

True / False (1/2 point each; 12 points total)

1. Regarding lipid rafts, including information in the review article by Nichols:
 they are disrupted by non-ionic detergents.
 they are a prerequisite for dynamic protein clustering.
 they are enriched in sphingolipids.
2. Regarding FRET:
 good FRET pairs must have non-overlapping excitation frequencies.
 a good FRET signal requires FRET pairs to be in close proximity.
 it is effective for monitoring lipid / protein interactions but not lipid / lipid interactions
 FRET pairs must be of the same molecule type (i.e. GFP variants or chemical fluorophores, but not a combination of the two).
3. Regarding protein secretion:
 type I prokaryotic secretion involves translocation of unfolded protein.
 GTP is required for docking of a translationally stalled ribosome onto the ER membrane.
 computer algorithms can accurately predict from primary sequence whether a protein will be an integral membrane protein or a secreted soluble protein.
4. Regarding lipid biosynthesis:
 serine can be incorporated into both glycerophospholipids and sphingolipids.
 ATP is the source of phosphate found on all glycerophospholipids.
 glycerophospholipids can be made by chemically modifying the head groups of other glycerophospholipids.
5. Regarding kinases:
 they can target amino acids other than serine, threonine, and tyrosine.
 they can target lipids.
 PKA is palmitoylated.
 PKC is palmitoylated.
6. G α subunits:
 are isoprenylated.
 bind GTP.
 can inhibit cAMP production.
7. Regarding ABC transporters,
 they can either import or export cargo, but no one transporter can do both
 they are always encoded by a single polypeptide
 they are part of the complex required for Type I bacterial secretion
 their ATPase activities can be probed using azido ^{32}P - γ ATP.

Multiple Choice (2 points each; 16 points total; one correct answer per question)

8. Approximately, what percentage of medically relevant drugs target membrane proteins?
A) 50%
B) 33%
C) 25%
D) Less than 1%
9. The biological membrane of what organelle allows for free movement of ions and small proteins?
A) endoplasmic reticulum
B) peroxisome
C) nucleus
D) lysosome

10. Which of the following organelles not utilize a post-translational translocation system for movement of protein across its membrane?
- A) Golgi
 - B) peroxisome
 - C) nucleus
 - D) mitochondria
11. The translocon and ribosome form a tight seal that prevents ion leaks from occurring during co-translational translocation. An experiment was discussed during class that supported this observation. Which of the following components was not used in that experiment?
- A) a fluorescently labeled amino acid
 - B) a protease
 - C) a pore forming toxin
 - D) an mRNA lacking a stop codon
12. The ENTH domain of Epsin is proposed to interact with:
- A) a glycerophospholipid
 - B) a sphingolipid
 - C) cholesterol
 - D) clathrin
13. Which of the following is not typically found in biological membranes?
- A) triacylglycerols
 - B) glycerophospholipids
 - C) sphingolipids
 - D) glycoproteins
14. Which of the following characteristics is not shared by cholesterol and steroidal hormones?
- A) a polar head group
 - B) a long alkyl side chain
 - C) a nonpolar core
 - D) buried C18 and C19 methyl groups
15. Which of the following does PKC not interact with?
- A) acidic phospholipids
 - B) calcium
 - C) diacylglycerol
 - D) IP3

Short Answer - These can be answered using one or two keywords, lists, diagrams, and/or a 1-2 sentences (1-5 points each; 38 points total)

16. List four agents that could be used to disrupt the association of peripheral but not integral membrane proteins. Your answers should reflect information obtained from class, the lecture notes, or assigned readings. (4 points)

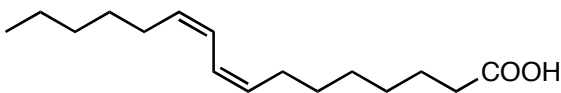
17. In class, there were two videos demonstrating the FRAP technique. One was of an ER-localized protein, and the other was of a Golgi localized protein.

What conclusions were drawn from the videos about the proteins? (2 points)

What specific aspect of the traditional fluid mosaic model did the observations challenge? (1 point)

18. Based on your readings of transmembrane segments and the class discussion of porins, what specific amino acids are commonly found at interfacial regions of membranes? (2 points)

19. Using the stick figure method, draw the chemical structure of a naturally occurring 16:2($\Delta^{8,10}$) fatty acid. (4 points)



20. Biotin is required for what specific step of fatty acid biosynthesis? (1 point)

21. Both radioactive and non-radioactive inorganic phosphate can be taken up by cells and incorporated into membrane lipids. Why is radioactive phosphate not a good tracer for sphingolipids? (1 point)

22. The reactive end of Coenzyme A (CoA) and the reactive transfer centers of the fatty acid transferase commonly utilize what atom for their chemical functions? (1 point)

23. What is meant by the term hydrophathy analysis and what is its utility? (3 points)
24. You were told to ignore the analogy of membrane proteins being like icebergs floating in an ocean of lipids. What new analogy were you presented with? (1 point)
25. What are the two main categories of lipids as defined in lecture? (2 points)
26. What nucleophiles are used in the process of cleaving peptide bonds in the context of a cysteine protease? Be sure to state the source of the nucleophile. Use words or a diagram. (4 points)
27. The C2 protein interaction domain binds what two components? (2 points)
28. What are the nucleophiles required for peptide bond hydrolysis in the synthesis of the hedgehog signaling molecule? You should not diagram the hedgehog biosynthesis pathway. (2 points)
29. What is unusual about the C-terminal amino acid of the Ras oncoprotein? Use words or a well-labeled diagram. (2 points)
30. What event lowers the desolvation energy barrier associated with the binding of Ca^{2+} ions to the Ca^{2+} P-type ATPase? (2 points)

31. By comparison to other ion conducting molecules, how does the interior of the gramicidin A conducting channel differ? (1 point)
32. Bacterial toxins such as those that cause cholera, pertussis, and anthrax generally have three functional domains. What are these? (3 points)

Long Answer - answer the following questions using several sentences, highly descriptive diagrams, and/or highly descriptive lists. (3-8 points each; 34 points total). For all questions in this section, *use sufficient detail to convey your knowledge of the material (i.e. do not use one word responses or diagrams without labels!)*.

33. Using a well-labeled diagram, depict the main features of the SRP (Signal Sequence Recognition Particle) in such a way that illustrates your knowledge of its chemical composition and properties that allow it to stall translation. Do not diagram the SRP cycle. (7 points)

34. In the article by Reguena *et al*, complexes of varying sizes were identified depending on the buffer conditions utilized. Explain how the choice of detergent impacted the results and the authors' rational explanations for the increased complex size. (4 points)

35. The serine proteases trypsin, chymotrypsin, and elastase have differential sensitivities to the classical serine protease inhibitors TPCK and TLCK. Explain why this is the case. (6 points)

36. Using the lecture as a reference, explain OR diagram the situations encountered by sic1 with regards to its role in regulating cell cycle progression. (8 points)

37. At the biochemical / molecular level, what would be the consequence of an adding an epitope tag to the C-terminus of proteins such as TRP, or forms of PLC and PKC, that are involved in phototransduction in *Drosophila*? (3 points)

38. Aquaporins have three features that allow for selective passage of water through biological membranes. Describe these features. (3 points)

39. Several biomedically relevant ABC proteins were discussed in class. Using one as an example, provide the name the ABC transporter, its normal cargo, and the biomedical importance of the transporter. (3 points)