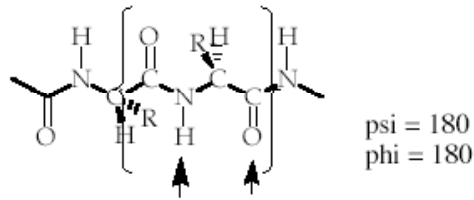


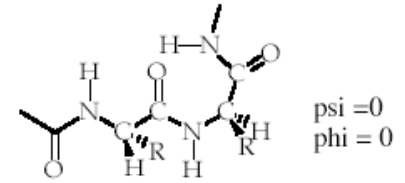
BMB 401 Spring 2004
Answers to Problem Set 3 Answers

1. α -helices, β -pleated sheets (β -conformations) and β -turns (hairpins) are the three primary protein secondary structures. Their common trait is that they are stabilized by hydrogen-bonding interactions between the amide proton and the carbonyl oxygen of the peptide backbone. In an α -helix, an amide proton forms a hydrogen bond with a carbonyl oxygen that is 4 residues away. The first four and last four amino acids in an α -helix cannot make intrachain peptide hydrogen bonds. The structure of the most common α -helix has 3.6 amino acids for every turn (360°), and traverses a distance of 5.4 Å per turn. Common phi and psi angles are -45° to -50° and -60° . By contrast, the β -conformation is a much more extended structure, in which interchain hydrogen bonds are formed rather than intrachain hydrogen bonds. Two types of β -conformations are recognized, the antiparallel β -sheet and the parallel β -sheet. The hydrogen-bonding interactions in an antiparallel β -sheet are more linear and stronger than in a parallel β -sheet. In each of these secondary structures, it is the need to form hydrogen-bonding interactions that promotes formation of these secondary structures as well as stabilizes them.
2. α -keratin is composed of 3.6₁₃ right-handed α -helices. The amino acid sequence of α -keratins causes two strands to wrap around each other in a left-handed helix or twist, because of hydrophobic amino acids that are being shielded from water. This left-handed twist is called a coiled coil. The points of contact in the coiled coil are the hydrophobic amino acids. Collagen is composed of left-handed helices because of a unique G-X-P (hydroxyP) repeating motif that induces this conformation. Three chains fold or twist about each other to form a coiled coil structure, which in this case is right-handed. The keratin structure is stabilized by hydrogen bonds and hydrophobic interactions between chains, as well as disulfide bonds in some cases. The collagen structure is stabilized by hydrogen bonds within the peptide chain itself, as well as hydrogen bonding between post-translationally modified amino acids. In addition, a number of covalent cross-linkings involving modified lysine amino acids permit added stability.

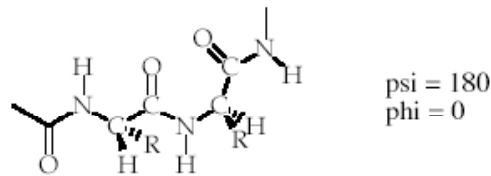
3.



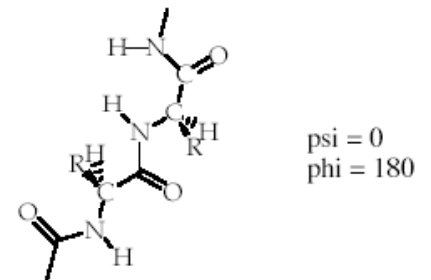
Not too bad a configuration. Might have steric problems with the amide hydrogen and the carbonyl oxygen.



Clear steric problems with the amid proton and carbonyl oxygen



Clear steric problems with the 2 carbonyl oxygens



The two protons would be closer than their normal van der Waals contact distances.

4. There are several ways to work this problem. Using a helix wheel is probably the easiest. You're trying to find how many amino acids are between two that are stacked exactly on top of each other in a 3.6_{13} α -helix. In other words, the two amino acids will lie in an imaginary line that traverses the helix. On a 3.6_{13} helix wheel, you know that each amino acid is 100° apart from its immediate neighbors. Therefore, you need to find a multiple of 360° that will give you a whole number when divided by 100. You will find that if you make five turns of the helix, you will have two amino acids to lie in a line that traverses the helix, and that is parallel to the helix axis. $5 \times 3.6 = 18$. Therefore, there are 17 amino acids between the starting one and the final one.

5. 660 mg of an oligomeric protein of molecular weight is 5×10^6 mol of that protein. You know that Sanger's reagent will react with the N-terminus of a protein. If there is only 1 polypeptide, then there should only be one equivalent of the compound shown. If there are 2 polypeptides, then there should be 2 equivalents etc. In order to establish the number of moles of the compound shown in the figure, you need the molecular weight of the compound, which can be generated by simply adding the molecular weights of all of the atoms (283 g). This gives 1.94×10^{-5} mol. 1.94×10^{-5} mol / 5×10^{-6} mol = 3.88. Therefore, this protein is most likely tetrameric.

6. Because of the amine group on the tryptophan side chain, it can hydrogen bond with other amino acids. The side chain of phenylalanine cannot

hydrogen bond. Although the side chain of tyrosine can hydrogen bond, this bond cannot prevent the chain from flipping, because it does not impede movement around the axis of rotation denoted by the β -carbon and the oxygen of the hydroxyl group. Even if tryptophan were not hydrogen bonded, because the axis of rotation is not symmetric through the R group, the volume that it would sweep out would be exceedingly large, inevitably leading to steric clashes.

7. This problem needs to be dealt with in terms of hydrophobicity, propensity for certain secondary structures, ability to form certain bonds, and any other special characteristics that a given amino acid might have.
 - a. Leucine to phenylalanine in general is a fairly conservative mutation. Both are hydrophobic, and both have same secondary structure preferences.
 - b. Lysine to glutamate can be dangerous if the lysine were involved in a salt bridge or hydrogen bond. Lysine typically acts as a hydrogen bond donor, whereas glutamate typically acts as a hydrogen bond acceptor. If the lysine were on the exterior of the protein, then the mutation would not be as critical. Both are well solvated by water.
 - c. Valine to threonine is not a particularly bad mutation. Valine is a little more non-polar than threonine. They both have similar secondary structure preferences.
 - d. Glycine to alanine is not too bad a mutation. In fact, alanine is the closest amino acid to glycine in structure. Glycine prefers β -turns, and doesn't like α -helices or β -sheets. They both are hydrophobic amino acids though. The reverse mutation would probably not be good, since alanine likes to be in α -helices, and glycine does not.
 - e. Methionine to proline is not a good mutation. Although they both are hydrophobic, methionine likes α -helices, whereas proline is a helix breaker.
 - f. Aspartate to asparagine is not too bad. They have similar secondary structure preferences. Aspartate is typically a hydrogen bond acceptor, whereas asparagines can act as both.
8. From the data given it is clear that the quaternary structure of this protein is tetrameric. The size of the entire protein is 360,000 Da, whereas the size of each subunit is 90,000 Da. The question is how much of the pyridoxal cofactor is bound to 1 mg of protein. 1 mg of tetramer (using 360,000 Da) corresponds to 2.8×10^{-9} mol. $1.86 \mu\text{g}$ of pyridoxal corresponds to 1.1×10^{-8} mol. $1.1 \times 10^{-8} \text{ mol} / 2.8 \times 10^{-9} \text{ mol} = 3.92$. Therefore, there are approximately 4 pyridoxals per tetramer, or 1 per subunit.
9. There is only one axis of symmetry (three-fold), which is perpendicular to the plane of the paper. Therefore the symmetry is C_3 . For the second molecule there is only one axis of symmetry (two-fold), which is in the plane of the

paper. Therefore, the symmetry is C_2 . In the third molecule there is a C_3 axis perpendicular to a C_2 axis. The C_2 axis while split one of the protomers in half. Therefore the symmetry designation is D_3 .