Lecture 3:
Protein Geometry & Role of Water

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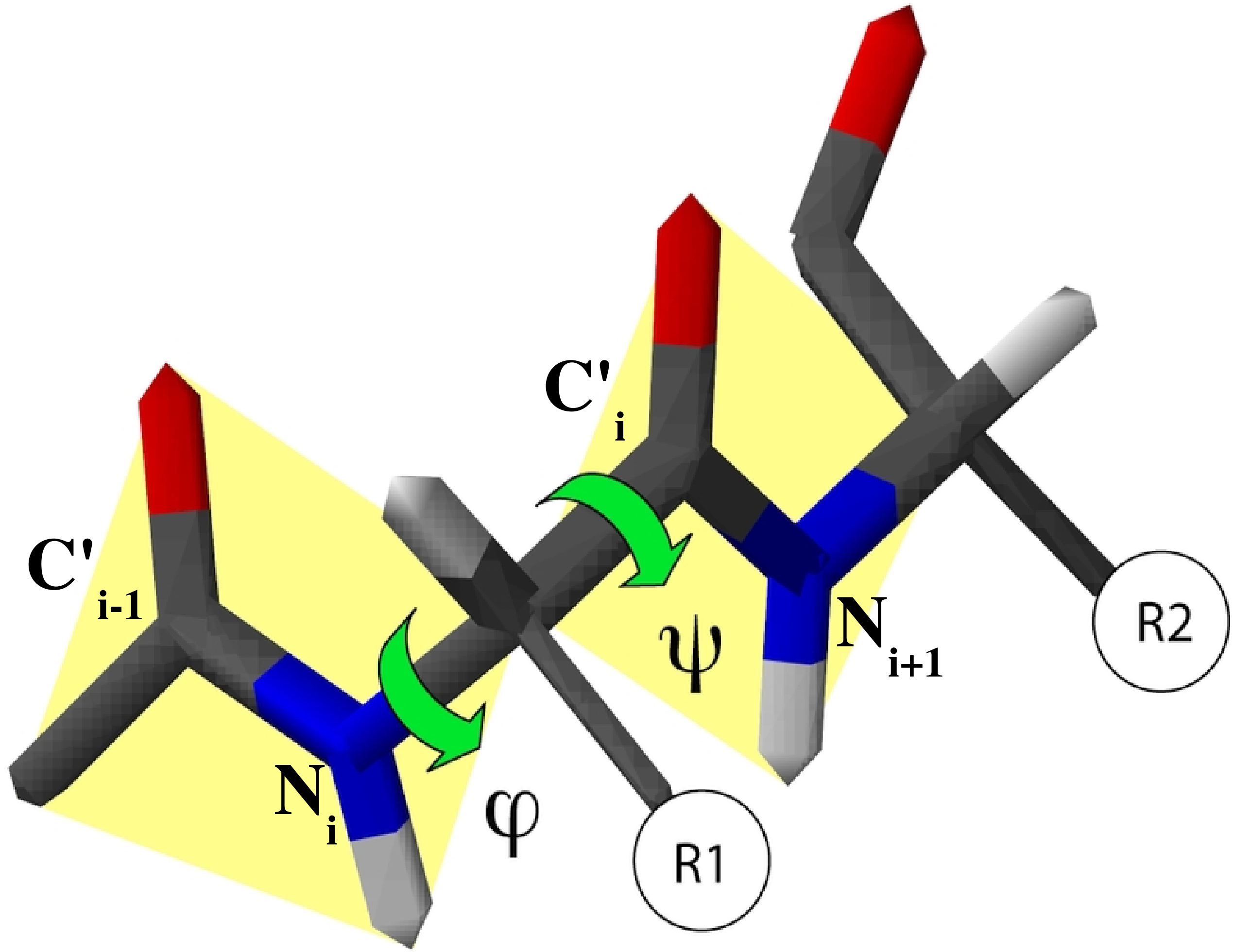
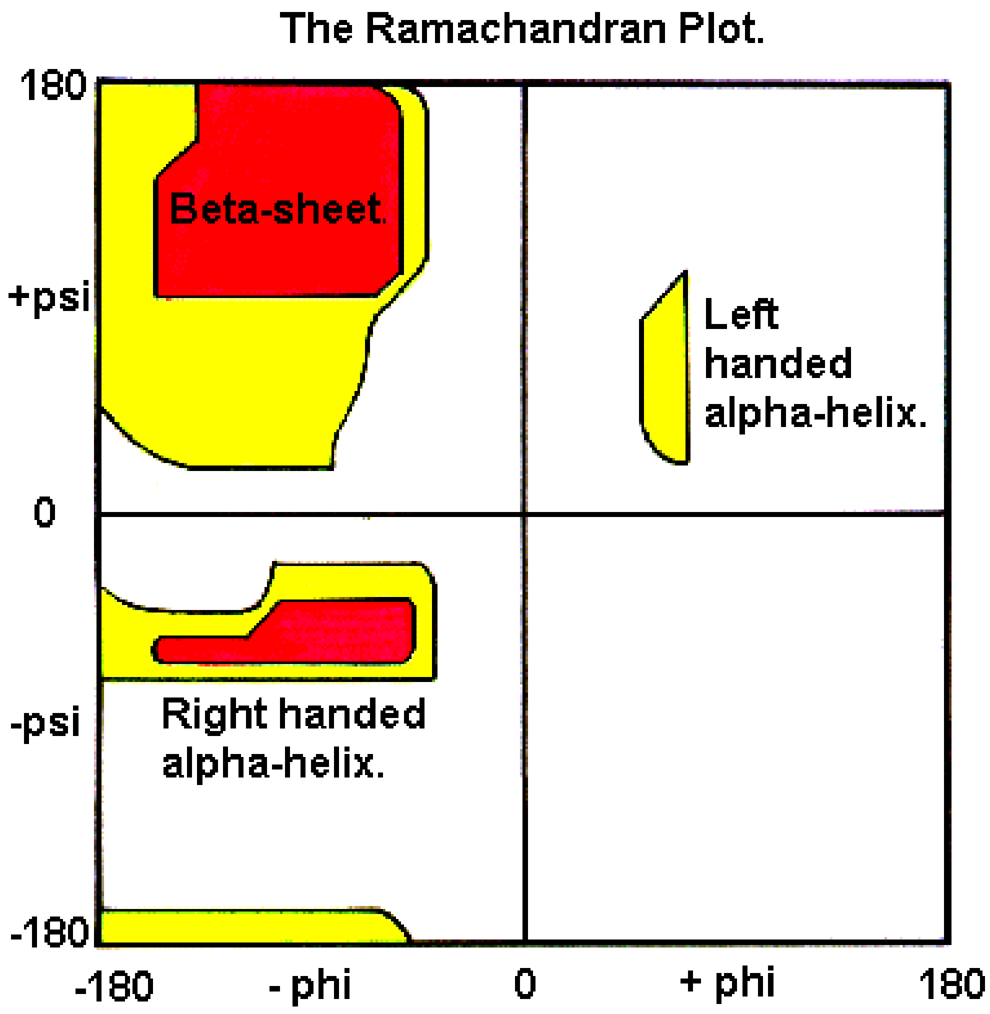
Protein Conformation - Dihedral angles (ϕ, ψ)



$\phi \cdots C'_{i-1} - C'_{i}$ & $\psi \cdots N_{i} - N_{i+1}$

White area on the RP NOT allowed: Why?



$\phi = 0$ … cis conformation for $C'_{i-1} – C'_{i}$

$\psi = 0$ … cis conformation for $N_{i} – N_{i+1}$

$\phi = 0$: distance between C' atoms 2.9 Å

$\psi = 0$: distance between N atoms 2.9 Å

Minimal interparticle distances r_{min} (VdW potential):

r_{min} (C'--C') = 3.0 Å & r_{min} (N--N) = 2.7 Å

↓

Disallowed versus Strained regions (Fig. 3.2. in Textbook)

Additional restrictions due to:

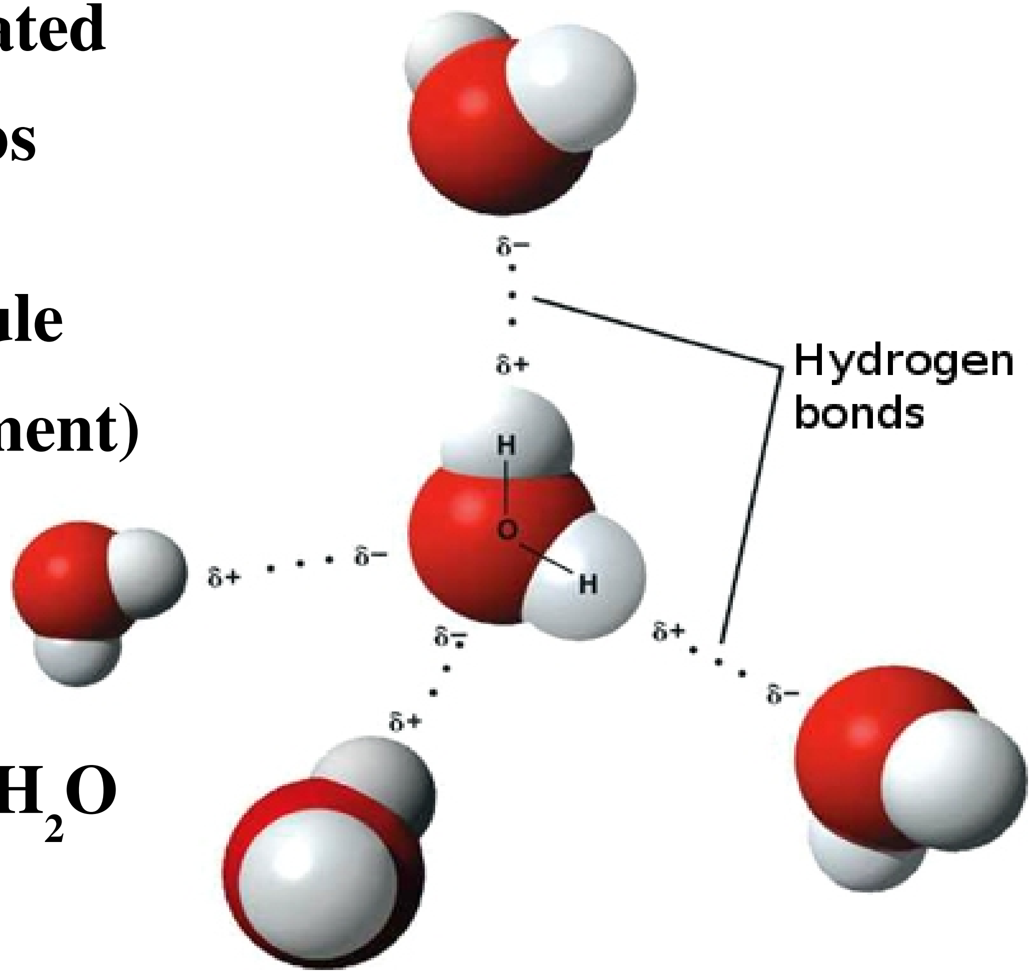
- **C_α and O-atoms attached to N and C';**
- **clashes of C' & N with C_β atoms;**
- **clashes with some C_γ atoms (valine, isoleucine, threonine);**

Outliers:

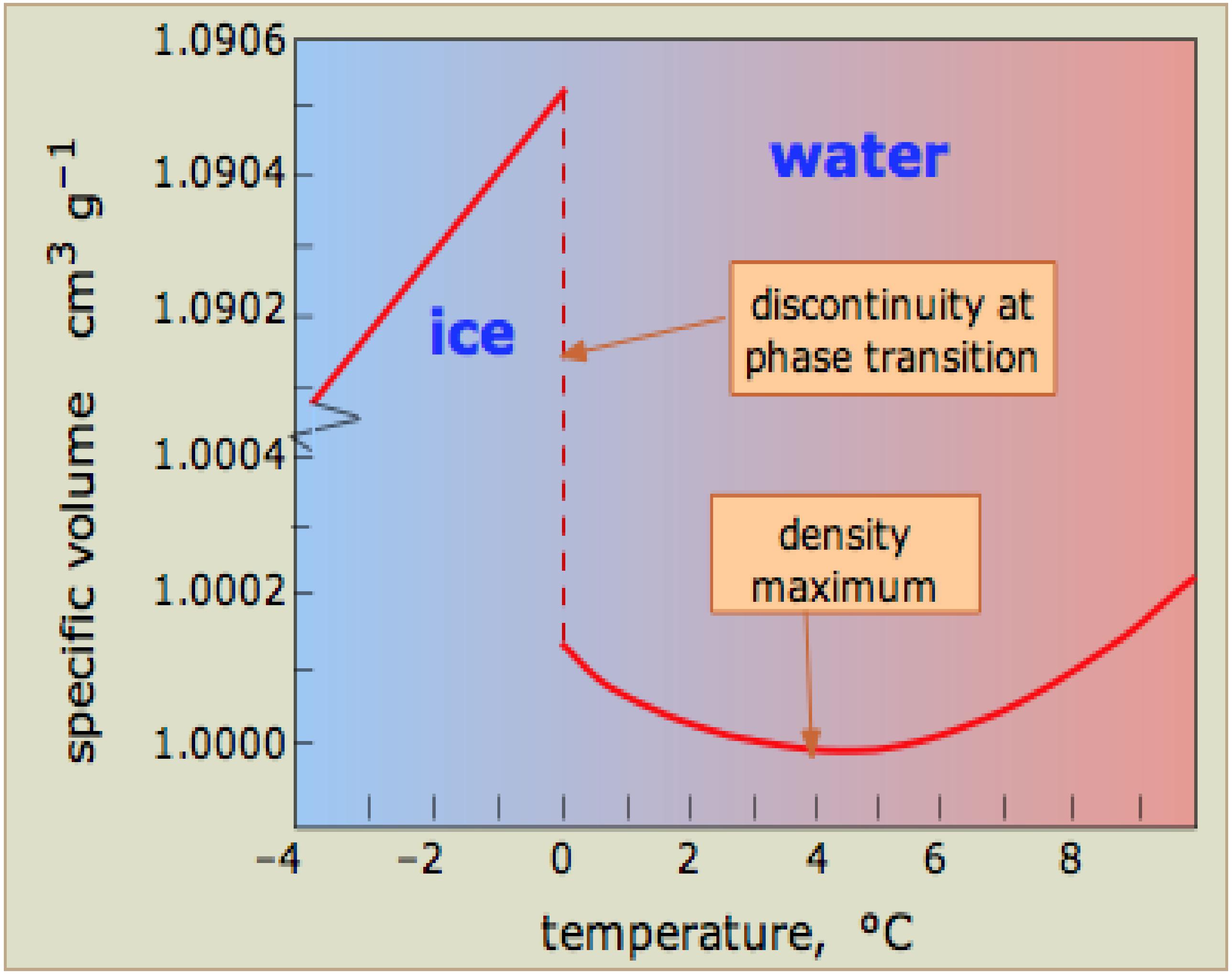
- **glycine (no C_β) → least restrictions: most flexible aa**
- **proline (imino acid): Pro ring created by the C_β atom bonded to the N-group → most restrictions ($\phi \sim -70^o$)**
- **extra restrictions on the aa preceding Pro**

Aqueous Environment – Properties of Water

- H_2O behavior dominated by the presence of Hbs
- H_2O is a polar molecule (electrical dipole moment)
- over 60 anomalies of H_2O presently described



Density maximum of liquid water at 4°C



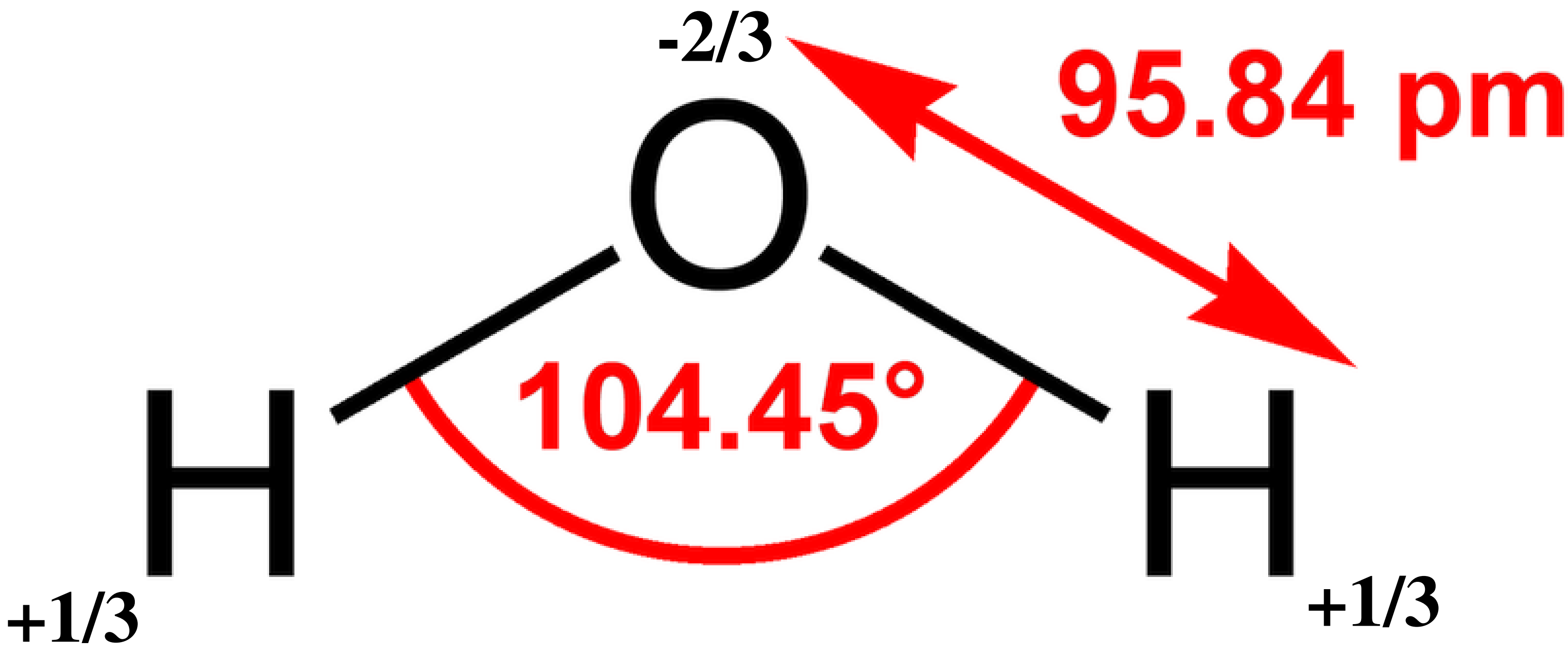
- **H_2O freezes (273 K) and boils (373 K) at abnormally high temperatures relative to**
  - **O_2 (54 K & 90 K)**
  - **H_2 (4 K & 20 K)**

- **extra stability of H_2O structure due to HBs**

- **INTs among electrons in H_2O responsible for HBs**

- **H_2O and D_2O similar melting and boiling temperatures (the mass of the nuclei is irrelevant)**

Partial charges within H_2O molecule:



Estimation of HB energy between H_2O molecules:

- **EIs between two oppositely charged atoms (of unit charges) At a distance of 1 Å**

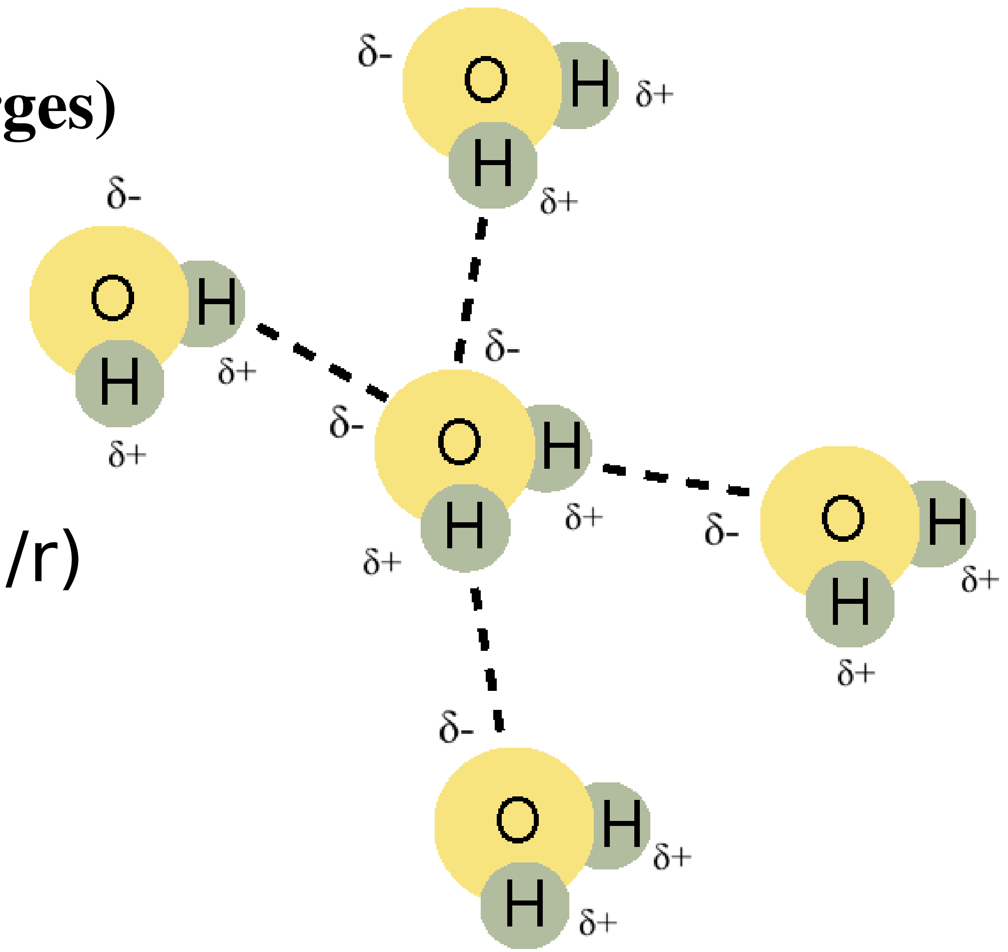
  ~ 330 kcal/mol

- **at a VdW distance ~ 3 Å (1/r)**

  ~ 110 kcal/mol

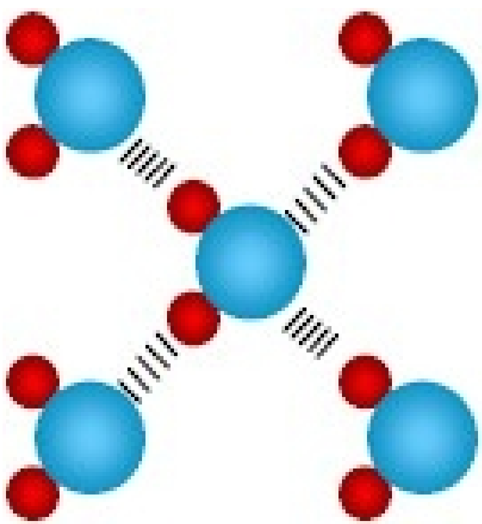
- **Due to partial charges**

  ~ 10 kcal/mol

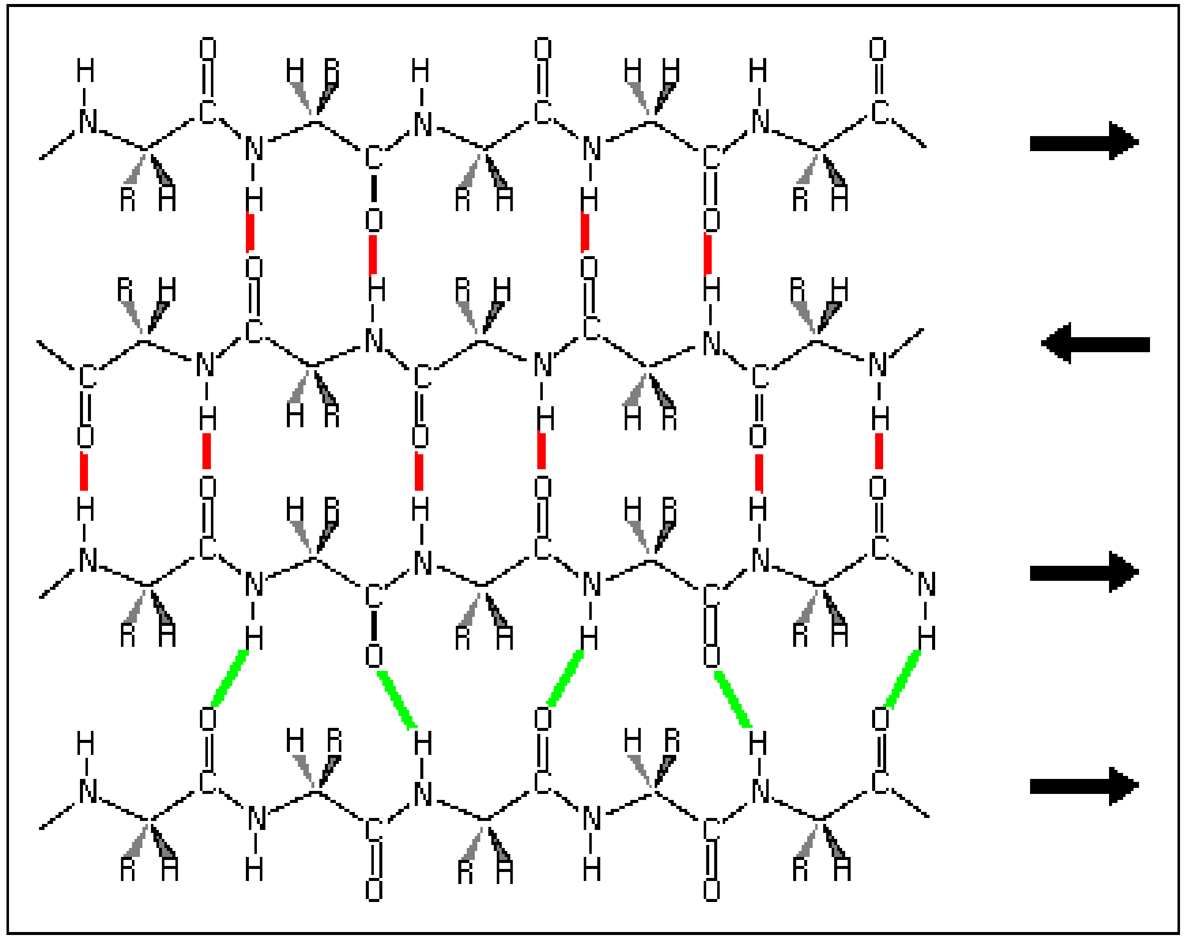


Donors versus Acceptors
(O – H & N – H versus C = O groups)

$$D^{\delta-} - H^{\delta+} :::: A^{\delta-}$$



H_2O



antiparallel vs parallel β-sheet

Orientational sensitivity of the HB:

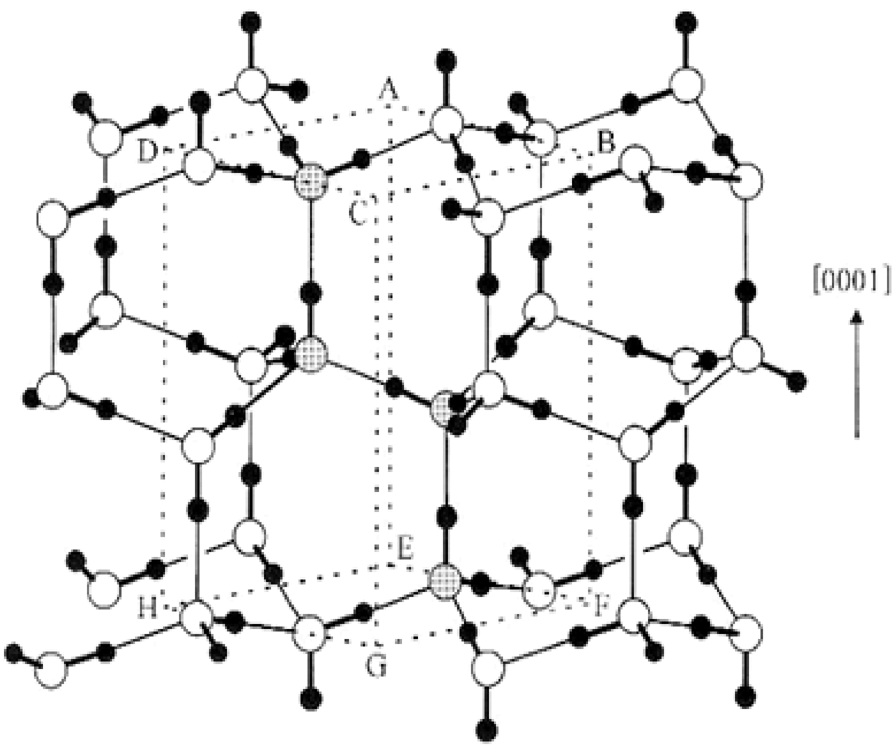
- the valence bond of the donor is directed towards the acceptor within 20-30^{o}
- orientation of the acceptor group is less important
- in H_2O, O-atom can participate as an acceptor of 2 HBs & H-atoms are donors of 1 HB each
- the energy of one HB: 5 kcal/mol (comparison of evaporation heats of similar compounds, one with HBs the other without)

How do HBs affect water properties?

- **Directionality of HBs → ice with open network structure**
- **ice less dense than water**
- **ice melts under strong pressure**

BUT

- **most of HBs existing in ice persist in liquid water**



Evidence:

IR absorption spectrum for O – H groups in ice, CCl_4, And liquid H_2O consistent with existence of loose HBs in liquid H_2O

In liquid water the molecular stretch vibrations shift to higher frequency, on raising the temperature: As HBs weaken, the covalent O-H bonds strengthen causing them to vibrate at higher frequencies.

Partial charges of protein backbone groups:

C' = O		Ca	N — H	
+2/5	-2/5	0	-1/5	+1/5

⬇

C' = O & N — H participate in HB formation

acceptor (↑ C' = O)   donor (↑ N — H)   → **2 backbone HBs per aa**

Polar side chain groups also form HBs.

E_{HB} = 5 kcal/mol versus E_T = 0.6 kcal/mol

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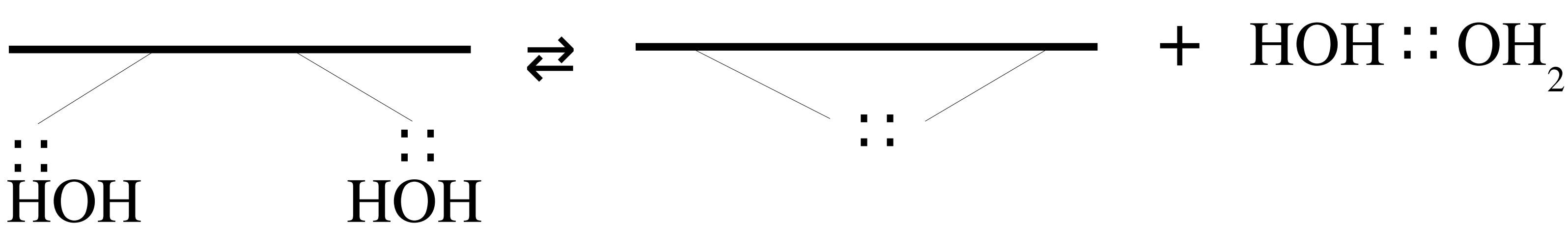
- **HBs mostly preserved by thermal motion/fluctuations**
- **HBs form between the protein and H_2O → this reaction has the energy balance close to zero:**



- **the entropy associated with "free" H_2O molecules increases (like in transition from ice to liquid H_2O)**

Entropy estimate upon formation of one protein HB:

Melting point for ice → liquid water at 0℃:

$$\Delta F = 0 \rightleftarrows \Delta E = T\,\Delta S$$

$$\Delta E = 80\ \text{cal/g} = 80\ \text{cal/g} \times 18\text{g/mol} = 1.44\ \text{kcal/mol}$$

⬇

The free energy of the protein in water decreases by ~ 1.5 kcal/mol upon formation of one HB and is almost entirely compensated by the loss of protein conformational entropy.

Entropy & Free Energy

➔A molecule M can be in one of the 2 states, a & b (a … at sea level, b … 5 km above the sea level). What is the probability for M to be in a or b, p_a versus p_b, at a constant temperature after a long time (M needs to be able to reach a and b)?

➔ probability of M being in a state of energy E:

$$\sim \exp(-E/k_B T) \quad \text{[Boltzmann Eq.]}$$

➔ $p_a : p_b$ .... $\exp(-E_a/k_B T) : \exp(-E_b/k_B T)$

- $E_a < E_b$ (gravity) → M will be mostly in state a (and 1.5-2 times less time in state b

- the above result only correct for equal volumes (e.g. lungs)

- the number of states visited by M is proportional to the available volume V_a or V_b

$$p_a : p_b = V_a \exp(-E_a/k_BT) : V_b \exp(-E_b/k_BT)$$

$$p_a : p_b = \exp(-E_a/k_BT + \ln V_a) : \exp(-E_b/k_BT + \ln V_b) =$$
$$[\exp(-(E_a - T\,k_B \ln V_a)/k_BT)] : [\exp(-(E_b - T\,k_B \ln V_b)/k_BT)]$$

Definition of Entropy and Free Energy:

$S = k_B \ln V$ → entropy of M in volume V

$F = E - T S$ → free energy of M

⬇

$$p_a : p_b = \exp(-F_a / k_B T) : \exp(-F_b / k_B T)$$

The most stable state of the system is that with the lowest free energy F.

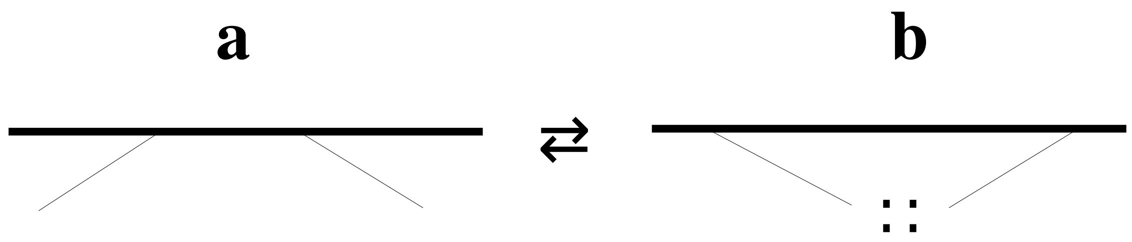
The energy change upon a → b, $E_b - E_a$, is the work required to transfer the body (M) from a to b, when there is no heat exchange with environment.

The free energy change upon a → b, $F_b - F_a$, is the work required to transfer the body (M) from a to b, when the body exchanges heat with environment.

Free energy of the HB formation in H_2O and protein:

- **$E_{HB} < 0$: hydrogen bond energy**
- **$S_{HB} > 0$: entropy of movements & rotations**
- **HBs are stable when $E_{HB} - T\,S_{HB} < 0$ ($\Delta F < 0$)**

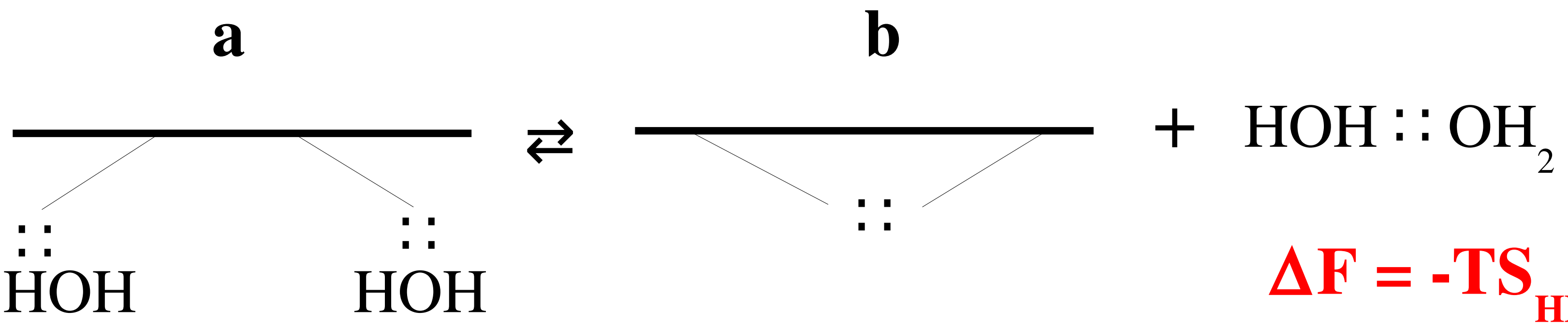
vacuum:



$\Delta F = E_{HB} < 0$

$E_a = 0$ & $S_a = 0$ $\quad$ $E_b = E_{HB}$ & $S_b = 0$

water:



$\Delta F = -TS_{HB} < 0$

$E_a = 2E_{HB}$ & $S_a = 0$ $\quad$ $E_b = 2E_{HB}$ & $S_b = S_{HB}$