Problem Set No. 1

Due: Thursday, 10/07/10 at the start of class

Objective: To understand the components and basic structure of cells, and to develop a picture of the molecular-scale interactions relevant to biological processes.

Review problems

You should pay special attention to these questions after reading. Note that the answers are given in the back of the book. Formulate your answers fully first and then check them. This can be a significant aid in your understanding of the material, and similar questions may be asked on the final. You do not need to provide written answers in the solutions you hand in.

- ECB 1-9
- ECB 1-11
- ECB 1-16
- ECB 2-17
- ECB 2-19
- ECB 3-11

Problem 1

Draw the chemical structure of a small fragment of DNA, a dinucleotide of thymine and guanine, whose sequence is abbreviated TG. Be sure to show all double bonds and formal charges. Be sure your sequence is not reversed.

Problem 2

Consider that the human genome (i.e., the total DNA content containing genetic information) consists of roughly 3 billion nucleotide pairs. When two unrelated human individuals are compared, roughly 0.1% of these nucleotides varies. The variable part of the DNA sequence gives rise to genetic diversity, while the remaining common 99.9% is what makes each individual distinctly human. Consider a random individual to whom you are not related. What is the probability that your genome is identical to theirs? Express your answer as 1 in 10^x. Recall that four bases are possible at each nucleotide in a DNA sequence.

Problem 3

In the largely aqueous environment of the cell, complementary DNA strands spontaneously self-assemble to form the iconic so-called "B" helical structure that is so familiar in popular science. At first glance, this may seem counterintuitive because the phosphate-deoxyribose backbone contains negative charges that would result in repulsive like-charge interactions between the two strands.

(a) The human genome consists of roughly 3 billion base-paired nucleotides. In eukaryotes, DNA exists inside of the cell nucleus, and a typical mammalian nucleus is spherical with a diameter of about 6 microns. What typical total concentration of positively-charged, monovalent species (e.g., Na+ or K+) must be present inside the nuclei of human cells in order to maintain an overall charge-neutral environment there? Express your answer in millimolar. Compare it to the value of ~150 mM often found in the cytosol.

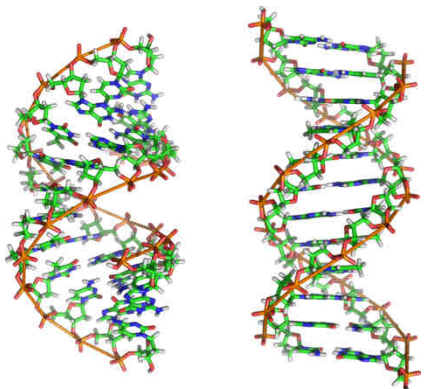
(b) In the B helical structure, negative charges on strand backbones opposite of each other are separated by about 12 Å in distance. Compute the potential energy of this interaction in kcal/mol assuming that the charges interact in a vacuum through Coulomb's law,

$$u(r) = \frac{q_1 q_2}{4\pi\epsilon\epsilon_0 r}$$

where $\epsilon = 1$ for a vacuum. Compare this to the average strength of a hydrogen bond in water, roughly 2 kcal/mol.

(c) Repeat part (b) in the case that the charges interact in water, where $\epsilon = 80$. How does water help stabilize the double helix structure?

(d) In dehydrated *in vitro* DNA samples, one often finds that DNA forms an alternate, wider "A" helical structure (see picture below). In this structure, negative charges on strand backbones opposite of each other are separated by about 19 Å. Is this consistent with your calculations in (b) and (c)?



A DNA B DNA

Problem 4

A large number of proteins are embedded in the phospholipid bilayer that forms cell membrane. Inside this bilayer, molecules see a highly hydrophobic environment formed by the hydrocarbon tails and with little water present. On the extracellular surface of the bilayer, molecules see a highly polar environment due to the presence of hydrophilic phospholipid head groups that are well-hydrated by water. Consider a very small protein—a peptide—that has reached a extracellular surface and comes to equilibrium. Assume that it can then either occupy one of two states: remaining associated at the extracellular interface (I), or inserting into the hydrophobic bilayer (B).

(a) In class, we discussed the Boltzmann law for the population of molecular states of a system,

$$\wp(\text{state}) \propto e^{-\frac{G_{\text{state}}}{k_B T}}$$

where $G_{\text{state}}$is the free energy of the state of interest. Using this expression, show that the fraction of time that the peptide will be inserted in the bilayer is given by

$$\wp(\text{B}) = \left[1 + e^{\Delta G/k_B T}\right]^{-1}$$

where ΔG relates to a free energy difference between the two states I and B. Indicate whether $\Delta G = G_I - G_B$ or $\Delta G = G_B - G_I$.

(b) An approximate change in free energy for moving a single glutamine amino acid from the bilayer interface to a hydrocarbon environment is 0.2 kcal/mol. Assume that a homopeptide made of N glutamines then has a transfer free energy of $N \times (0.2\ \text{kcal/mol})$. What fraction of the time will a *hexa*-glutamine peptide be inserted in the bilayer? What for a *deca*-glutamine peptide? Assume the temperature is 300 K.

(c) Repeat part (b) for homopeptides made of phenylalanine, for which the transfer free energy per amino acid is approximately -0.6 kcal/mol. Which amino acid type is more hydrophobic?

(d) In general, transfer free energies can measure the favorability of moving a molecule from a polar to a hydrophobic environment, and these can serve to measure how "hydrophobic" the molecules themselves are. Such measurements can also be made at many different temperatures. For highly hydrophobic molecules, it is often found that the transfer free energy varies significantly with temperature around 300 K. What do you think is the larger contribution to ΔG in this case: the entropic or enthalpic component?

Problem 5

(a) A system exhibits a heat capacity C_P that is approximately constant with temperature. Using fundamental thermodynamic definitions, show that at fixed pressure the following relations then apply:

$$S = S_0 + C_P \ln\left(\frac{T}{T_0}\right)$$

$$H = H_0 + C_P(T - T_0)$$

where T_0 is a reference temperature at which the entropy (S_0) and enthalpy (H_0) are known. Assume quantities are on a per-mol basis.

(b) Proteins can generally exist in two different states: an unfolded state consisting of many unstructured conformations (e.g., the protein "flops" around a lot) and a folded state, a highly-ordered and stable three-dimensional structure. Typically, as one increases the temperature, a protein transitions from folded to unfolded states. The quantity that determines which state will be dominant at any one temperature is the Gibbs free energy.

Using the constant heat capacity approximation shown in part (a), find an expression for $\Delta G_{\text{fold}} = G_{\text{folded}} - G_{\text{unfolded}}$. Show that this can be written as

$$\begin{aligned}\Delta G_{\text{fold}} &= G_{\text{folded}} - G_{\text{unfolded}} \\ &= \Delta H_{\text{fold}} - T\Delta S_{\text{fold}} \\ &= \Delta H_{\text{fold},T_f}\left(1 - \frac{T}{T_f}\right) + \Delta C_P\left[T - T_f - T\ln\left(\frac{T}{T_f}\right)\right]\end{aligned}$$

where

$$\Delta H_{\text{fold},T_f} = (H_{\text{folded}} - H_{\text{unfolded}})_{T_f}$$

$$\Delta C_P = C_{P,\text{folded}} - C_{P,\text{unfolded}}$$

and T_f is the folding temperature, i.e., the temperature at which the protein switches from folded to unfolded. Hint: write separate expressions for G_{folded} and G_{unfolded} using the models above and then take their difference. Let the reference temperature $T_0 = T_f$. What must be the value of ΔG_{fold} at T_f?

(c) Show that the ratio of the probability of finding a protein in the folded versus unfolded states is given by

$$\frac{\wp(\text{folded})}{\wp(\text{unfolded})} = e^{\frac{-\Delta G_{\text{fold}}}{RT}}$$

(d) Experiments can measure $f \equiv \wp(\text{folded})/\wp(\text{unfolded})$ as a function of temperature T, where f is the fraction of molecules of a particular protein in solution that are folded. If the heat capacities of the unfolded and folded states are roughly equivalent, what would you plot in order to estimate $\Delta H_{\text{fold},T_f}$?

Problem 6

Avidin is a protein found in avian and reptile eggs that binds the small molecule biotin (vitamin H) with an extremely high affinity. This is one of the strongest known noncovalent interactions, and the avidin-biotin system is a major bioengineering tool. For example, solutions of protein can be separated by tagging the target molecule with biotin during synthesis and then passing over an avidin-coated solid surface, where they bind and become immobilized. The interaction can be described by the following equation:

$$A + B \underset{K_D}{\leftrightarrow} AB$$

Here, the equilibrium constant K_D is defined by the equilibrium molar concentrations as

$$K_D = \frac{[A][B]}{[AB]} \approx 10^{-15}\ \mathrm{mol/L}$$

(a) An equimolar solution of avidin and biotin is prepared, each with *total* molar concentration c_0. At what c_0 will exactly half of the biotin be bound to avidin?

(b) Consider that a cell contains total concentrations of avidin and biotin each of the value c_0 that you found in part (a). If the cell is spherical and roughly 100 microns in diameter, how many *molecules* each of avidin and biotin are there? What does this mean if only one of each is present in the cell? (Perhaps this attests to the strength of the interaction).

(c) Lets say that you are able to prepare a series of different solutions as in part (a) for different values c_0, e.g., by titration. You might then be able to measure the fraction of biotin bound to avidin, $x = [AB]/([B] + [AB])$, by calorimetric or other means. How would you plot quantities involving x and c_0 such that they gave you a linear relation with slope K_D?

Problem 7

Many Nobel prizes have gone to scientists that have elucidated the structure and function of important biomolecules, or who have engineered novel biomolecules that have had a profound impact on biology and biotechnology. In a few (3-5) sentences, describe an important biomolecule that has been at the center of a recent Nobel prize, say in the past 10 years. Be sure to indicate to whom the prize went, what the basic function of the molecule is, and why the discovery has been important. Be sure to use correct grammar & spelling, and clear sentence structure.

Problem 8

Did you have an internship or research experience this summer? If so, and if you are willing to share information related to it, please let us know (a) the company or organization and its location, (b) the topic or title of the project, and (c) the name and contact information (email or phone) of the person from whom you were able to obtain the internship.