Biochemistry CUME October 2018

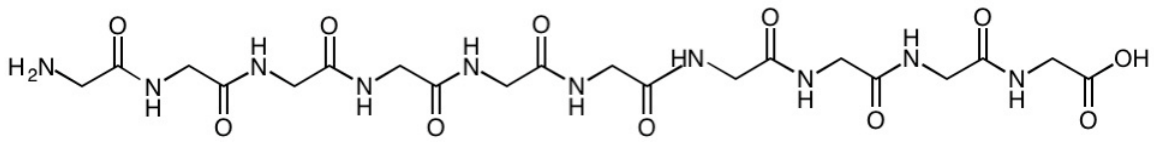
Topic: Protein Structure Analysis

Protein structures can be described and characterized at four different levels - primary structure (1°), secondary structure (2°), tertiary structure (3°) and quaternary structure (4°).

1. Primary Structure (25 points)

(a) **(10 points)** The primary structure of a protein refers to its amino acid sequence. Please draw out the chemical structure of the given peptide based on its sequence. Add the side chains to the protein backbone from the amino-terminus to the carboxyl terminus.

Peptide sequence: `LHAVNDRQKW`



(b) **(5 points)** Which group (charged, polar and non-polar) does each of these 10 amino acids in **1(a)** belong to? Please use the 3-letter codes to identify amino acids.

- Charged:
- Polar:
- Non-polar:

c) **(10 points)** What biophysical or biochemical methods can be used to determine protein sequences? Explain the principles behind such experimental method(s).

2. Secondary Structure (25 points)

(a) **(15 points)** Alpha-helix is an important element of protein secondary structure. A convenient way to illustrate the amino acid sequences in helices is the helical wheel or spiral. Since one turn in an α-helix is 3.6 residues long, each residue can be plotted every 100º (=360/3.6) around a circle or a spiral. Such a plot shows the projection of the position of the residues onto a plane perpendicular to the helical axis. Residues on one side of the helix are plotted on one side of the wheel. Please map the following three protein sequences onto a helical wheel. Based on the positions of residues in the helical wheel, which of the three helices (1, 2 and 3) is most likely part of a trans-membrane protein. Explain why?

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Helix 1: LSFAAAMNGLA
Helix 2: INEGFDLLRSG
Helix 3: KEDAKGKSEEE
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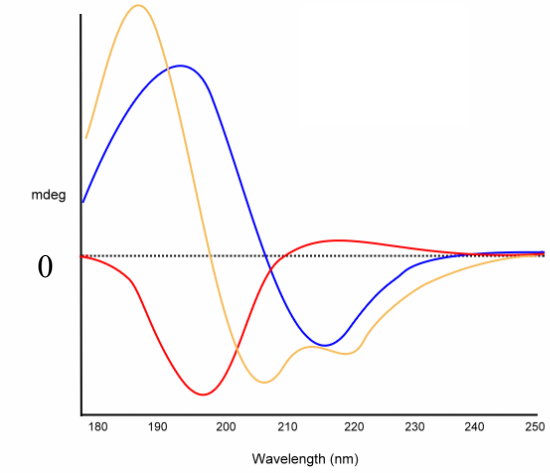
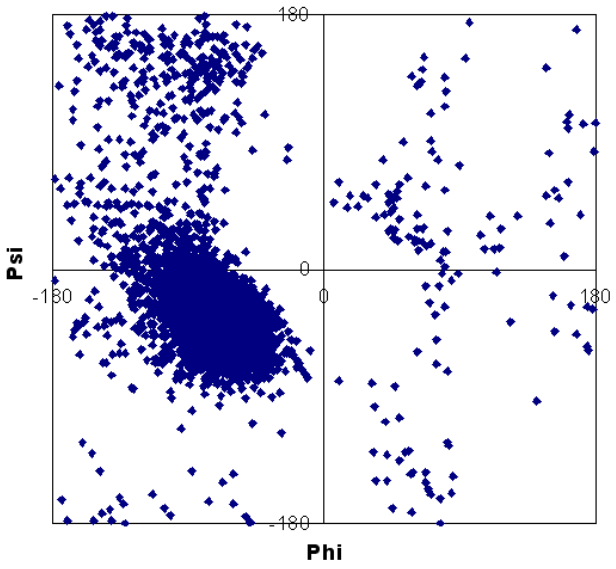
(b) **(10 points)** Circular dichroism (CD) spectroscopy is often used to examine protein secondary structures. In the figure shown below, each colored curve represents the characteristic CD spectrum for one of the three most common secondary structures, namely alpha-helix, beta-sheet and random coil. Can you find the correspondence between these curves and the named secondary structures? Explain the origin for such optical activities.



3. Tertiary Structure (25 points)

(a) **(5 points)** Protein folding is a physical process by which the linear polypeptide chain of a protein is folded into a specific and functional conformation in the three-dimensional space. What are the major interactions or forces that drive the protein folding process?

(b) **(10 points)** The Ramachandran diagram shows the distribution of the backbone dihedral angles (ϕ, ψ) in a protein structure. Please **circle** and **label** those region(s) in the ϕ−ψ plot where you expect to find residues from alpha-helix and beta-strand, respectively.



(c) **(10 points)** Several biophysical methods such as X-ray crystallography, Nuclear Magnetic Resonance (NMR) and cryo-electron microscopy have been widely used to determine the tertiary protein structures at atomic or near atomic resolution. Discuss their strengths and limitations in protein structure determination.

4. Quaternary Structure (25 points)

(a) **(15 points)** Human hemoglobin (Hb) is a hetero-tetramer consisting of four subunits, two α-subunits and two β-subunits. What function or property of Hb requires such tetramer? Explain why.

(b) **(10 points)** Discuss one or two biophysical or biochemical methods that are commonly used to determine the quaternary structure of a multi-subunit biological assembly. Explain why and how.