Biol 591 Introduction to Bioinformatics (Fall 2002): Problem Set IntroM

Note: *Multiple choice questions in these problem sets and on the exams may have multiple answers or none at all. Choose all answers that apply. Choices may be ambiguous. In such cases, make what you think is the most reasonable interpretation, and be prepared to justify it.*

Questions relating to **What is a cell? How does it work?**

PIM.1. Which of the following are hydrophobic? Hydrophilic? Amphipathic?

A. vinegar
B. skin
C. tooth paste
D. sugar
E. wax
F. rabid dogs

PIM.2. In general, hydrophilic molecules have a difficult time passing cell membranes unless the cell makes accommodations for them. Presuming there are no such accommodations, which of the following molecules would not easily get into a cell?

A. sodium
B. sugar
C. ethanol
D. amino acids

PIM.3. Consider that at an air-water interface, amphipathic molecules expose their hydrophobic surface to air. Draw a picture of what a soap bubble might look like at the molecular level, using a long-sticked popsicle to represent a molecule of soap.

PIM.4. Many local anesthetics, like procaine, are positively charged amphipathic molecules that anesthetize by acting on cell membranes. How do you suppose they fit into a membrane? (Draw a picture)

PIM.5. Imagine building a house, not in the conventional way, but the way DNA builds a cell. How would you do it?

Questions relating to **Proteins: What are they? What do they do?**

PIM.6. One common way of assaying for the presence of protein in a solution is to measure the ability of the solution to absorb medium wavelength UV light. Since absorption at these wavelengths is due to aromatic structures (rings with double bonds), which amino acids do you think are responsible for the absorbance?

PIM.7. Proteins that bind to DNA are typically rich in arginines and lysines? Why?

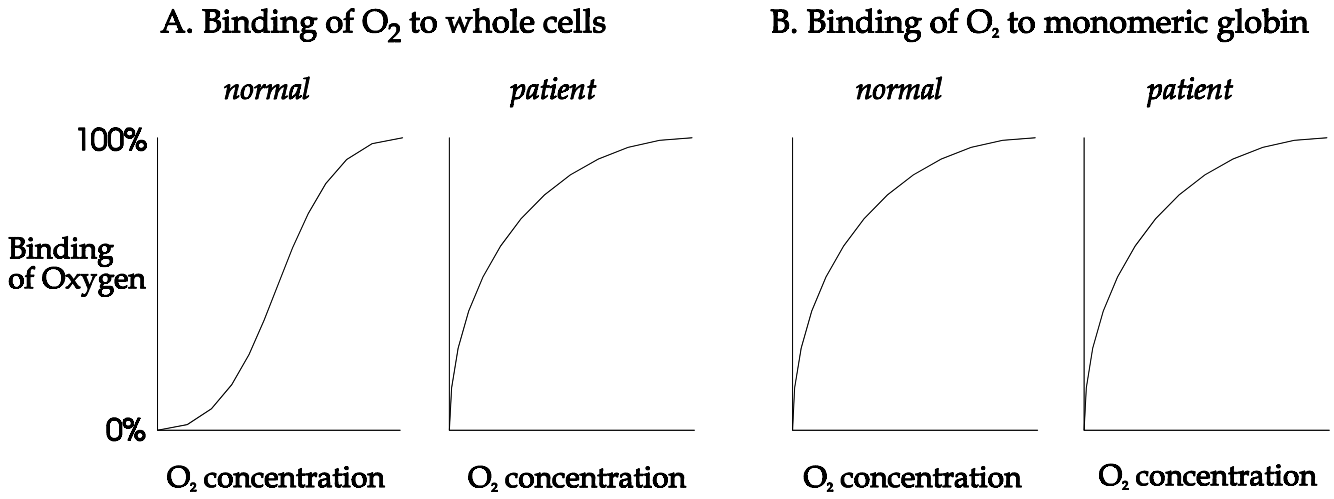
PIM.8. Some antibiotics form rings that stack and create a pore through the membrane. Consider a cyclic polypeptide antibiotic composed of the four amino acids: serine, glycine, threonine, and alanine. If each atom of the backbone is about 2 angstroms in length, estimate the circumference of the pore (presume it to be a circle) and the diameter of a molecule that could fit through it. Approximate the circumference ($\pi \cdot$diameter) to be $3 \cdot$diameter. (Show work)

PIM.9. Peptide hormones can be as small as five amino acids in length, but most protein have polypeptide chains ranging from 70 to 1000 amino acids. Suppose you have some reason to believe that a small protein, precisely 100 amino acids in length, can transform glucose into gold. You set out to synthesize every possible 100-amino acid protein until you find the one you want. How many such proteins may you have to go through? How long would it take?

PIM.10. Before you cook an egg, the egg "white" is not at all white: it's clear. After you cook the egg, the "white" is white, because the large amount of globular protein has denatured (i.e., unfolded), and as a consequence, the protein has precipitated. Why should unfolding globular protein that are normally soluble in water cause them to stick to each other (which is what "precipitate" means)?

PIM.11. Lactate dehydrogenase (the last enzyme in human anaerobic glycolysis) is a soluble, multimeric protein. If you were to try to fold a single linear polypeptide chain of lactate dehydrogenase, you would find it impossible to do so without leaving a large number of hydrophobic amino acids exposed to water. Explain.

PIM.12. A patient exhibits signs of anemia. The red cell count is normal, as is the amount of hemoglobin, as judged by the binding of antibody directed against hemoglobin, but the binding of oxygen to whole cells is atypical (Fig. A below). You isolate hemoglobin from the patient and test binding of oxygen to the monomeric globin subunits. It is normal (Fig. B below). What mutation might account for these findings?



PIM.13. The gene encoding human interferon, the natural defense against viral infection, has been cloned in *E. coli* and **You Are There!** You can make a million bucks if you can only figure out how to make *E. coli* into an interferon factory. Problem: cloned interferon accumulates inside of *E. coli* cells, making them sick and greatly complicating purification of the protein. How might you engineer the gene to solve these problems?

PIM.14. Many proteins that form channels through membranes pass through the membrane multiple times. For example, rhodopsin, the light receptor protein in the rod cells of the retina, passes through the membrane seven times as alpha-helical chains. Below is a cartoon showing the side view of part of a hypothetical channel-forming protein -- call it rhodopsin. The circles are amino acid residues, the number of each corresponding to the amino acid's position in the chain. The roman numerals refer to membrane-spanning alpha-helical segments of the protein (only the first two are shown here). The top view shows how the seven α-helices participate in the formation of a pore through the membrane. The pore serves as the means by which protons can pass the membrane in response to light.

Congenital retinitis pigmentosa is a genetic disease leading to night-blindness. The disease exhibits a variety of symptoms of different severities, which, in many cases, have been linked to specific mutations in rhodopsin. For each given molecular outcome, choose one or more plausible amino acid mutations that could account for it. In each case, explain, briefly, why your choice(s) would lead to the outcome.

a. Rhodopsin found in cytoplasm, fails to insert in membrane.

b. Radical change in structure of rhodopsin. Channel doesn't form properly.

c. Overall structure of rhodopsin normal, but channel does not conduct protons.

d. Structure and function of rhodopsin normal.

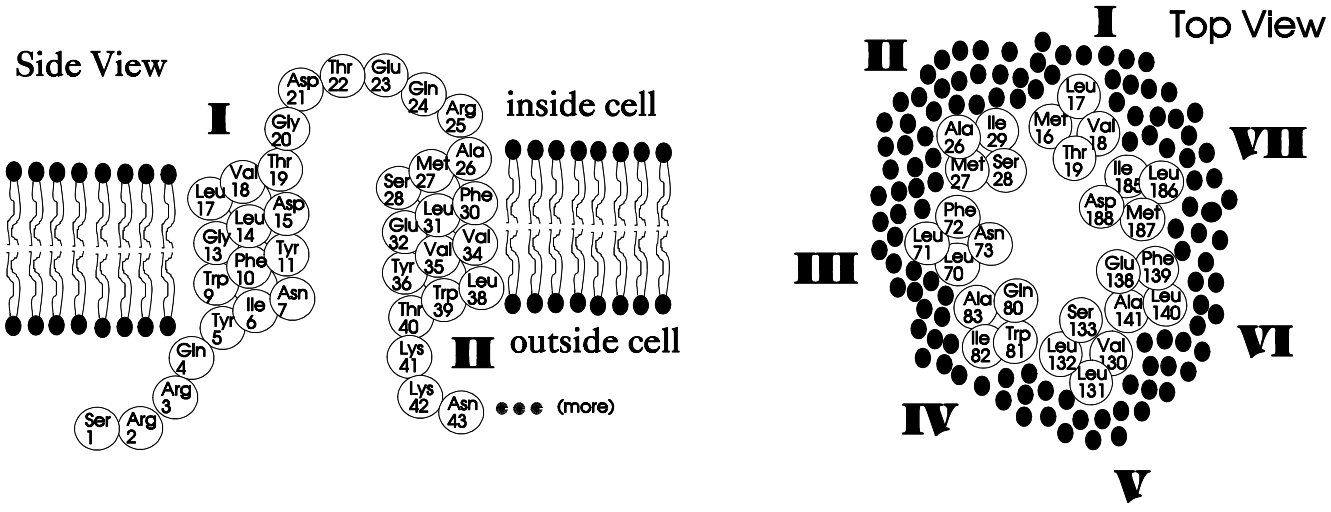
A. Insertion of three glutamates between Thr_{22} and Glu_{23}.

B. Insertion of three glutamates between Phe_{30} and Leu_{31}.

C. Glu_{138} mutated to arginine.

D. Asp_{188} mutated to leucine.

E. Mutation in amino acid not found in mature rhodopsin.



Abbreviations: **Ala**=alanine, **Arg**=arginine, **Asn**=asparagine, **Asp**=aspartic acid, **Cys**=cystine, **Gln**=glutamine, **Glu**=glutamic acid, **Gly**=glycine, **His**=histidine, **Ile**=isoleucine, **Leu**=leucine, **Lys**=lysine, **Met**=methionine, **Phe**=phenylalanine, **Pro**=proline, **Ser**=serine, **Thr**=threonine, **Trp**=tryptophan, **Tyr**=tyrosine, **Val**=valine

PIM.15. Of the 19 amino acids of glycophorin that lie within the membrane, some are hydrophilic (see Fig. 6 in notes entitled, *Protein Structure/Function*). What do you think may be the significance of this?

PIM.16. A child presents to you, her pediatrician, with all the classical symptoms of diabetes. Upon testing, you find that antibody against insulin detects only very low levels of insulin in her blood, but she responds normally to administered insulin. You are surprised to find, however, that the same antibody detects levels of insulin in the pancreas that are grossly higher than normal. What mutation might account for these findings?

PIM.17. An enzyme has a molecular weight of 60,000 daltons. When it is exposed to detergent, the protein breaks up to identical inactive components with molecular weights of 20,000 daltons. If the detergent is removed by dialysis, the 60,000-dalton protein reforms and regains enzymatic activity. You have isolated two mutant proteins. Mutant 1 shows no enzymatic activity and has a molecular weight of 20,000 daltons whether or not detergent is present. Mutant 2 has a molecular weight of 60,000 without detergent and 20,000 with detergent but shows no enzymatic activity in either case.

a. Suggest defects to explain the behavior of each of the mutant enzymes.

b. A person is heterozygous for Mutant 2 (i.e., has 50% Mutant 2 enzyme and 50% normal enzyme). How would you explain an observation that the person has 87.5% of the enzymatic activity of a normal person? How would you explain an observation of 12.5% activity?

c. Ascribe the terms "dominant" or "recessive" to the mutation leading to Mutant 2, according to the two situations presented in **b**.

Other matters of interest

Societal dilemmas posed by genetic advances

The ultimate in genetic engineering is to make your own gene that will encode a protein with whatever properties you like. We already have crude ideas at how the sequence of amino acids determines the three-dimensional structure of the protein and a few hints as to how the structure determines function. We're not that far away from designer enzymes. On one hand, it would be wonderful to be able to design protein catalysts for organic reactions that are currently impractical – the materials, pharmaceuticals, and other products that could result from this advance is staggering. Some have worried about a dark side. Proteins are most readily synthesized in organisms. Transgenic cows and other animals are already used to make proteins, e.g. insulin, for human use. Is there something lost by making animals into organic machines? Do we cross a line that should not be crossed by altering the genetic makeup of a cow to make it less than a cow and more of a factory?