BCHM 463 GSS - 09/19/2022

Protein Structure and Thermodynamics

(1.)

a.) Are peptide bonds generally trans-isomerized or cis-isomerized? Why?

Peptide bonds are generally **trans-isomerized.** The R-groups at the ends of the cis-isomerized peptide bonds tend to have steric clashes which destabilize the overall peptide structure and is thus considered to be unfavorable.

b.) Are there any amino acids which are an exception to this rule?

Proline exists in the trans-isomerization or the cis-isomerization in about a 50/50 mix. Neither state is especially preferred due to proline's unique molecular geometry - it is the only amino acid which connects with the amine group of its own amino acid, thus less steric hindrance is present in the cis-conformation.

(2.) Protein folding is heavily dependent upon the amino acid interactions both within the protein itself and the protein's environment.

a.) Using your knowledge of the types of interactions discussed in class, rank the importance of different types of interactions in determining the tertiary and quaternary structure of a protein.

In order of importance:
1 - Induced Dipole Interactions
2 - Dipole-Dipole Interactions and Hydrogen Bonding
3 - Ionic Interactions and Covalent Bonds

b.) Why is the weakest interaction between molecules actually the strongest determinant of tertiary and quaternary structure in proteins?

Protein folding for tertiary and quaternary structures is dependent solely on the change in the system's entropy. Thus, the largest determinant of entropy in physiological systems is the hydrophobic effect - regions within the protein which have largely hydrophobic residues will tend to sequester themselves away from the outside and coalesce at the center of the protein, resulting in the formation of tertiary and quaternary protein structures that depend heavily on the hydrophobic or hydrophilic characteristics of amino acid residues.

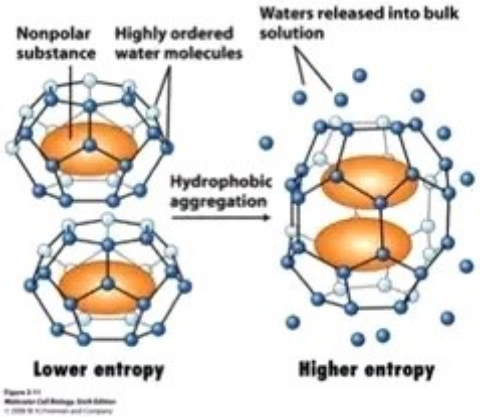
c.) Suppose you had a small protein with the sequence, N' - MLIIFGAAVPAALFGPIALILFA - C'. How would you expect this protein to behave in an environment like human blood?

This protein is notable in that it is constituted of exclusively nonpolar amino acids. There is not a single hydrophilic residue in this sequence, and as a result, this protein would fail to fold in an aqueous environment such as human blood. Furthermore, due to hydrophilic separation, it is likely that adding multiple proteins of this kind in the blood would result in clumping and aggregation of the nonpolar protein in the bloodstream, which would not be ideal for the organism.

(3.) You isolate a small transmembrane protein which associates with the phospholipid bilayer of animal cells. This protein is anchored to the membrane through a polypeptide sequence N' - RSTYKRVIH - C'. Would this anchor sequence most likely be present *within* the phospholipid bilayer or on the *outer-facing* surfaces of the membrane? Explain how you arrived at your conclusion.

There are some nonpolar residues in this sequence, but the sequence primarily consists of charged or polar amino acid residues (such as R, S, T, Y, etc...). It would not be entropically favorable for this anchor sequence to be found within the hydrophobic environment of the phospholipid bilayer. Thus, it is most likely that this protein anchor is found outside of the phospholipid membrane, facing either the cytoplasmic side or the extracellular side.

(**4.)** Briefly explain the entropic contribution to the hydrophobic effect. Illustrating your answer may help with answering this question.



Hydrophobic substances in solution restrict movement of water molecules around the substance, forming what is known as a structured "water cage" around the nonpolar surface, decreasing the entropy of the system. When hydrophic substances aggregate, the overall surface area of restricted movement is lowered, increasing the overall entropy of the system. Furthermore, the release of waters between hydrophobic substances during hydrophobic aggregation additionally confers entropy to the system.

(5.) What is the difference between ΔG and $\Delta G^{o'}$? In differentiating between the two, also describe their relationship in terms of Q and K_{eq}.

$\Delta G^{o'}$ describes the reaction at equilibrium, whereas ΔG describes the energy being exchanged during a reaction. Since $\Delta G^{o'} = -RT\ln K$ and $\Delta G = \Delta G^{o'} + RT\ln Q$, we see that ΔG, which is directly dependent on $\Delta G^{o'}$, may be simplified to the expression: $\Delta G = \Delta G^{o'} + RT\ln Q = -RT\ln K + RT\ln Q = -RT(\ln K - \ln Q)$ since we assume T is constant in biological systems. Thus, it follows that when $K > Q$, ΔG is necessarily (-) since $\ln K - \ln Q$ would result in a positive value, and when $K < Q$, ΔG is necessarily (+) since $\ln K - \ln Q$ would result in a negative value. When $K = Q$, $\Delta G = \Delta G^{o'}$, and the system is in equilibrium. This is generally bad for the organism, as reaching equilibrium in biochemistry typically means death.

(6.) A 1M HCl solution is dissolved in water at 25°C and mixed. The enthalpy change of the water-HCl system is $\Delta H = -74.84$ kJ/mol.

a.) Is this process endothermic or exothermic?

ΔH is negative. The process is exothermic.

b.) Overall, would there be an increase or a decrease in the entropy of the system?

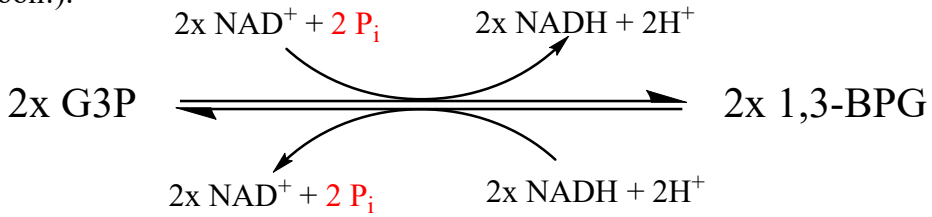
Generally, the solvation of a solute in water results in an increase in entropy of the system (positive ΔS). The fact that the system is mixed would also result in an increase in the entropy of the system.

c.) What does this say about the spontaneity of HCl dissolution in water with mixing? Explain mathematically.

$\Delta G = \Delta H - T\Delta S$. In this case, the change in enthalpy is negative and the change in entropy is positive. Thus, ΔG is necessarily negative (there is no way for ΔG to be positive). Therefore, the dissolution of HCl in water at 298 K is a spontaneous process.

(7.) Patients suffering refeeding syndrome, a metabolic disturbance caused by the sudden reinstitution of nutrients after a long period of starvation, may experience hypophosphatemia (low concentration of phosphate compounds in blood), a potentially deadly complication that interrupts hundreds of metabolic pathways dependent on normal phosphate concentrations.

a.) One key phosphate-dependent step in metabolism is the sixth step of glycolysis (you will learn more about this process very soon!):



Under standard physiological conditions, $\Delta G^{o'} = +6.3$ kJ/mol, and ΔG is between -2 kJ.mol and +2 kJ/mol. How would a decrease in $[P_i]$ (phosphate concentration) affect the spontaneity of this reaction, if at all? Explain mathematically, using the relationship between $\Delta G^{o'}$ and ΔG.

First, we get the equilibrium constant expression: $([NADH][H^+][1,3\text{-BPG}]) / ([NAD^+][P_i][G3P])$. A decrease in $[P_i]$ would result in $Q > K$, shifting the reaction to the left (You will soon find out that this is not the path that glycolysis takes!).

$\Delta G = -RT(\ln K - \ln Q)$, since $Q > K$, $\ln Q > \ln K$ and $\ln K - \ln Q$ will be negative. Thus, ΔG will be a positive value, and the forward reaction (G3P to 1,3-BPG) will become thermodynamically unfavorable.