Biology 5357

Chemistry & Physics of Biomolecules

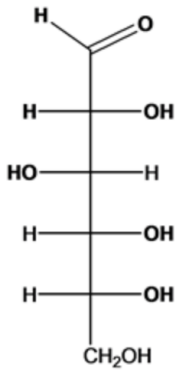
Examination #3

Glycobiology, Membranes
& Membrane Proteins Module

December 9, 2019

Name: _____

Question 1. (8 points, A=6, B=2 pts) The Fischer projection of D-glucose is shown in the figure below.



(A) Trehalose is the disaccharide Glc-α1-Glc-α1. Draw the structure of trehalose, showing the stereochemistry at each chiral center. You may draw your answer using either Haworth projections or 3-D "chair" structures.

(B) Trehalose is a non-reducing sugar. What is the definition of "non-reducing"? Indicate the non-reducing feature(s) on your structure drawn in part (A).

Question 2. (8 points, 2 pts each) In a couple of sentences, briefly describe or define each of the following terms, and its importance in glycobiology:

(A) Mucin

(B) Lectin

(C) Fucose

(D) Ceramide & Glycosphingolipid

Question 3. (8 points, 4 pts each)

(A) End labeling of a glycan with 2-aminobenzamide, followed by sequential digestion with different glycosidase enzymes can be used to analyze glycan structure. What does the end labeling step do, and how is it detected? How can the results of the enzymatic reactions be detected?

(B) Explain the basic ideas behind MS/MS analysis of glycans, and how describe how this technique can be used in determining the structure of a glycan.

Question 4. (8 points, A & C=2, B=4 pts)

(A) Which amino acid is the most common attachment point in *N*-glycoproteins? Which amino acids are most often modified in *O*-glycosylation?

(B) Many of the initial steps in *N*-glycosylation occur in the endoplasmic reticulum. Explain how UDP-Glc, GDP-Man and dolichol are involved in this process. A diagram of the ER may be helpful. What is meant by "high mannose glycan"?

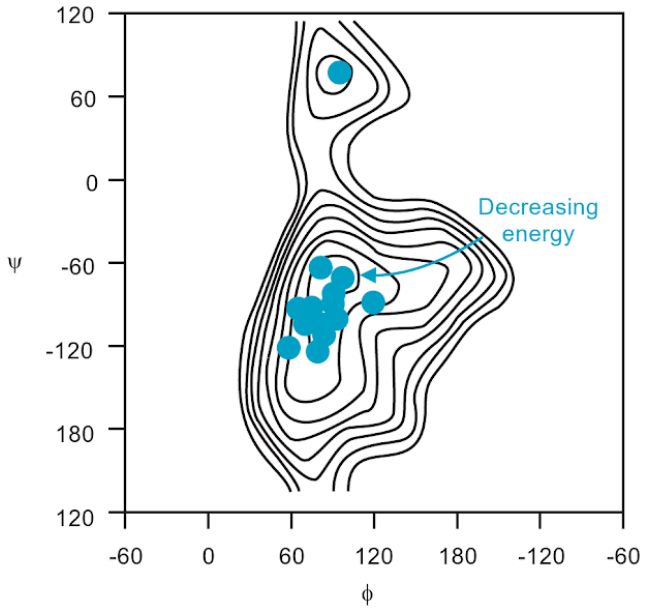
(C) After processing in the ER, a future secreted protein moves to the Golgi apparatus. What steps occur in the Golgi?

Question 5. (8 points, A & B=2, C=4 pts)

(A) Which isolated reaction has a positive free energy, formation of a glycosidic bond or breakage of a glycosidic bond?

(B) What is the approximate free energy value associated with conversion of ATP to ADP? How many ATPs are consumed in driving the reaction you chose in part (A)?

(C) Below is the conformational potential energy surface for a D-Man-α1-3-D-Man disaccharide unit. Draw the structure of D-Man-α1-3-D-Man (Note mannose is a hexopyranose, like glucose. You *do not* need to include the stereochemistry.) Mark the torsion angles in your structure corresponding to ϕ and ψ in the figure.



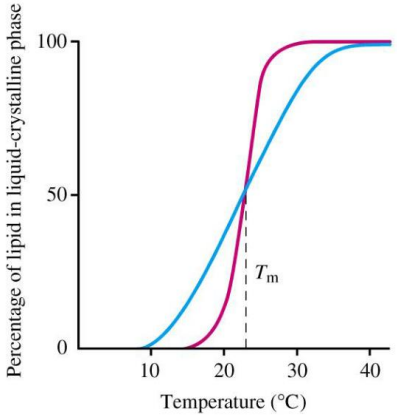
Question 6. (8 points, B & D=2 pts, others 1 pt) Cholesterol is an important component of biological membranes:

(A) What class of lipid is cholesterol?

(B) What is the proportion of cholesterol in the endoplasmic reticulum *vs*. plasma membrane?

(C) What does cholesterol do to the membrane thickness?

(D) The figure below shows results from a differential scanning calorimetry (DSC) experiment of membranes with pure phospholipids (magenta) and phospholipids + cholesterol (blue). Draw the corresponding DSC thermatograms each sample.



(E) Why does the addition of cholesterol increase membrane fluidity at 15°C?

(F) Why does the addition of cholesterol decrease membrane fluidity at 30°C?

Question 7. (6 points, 2 pts each) Describe how the following factors affect the free energy of micelle formation, and provide an explanation:

(A) Increasing the chain length

(B) Increasing the salt concentration

(C) Increasing the temperature

Question 8. (8 points, 2 pts each) Describe how the following factors affect the lipid area in a fluid bilayer, and in each case provide an explanation.

(A) Increasing chain length for saturated chains

(B) Saturated *vs*. unsaturated

(C) For unsaturated chains, moving the position of the double bond

(D) Cis *vs*. trans unsaturation

Question 9. (4 points) What are four material properties of membranes?

Question 10. (20 points, A=10, B=4, C=4, D=2 pts) Consider the five different amino acid partitioning scales that were discussed in class: (i) Radzika, (ii) Wimley, (iii) Moon, (iv) Hessa and (v) MacCallum.

(A) Describe the setup of each experiment or calculation. Draw a cartoon depicting the change in states from water to the "lipid" model.

(B) The results of each of these methods give different values for the partitioning free energies of side chains from water to lipid bilayers, with the Hessa scale providing the lowest free energies. Provide a ranking of the other four methods relative to the Hessa partitioning scale.

Highest: (1)

(2)

(3)

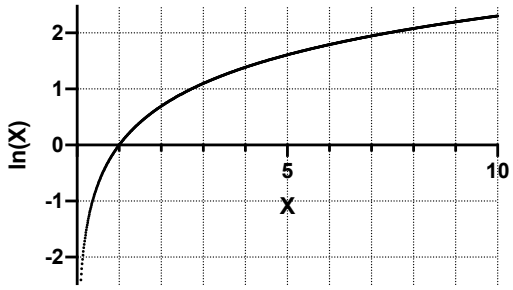
(4)

Lowest: (5) Hessa

(C) Provide an explanation of why the Radzicka method measures larger free energies than the Hessa method.

(D) Often times, hydropathy analysis based on these partitioning scales incorrectly predicts transmembrane helices in membrane proteins. Provide one reason why and explain.

Question 11. (2 points) Consider a cell in seawater ($[K^+]$ ~ 10 mM, $[Na^+]$ ~ 500 mM, $[Cl^-]$ ~ 500 mM). What would be the membrane potential of a cell (internal ion concentrations $[K^+]$ ~ 4 mM, $[Na^+]$ ~ 100 mM, $[Cl^-]$ ~ 100mM) if the membrane contained open Na^+ selective leak channels?



Question 12. (2 points) Explain why the cell membrane is a physical barrier to charged species?

Question 13. (6 points) What are the three main methods of passive transport across membranes? Describe the physical mechanism of each.

Question 14. (4 points) What are the two main methods of active transport across membranes? Explain what drives the uphill movement of species in each situation.