Biology 5357

Chemistry & Physics of Biomolecules

Examination #3

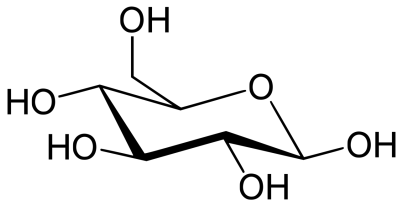
Glycobiology, Membranes
& Membrane Proteins Module

December 8, 2023

Name: _____

Question 1. (15 points; A-C, 2 pts each; D-F, 3 pts each)

Shown below is a possible chair conformation for D-glucose.



(A) Does the structure show the α- or β-form of glucose?

(B) Is glucose an aldose or a ketose? Explain.

(C) Label the anomeric carbon on the structure.

(D) Draw the Fischer projection for the "open" form of glucose.

(E) *N*-Acetylglucosamine (GlcNAc) is a very common derivative of glucose, which is modified at carbon-2. Show its structure.

(F) Chitin is a β-1,4-linked polymer of GlcNAc, occurring in the exoskeleton of hard-shelled animals, the cell walls of some fungi, and other natural sources. Over 1 billion tons of chitin is produced each year in Earth's biosphere. Draw the structure of a GlcNAc dimer from chitin, showing a glycosidic bond.

Question 2. (9 points)

Your research group is working on a cell surface receptor that has three potential *N*-linked glycosylation sites (Asn-X-Ser/Thr) and a proline-rich sequence containing several threonine residues in its extracellular region. You have expressed a soluble form of the receptor in Chinese hamster ovary (CHO) cells and can purify roughly 1 mg from the medium. Discuss some of the experimental approaches you would use to characterize the glycosylation of the expressed form of the receptor.

Question 3. (8 points; A-B, 4 pts each)

In the *N*-linked glycoprotein biosynthetic pathway, partially processed proteins transfer from the endoplasmic reticulum (ER) to the Golgi apparatus. "Misfolded" proteins encounter lectins in the membrane of the ER, which triggers a process known as endoplasmic reticulum-associated protein degradation or ERAD. Proteins involved in ERAD get translocated to the cytoplasm and tagged with ubiquitin to make them substrates for the proteasome.

(A) Why might lectins be well-suited to recognition of damaged or misfolded proteins exiting the ER?

(B) In mammalian cells, the ERAD mechanism involves removal of proteins with most or all of the α-1,2-linked mannose residues. Targeting via ERAD involves M-type lectins that are structurally related to the mannosidase ERManI. Explain why this makes sense in terms of your knowledge of glycoprotein biosynthesis.

Question 4. (12 points; A-D, 3 pts each)

Describe an experiment and findings that provided support for the following hypotheses about cellular boundaries.

(A) Cellular boundaries are oil-like

(B) Cellular boundaries are lipid bilayers

(C) Cellular boundaries are fluid

(D) Cellular membranes have proteins embedded within

Question 5. (6 points; A-B, 3 pts each)

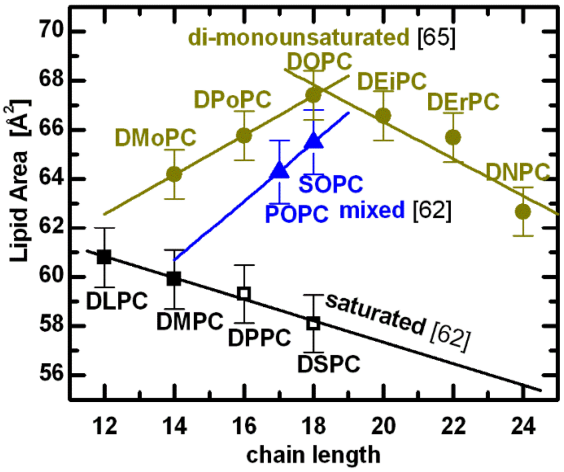
Cellular membranes have diverse phospholipid compositions. For each of the following, describe three types of modifications/variations in lipid chemistry and provide examples.

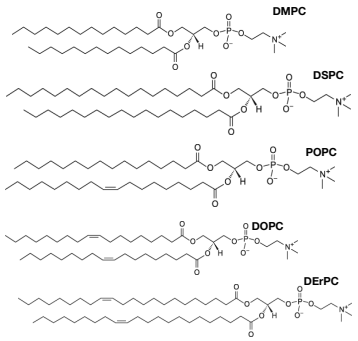
(A) Headgroup chemistry

(B) Acyl chain chemistry

Question 6. (8 points; A-D, 2 pts each)

Consider the following plot from lecture that shows lipid surface area vs. chain length. Example lipid structures are also shown for reference.





Describe the changes in intermolecular interactions that lead to altered lipid configurations in the trends observed for:

(A) Saturated lipids

(B) Mixed chain lipids

(C) Di-monounsaturated lipids from DMoPC to DOPC

(D) Di-monounsaturated lipids from DOPC to DNPC

Question 7. (8 points; A-D, 2 pts each)

Please answer each of the following questions:

(A) What is the main lipid bilayer phase transition? What are the molecular changes?

(B) Describe a change in the lipid chemical structure that will yield an increase in T_m. Explain why on a molecular level.

(C) Describe a change in lipid chemical structure that will decrease T_m. Explain why on a molecular level.

(D) Describe a change in lipid composition that will make the phase transition broader. Explain why on a molecular level.

Question 8. (10 points; A = 4 pts, B = 6 pts)

Consider energetics of amino acids and proteins in membranes.

(A) In the amino acid partitioning studies, the free energy of transferring an arginine side chain from cyclohexane into water is measured to be +14.92 kcal/mole. However, in the OMPLA equilibrium folding experiments, the cost of partitioning arginine into the center of the lipid bilayer is much lower, just +2.14 kcal/mole. Explain the differences between these two experimental approaches and why the energy is lower in the second experiment.

(B) Describe three different considerations that must be made to measure equilibrium membrane protein folding in membranes, compared to soluble protein folding equilibrium in water/buffer.

Question 9. (8 points)

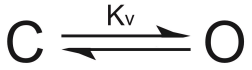
Consider the different mechanisms of membrane transport/permeability. Describe the main mechanistic differences between (a) ion channels, (b) uniporters, (c) secondary active transporters, and (d) pumps. Please include a description of the driving force for the transport or permeation.

Question 10. (8 points)

Nerves have long processes called axons which can project very large distances compared to the size of the cell body. Electrical signals travel through these projections at a rapid pace much faster than simple chemical diffusion. Explain briefly why that is the case. Feel free to draw a diagram if that is easier.

Question 11. (8 points)

Consider a simple two state system describing a voltage-gated ion channel as shown below.



Write the equation describing open probability, Po, in terms of the equilibrium constant Kv. What is the key feature of the charge (Q)-voltage and Po *vs*. voltage curve that characterizes this system?