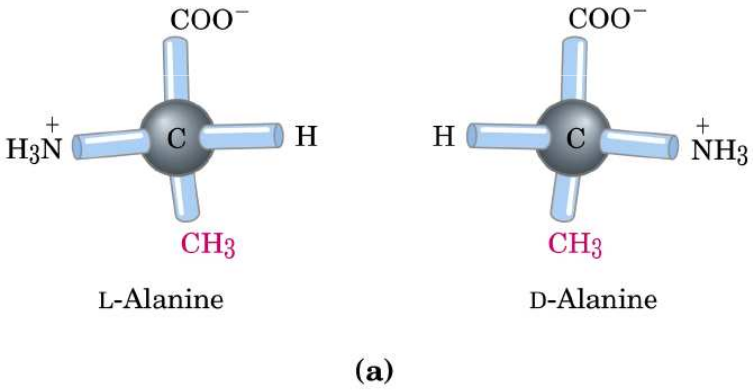




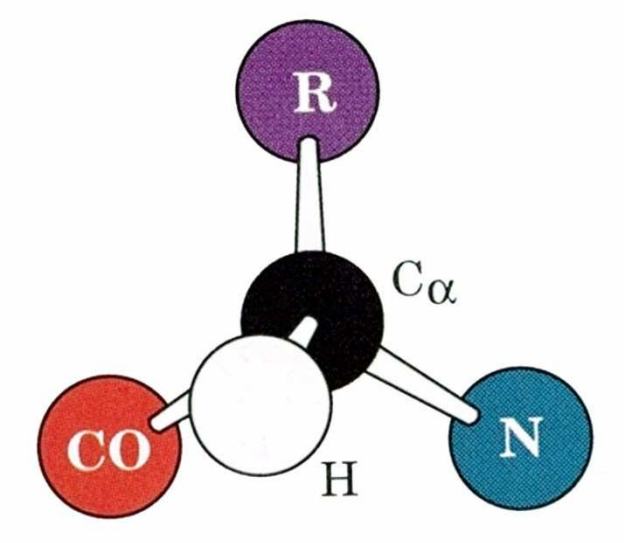
Lysine

Amino acids are chiral molecules





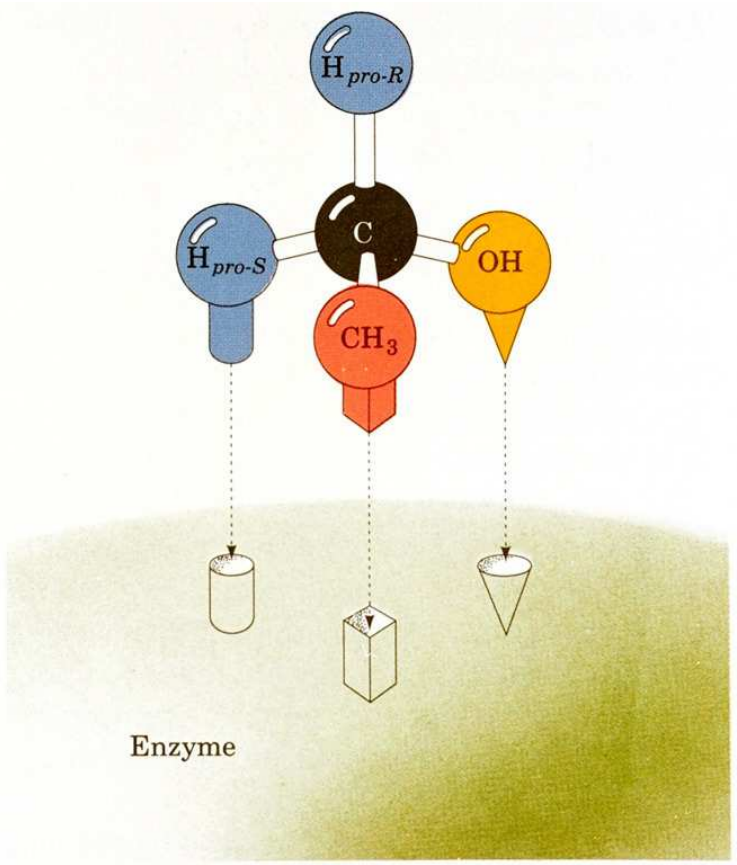
All amino acids found in proteins encoded by the genome have the L-configuration



The CORN law:

Imagine looking along the H-Cα bond with H atom closer to you. When read clockwise, the groups attached to the Cα spell the word CORN.

Chirality is important in biochemistry



Enzymes are highly specific both in binding chiral substrates and in catalyzing their reactions. This stereospecificity arises because enzymes, by virtue of their inherent chirality, form asymmetric active sites. They are absolutely stereospecific in the reactions they catalyze.

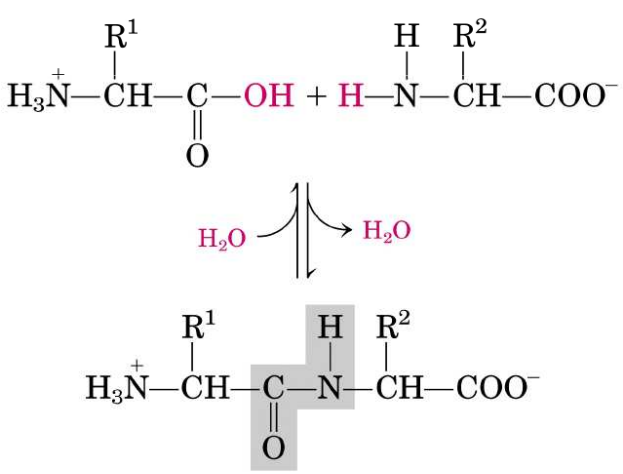
Side chains confer functional specificity

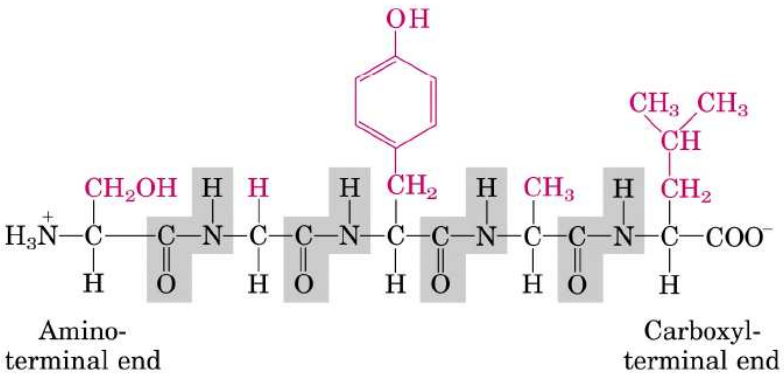
- Side chains are the determinants for proteins' structure and function.

- Charged and polar side chains are chemically active. They usually provide important catalytic residues in active sites. Their abilities to form specific hydrogen bonds often confer specificity in protein-ligand, protein-protein interactions.

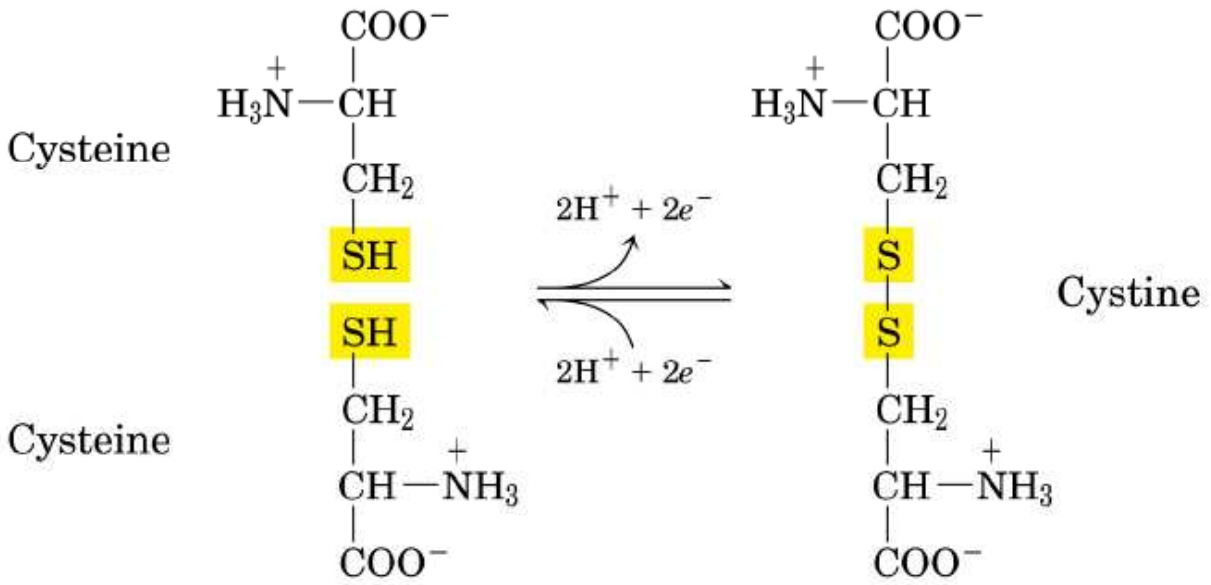
- Non-polar side chains are very important in supporting the overall protein architecture. They are the driving force for protein folding and protein-protein interactions.

Amino acids in proteins are linked together by peptide bonds





Polypeptide chains can be cross-linked together by forming disulfide bonds



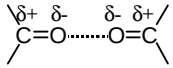
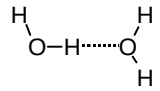
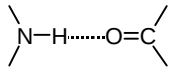
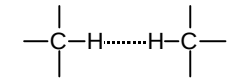
Noncovalent interactions are of outmost importance for biological systems

- Noncovalent interactions drive the spontaneous folding or polypeptide and nucleic acid chains and the spontaneous formation of membranes.

- Noncovalent interactions are one to three orders of magnitude weaker than covalent interactions.

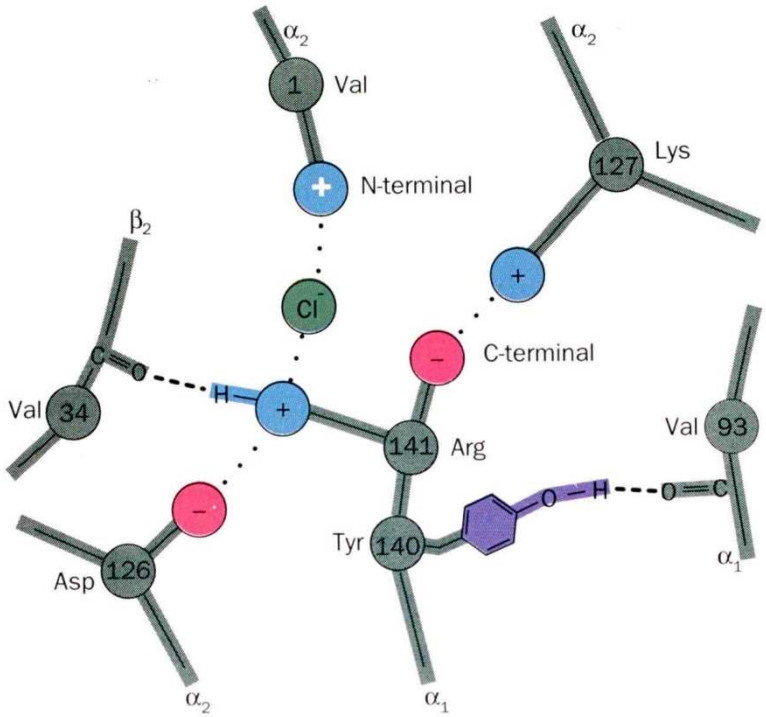
- A major contribution to protein stability originates from the surrounding liquid medium and is of an entropic nature. Therefore, the whole system, protein and solvent, has to be taken into account.

Types of noncovalent interactions important for protein structures

Type	Examples		Binding energy (kcal/mol)	Change of free energy water to ethanol (kcal/mol)
Electrostatic interaction	Salt bridge	—COO $^-$·····N^+H_3—	-5	-1
	Dipole-dipole		+0.3	
Hydrogen bond	Water		-4	
	Protein backbone		-3	
Dispersion forces	Aliphatic hydrogen		-0.03	
Hydrophobic forces	Side chain of Phe			-2.4

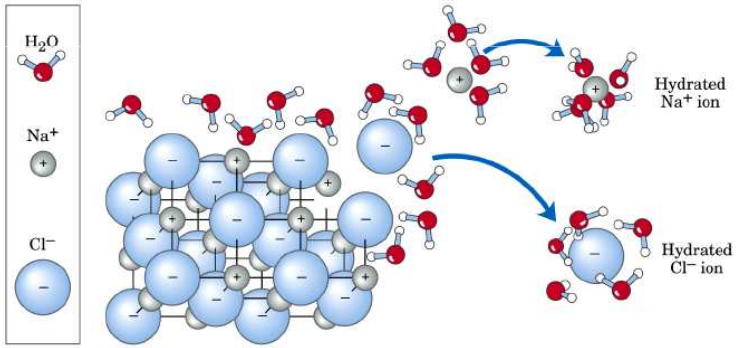
Salt bridges are observed in proteins

- Two ionic protein groups with opposite charges can form an ion pair or salt bridge.
- These groups include:
  - Positively charged: side chains of arginine, lysine and histidine (at neutral pH), the amino terminus of the backbone.
  - Negatively charged: side chains of aspartic acid and glutamic acid, the carboxyl terminus of the backbone.

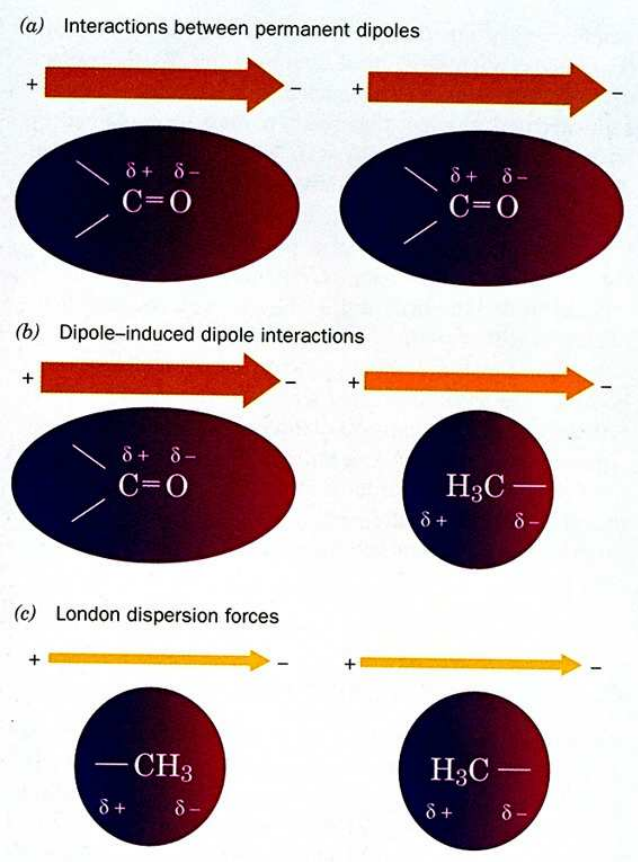


Salt bridges are strong but do not contribute greatly to protein stability

- Free ions in aqueous solution are highly solvated.
- Most of the ion pairs in proteins are located on the surface and tend to be poorly conserved in evolution.
- Buried ion pairs are usually critical for protein functions, such as the catalytic triad Asp-His-Ser observed in serine protease.

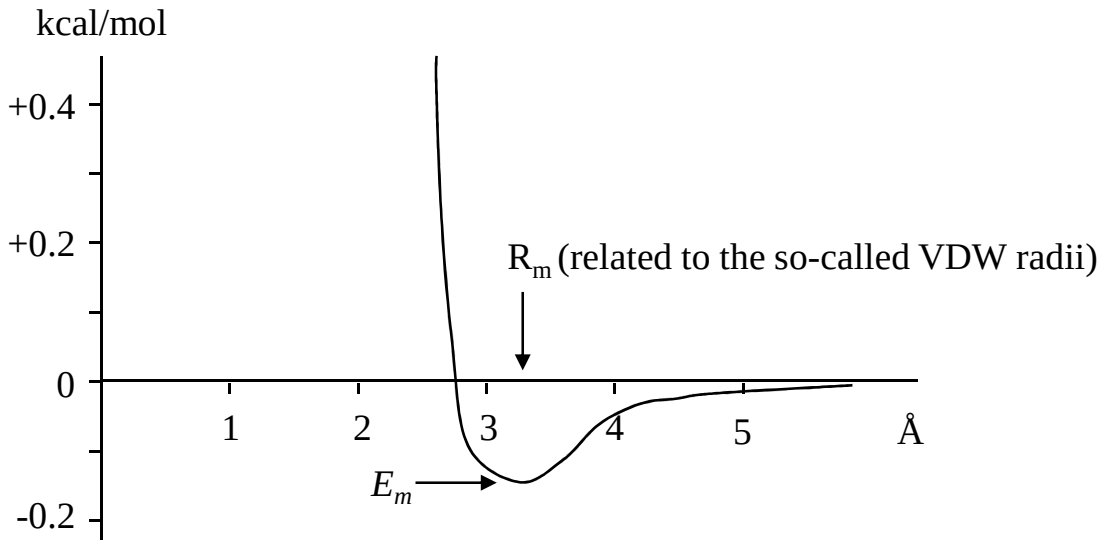


Van der Waals interactions arise from electrostatic interactions among dipoles



- Permanent dipoles arise from uneven distribution of electrons between two covalently-bonded atoms.
- Permanent dipoles can induce a dipole moment in neighboring group to form an attractive interaction.
- London dispersion forces arise from rapid fluctuating motion of electrons, which creates instant small dipole moments.

Dispersion forces are counterbalanced by the repulsion of the electronic shells

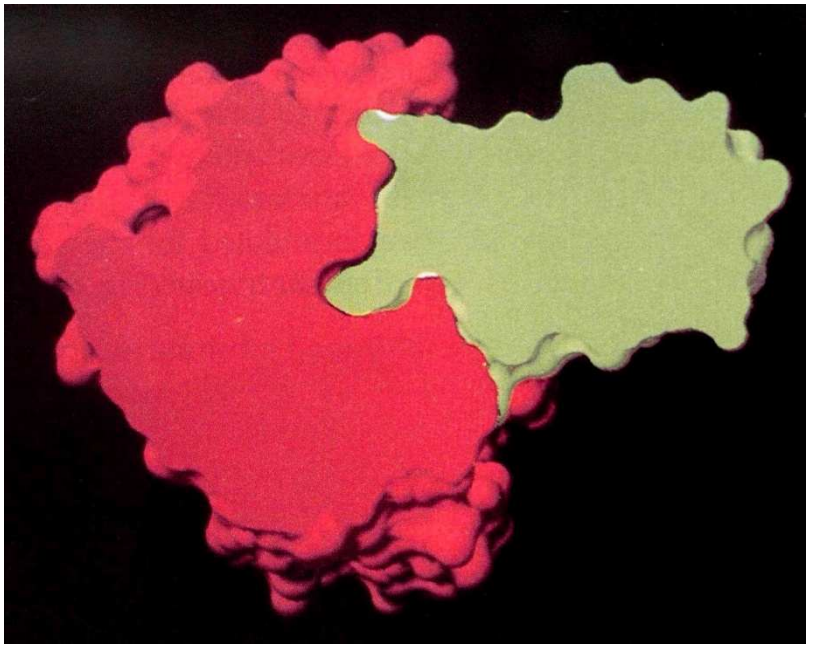


Lennard-Jones 6-12 potential for dispersion forces and electron repulsion

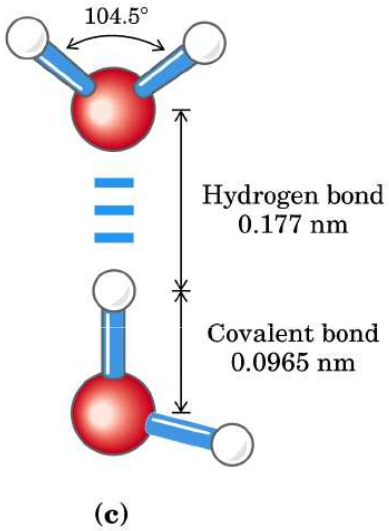
$$E = \frac{A}{R^{12}} - \frac{B}{R^6} = E_m \bullet [- (\frac{R_m}{R})^{12} + 2 (\frac{R_m}{R})^6]$$

Van der Waals interactions are weak but significantly stabilize protein structures

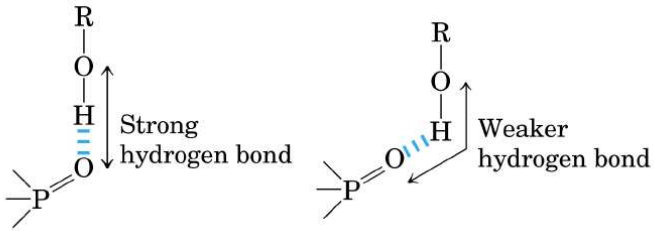
- Van der Waals interactions are weaker than the charge-charge interactions of ion pairs.
- Van der Waals interactions are short-range, local interactions.
- The great numbers of interatomic contacts (surface complementarity) in proteins make Van der Waals a major influence in determining their structures.



Hydrogen bond is a special form of electrostatic interactions



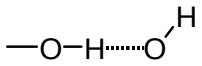
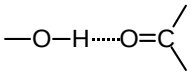
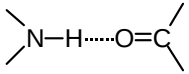
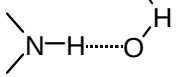
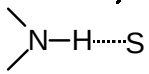
(c)



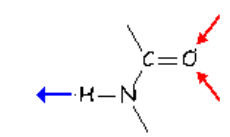
- The strength of hydrogen bond is intermediate between the energy of van der Waals interactions and covalent bonds.

- Hydrogen bond tends to be linear with the donor-hydrogen bond pointing along the acceptor's lone pair orbital. Deviations from the ideal geometry are not unusual.

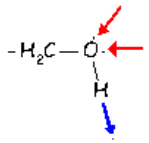
Hydrogen bonds found in proteins

Type of hydrogen bond	Donor-acceptor distance (Å)
Hydroxyl-hydroxyl 	2.8 ± 0.1
Hydroxyl-carbonyl 	2.8 ± 0.1
Amide-carbonyl 	2.9 ± 0.1
Amide-hydroxyl 	2.9 ± 0.1
Amide-imidazole nitrogen 	3.1 ± 0.1
Amide-sulfer 	3.7 ± 0.1

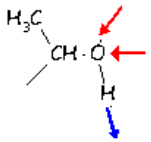
Many side chain groups can participate in forming hydrogen bonds



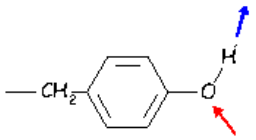
peptide



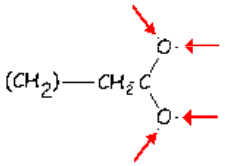
serine



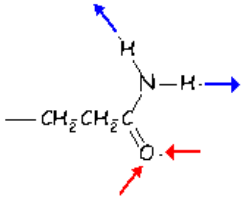
threonine



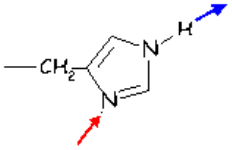
tyrosine



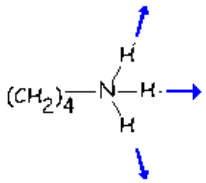
glutamic/aspartic acid



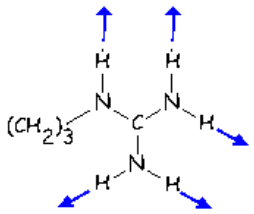
glutamine/asparagine



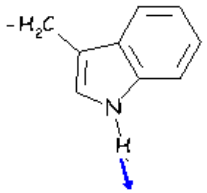
histidine



lysine



arginine



tryptophan

hydrogen bond donor →

hydrogen bond acceptor ←

The role of hydrogen bonds in stabilizing protein structures

- Most potential hydrogen bond donors and acceptors in a protein are involved in hydrogen bonds.

- Internal hydrogen bonding cannot significantly stabilize the structure of a native protein relative to its unfolded state.

- The internal hydrogen bonds of a protein do provide a structural basis for its native folding pattern.

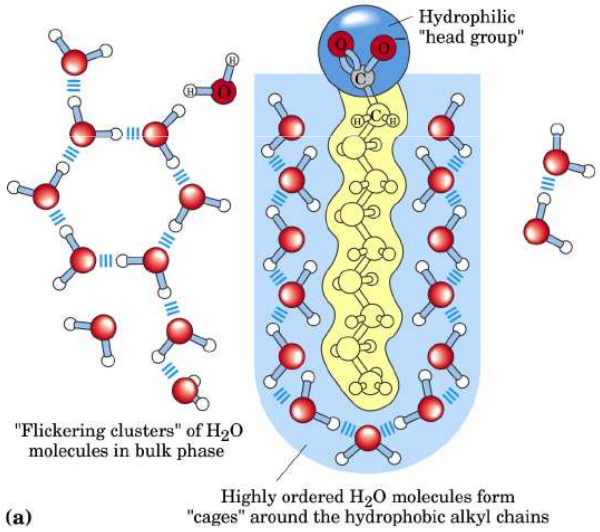
The spontaneity of a process is governed by the change in Gibbs free energy

$$\Delta G = \Delta H - T \bullet \Delta S$$

- The overall change in Gibbs free energy (ΔG) for a spontaneous process is negative.
- Hydrophobic effect: transfer of a nonpolar molecule from water to a nonpolar solvent is a spontaneous process (oil and water do not mix).

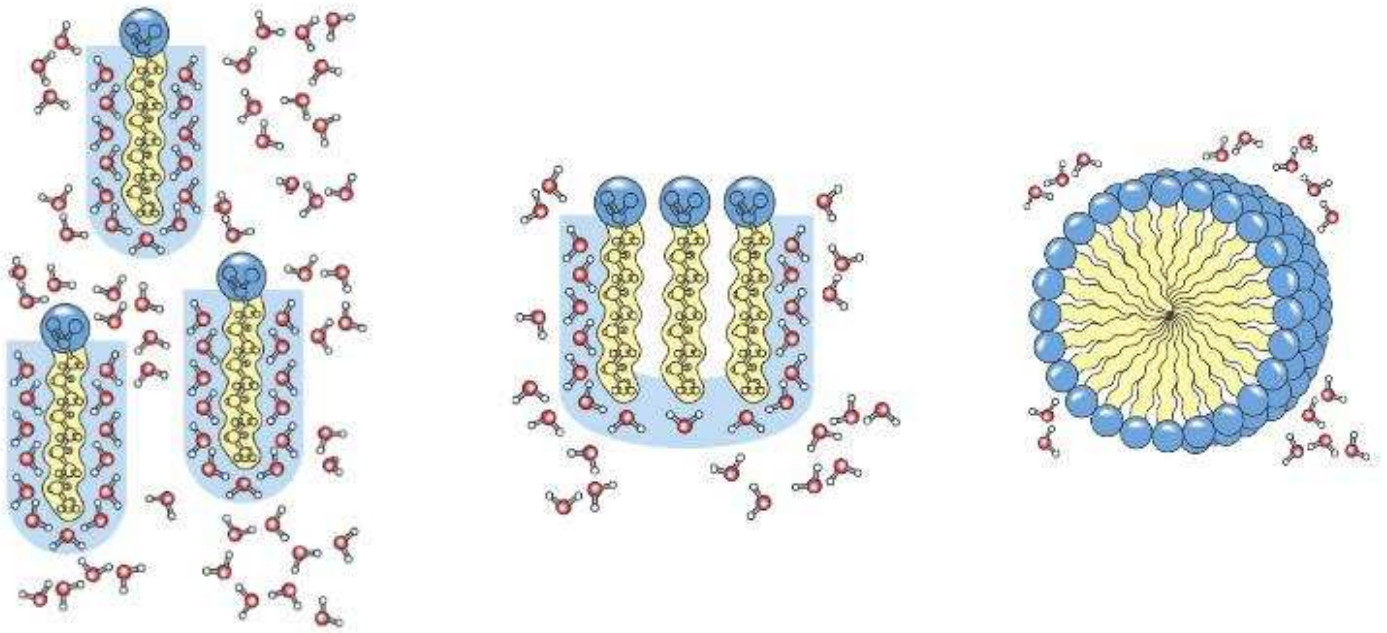
Process	ΔH ($kJ \bullet mol^{-1}$)	$-T \bullet \Delta S$ ($kJ \bullet mol^{-1}$)	ΔG ($kJ \bullet mol^{-1}$)
CH_4 in $H_2O \Leftrightarrow CH_4$ in C_6H_6	11.7	-22.6	-10.9
CH_4 in $H_2O \Leftrightarrow CH_4$ in CCl_4	10.5	-22.6	-12.1
C_2H_6 in $H_2O \Leftrightarrow C_2H_6$ in benzene	9.2	-25.1	-15.9
C_2H_4 in $H_2O \Leftrightarrow C_2H_4$ in benzene	6.7	-18.8	-12.1
C_2H_2 in $H_2O \Leftrightarrow C_2H_2$ in benzene	0.8	-8.8	-8.0
Benzene in $H_2O \Leftrightarrow$ liquid benzene	0.0	-17.2	-17.2
Toluene in $H_2O \Leftrightarrow$ liquid toluene	0.0	-20.0	-20.0

The physical mechanism of hydrophobic effect



- Water molecules form a cagelike structure around the nonpolar molecule.
- The positive ΔH is probably due to the fact that the cage has to be broken to transfer the nonpolar molecule.
- The positive ΔS is probably due to the fact that the water molecules are less ordered (an increase in the degree of disorder) when the cage is broken.

Formation of lipid micelles: burial of hydrophobic tails



Protein folding towards its native state is driven by the hydrophobic effect

- A natural polypeptide chain contains a specific pattern of polar and nonpolar groups.
- In native proteins the majority of nonpolar side chains are removed from the water and assembled in hydrophobic cores.
- Burial of polar backbones in the interior without loss of free energy is achieved only if they form hydrogen bonds.
- Charged groups are either on the surface (common) or in the interior by forming salt bridges.