

April 1, 2008

True / False (0.5 point each; 10 points total)

1. Regarding integral membrane proteins:
 they typically have transmembrane segments that are 10 nm in width.
 they typically constitute 25% of the genes in a typical genome.
2. Regarding naturally occurring fatty acids:
 they typically have an even number of carbon bonds.
 they typically have *trans* double bond configurations when unsaturated.
 they are relatively insoluble in water.
3. Regarding prokaryotic protein secretion:
 the Sec-system translocates unfolded proteins.
 the signal sequence targeting proteins to the Sec-system is retained on secreted proteins.
 Gram positive prokaryotes make extensive use of Type III systems for infection.
4. Proteins with a PDZ domain:
 typically bind a phosphorylated target.
 interact with the N-termini of proteins that are modified by the methionine aminopeptidase.
5. In terms of protease mechanisms:
 an aspartic protease forms an acyl intermediate with its substrates.
 the N-terminal product produced by all 4 proteolytic mechanisms would be radiolabeled in the presence of $^{18}\text{O}\text{-H}_2\text{O}$.
6. Regarding protease inhibitors:
 PMSF inhibits elastase, chymotrypsin, and trypsin.
 TLCK inhibits chymotrypsin.
7. GPI-linked proteins:
 are found in single celled parasites on the extracellular face of the plasmamembrane.
 are anchored in plasma membranes because of their conjugation with a sphingolipid.
8. On the topic of protein channels:
 they can be either gated or ungated.
 the aquaporin channel is 5.8 Å in diameter at its narrowest.
9. Regarding the 4 types of transport ATPases discussed in class:
 they can all be manipulated to hydrolyze ATP.
 the P-type ATPases represent the largest transporter gene family.

Multiple Choice (2 points each; 28 points total; one correct answer per question)

10. Which of the following biological membranes does not actively restrict ion movement across it?
 - A) endoplasmic reticulum
 - B) nucleus
 - C) peroxisome
 - D) plasma membrane

11. Which of the following amino acids is not a part of glutathione?
A) cysteine
B) glutamate
C) glycine
D) serine
12. Which of the following is the best source of dietary docoahexanoic acid?
A) beef
B) fish
C) leafy vegetables
D) nuts and seeds
13. The polar head group on cholesterol is derived from which functional group?
A) an amine
B) a hydroxyl group
C) a phosphate group
D) a sulfhydryl group
14. Cholesterol is similar in size to a fatty acid of which length?
A) C12
B) C14
C) C16
D) C18
15. What is the relative abundance of docosahexaenoic acid in the plasma membranes of neurons (by weight relative to other lipid components)?
A) 50%
B) 33%
C) 25%
D) 10%
16. Approximately, what percentage of medically relevant drugs target membrane proteins?
A) 50%
B) 33%
C) 25%
D) 10%
17. Which of the following is the binding target of the FYVE domain:
A) PI(3)P
B) PI(3,4)P2
C) PI(4,5)P2
D) PI(1,4,5)P3 – more commonly known as IP3
18. The FYVE domain requires which ion cofactor:
A) Ca^{2+}
B) Fe^{2+}
C) Mg^{2+}
D) Zn^{2+}

19. Which of the following is not known to be nucleophile in proteolytic mechanisms?
- A) cysteine
 - B) histidine
 - C) serine
 - D) water
20. Which of the following eukaryotic organelle membranes is not known to have an associated ABC transporter?
- A) endoplasmic reticulum
 - B) lysosome
 - C) nucleus
 - D) plasma membrane
21. Which of the following typically exists in monomeric form in biological membranes?
- A) ABC-type ATPase
 - B) aquaporin channel
 - C) OmpF general porin
 - D) P-type Ca^{2+} ATPase
22. Based on class discussion, which of the following amino acids is least likely to be at the interfacial region of a porin or channel ionophore?
- A) His
 - B) Phe
 - C) Trp
 - D) Tyr
23. Which of the following does not contribute to the selectivity of the aquaporin channel?
- A) desolvation filter
 - B) dipole filter
 - C) proton filter
 - D) size filter

Short Answer - These can be answered using one or two keywords, lists, diagrams, and/or a few sentences (1-4 points each; 31 points total)

24. Using the stick figure method, draw the chemical structure of a naturally occurring 18:2($\Delta^{9,12}$) fatty acid. (4 points)

25. Transmembrane segments of which type of secondary structure are not readily identified by existing computer algorithms? (1 point)
26. What is the common over-the-counter name for dietary docoahexanoic acid? (2 points)
27. List the predominant sequence features and/or protein domains of a prokaryotic protein that is destined for secretion via the autotransporter mechanism? (3 points)
28. List two protein interaction domains that were discussed in class that function similarly to the FYVE domain. (2 points)
29. What two types of proteases are generally required for production of peptide hormones such as adreno corticoprophin hormone (ACTH) and β -endorphin? (2 points)
30. Why is palmitoylation not an accurate descriptor for this type of lipid-based protein modification? (2 points)

31. List the types of lipid-modified proteins that do not have a free carboxylic acid at the C-terminus? (3 points)
32. Which lipid is typically found conjugated to mammalian retroviral *Gag* polypeptides such as HIV *Gag*? (2 points)
33. What new paradigm (i.e. pathway) did Dr. Schmidt describe that expands the known paradigms for mediating signal transduction across biological membranes. (2 points)
34. List any four of the six domains that make up the Ca^{2+} P-type ATPase? (4 points)
35. List the two types of energy potential used to drive ATP synthesis? (2 points)
36. What bacterially derived reagent was essential for the experiments used in determining that the translocon channel was open and continuous with the exit channel of translocating ribosomes? (2 points)

Long Answer - answer the following questions using several sentences, highly descriptive diagrams, and/or highly descriptive lists; in the case of the experimental diagram, just fill in the appropriate banding pattern (4-8 points each; 31 points total). *Use sufficient detail to convey your knowledge of the material; do not use one-word responses or diagrams without labels!*

37. As discussed in class, in what two major ways do diagrams/cartoons of biological membranes typically deviate from reality? (4 points)

38. Using the blank template provided, diagram the results of a protease protection experiment that you would expect for a plasma membrane (PM)-localized integral membrane protein having an extracellular N-terminus. Assume that you are using whole cells (not microsomes) as the source material for the experiment and that your detection assay is based on the immunoblot/Western technique. (4 points)

Location of reporter:	N-term				C-term			
TX-100:	-	-	+	+	-	-	+	+
Prot. K:	-	+	-	+	-	+	-	+
PM candidate blots:	<div style="border: 1px solid black; height: 40px; width: 100%;"></div>				<div style="border: 1px solid black; height: 40px; width: 100%;"></div>			
Intracellular control blots:	<div style="border: 1px solid black; height: 40px; width: 100%;"></div>				<div style="border: 1px solid black; height: 40px; width: 100%;"></div>			

39. During the course of your thesis research, you have identified two proteins (proteins A and B) that you believe must form a heterodimer. Consistent with your hypothesis, you have determined that you can co-immunoprecipitate both proteins using the anti-myc antibody when only protein B is tagged at its C-terminus with the myc epitope. However, the opposite is not true. Protein B is not recovered when protein A is tagged with a C-terminal myc epitope. Given your knowledge of protein-protein interactions, what type of domain might you be dealing with and on which protein is it likely to be found? (8 points)

40. Explain with some structural detail the means by which syntrophin and neuronal nitric oxide synthase physically interact. (6 points)

41. Diagram the key functional domains of a *Clostridial* neurotoxin, elaborate on the function of each domain, and indicate the cytosolic component that helps activate the neurotoxin. Do not describe the endocytotic uptake of this neurotoxin! (9 points)

Bonus Questions – (5 points total)

1. List the names of two members of your group as assigned in class. (2 points)
2. As discussed in class, what is an intriguing new therapeutic target for treating the cellular affects (not the infection) associated with many A-B type bacterial toxins. (2 points)
3. What amino acid is phosphorylated during the transport cycle of P-type ATPases? (1 point)

The End. Good luck with your careers!