

## **BMB623 EXAM II**

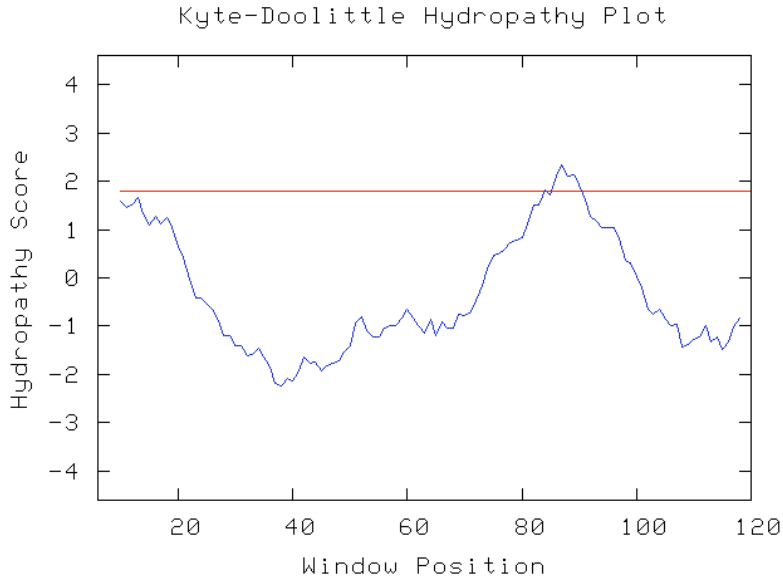
### **March 14, 2007**

- Place name on all pages.
- Show all reasoning and work clearly (if you want a chance to get partial credit even when your final answer maybe wrong).
- Letter each part of the answer.
- Exams will be collected at 11AM. Good Luck!

- 1. (21 pts total)** COPII and “COPI” vesicles are involved in protein trafficking in the secretory pathway.
- (A) Describe how the cargo selection process is controlled by COPII coat proteins. (6 pts)
  - (B) Describe how the budding process is controlled by COPII and its cofactors (diagram the coat recruitment process naming the components involved, 6 pts).
  - (C) How could you experimentally accumulate COPII vesicles for their isolation and characterization if you have a purified ER preparation to start with (3 pts)?
  - (D) What region of the “COPI” coat mediates membrane targeting (3 pts)?
  - (F) If you use “COPI” antibodies to immunostain a cell, what regions of the cell will be visualized? (3 pts).

2. (40 pts) (56) The murine protein Y was discovered and sequenced. It has the following sequence and corresponding hydrophobicity plot.

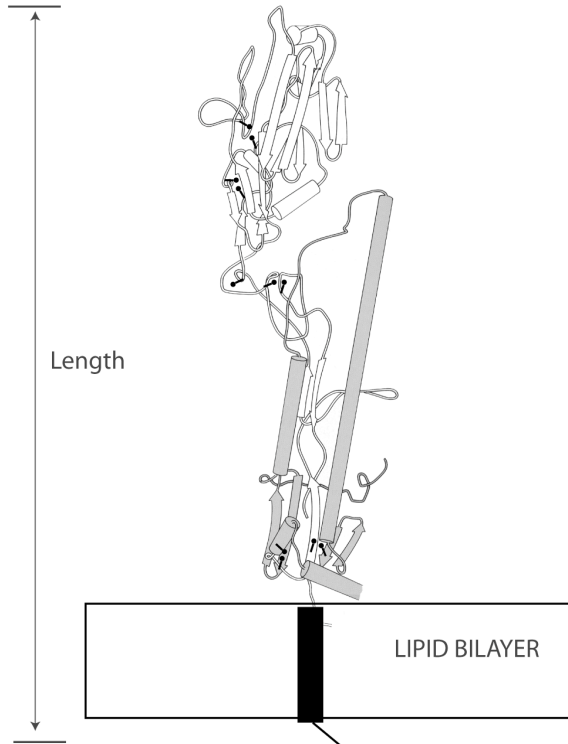
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mrkmpctll lllaallapt qtrgagagqt rnasagrprd rkaprrpgpg
msgpanwtyv yekrysgafp nqlqagglg aamvgavlta llaglvsikk
rgagagrprt rgagagrnpq lraqcaat
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- A)** Draw a cartoon of the expected topology of Protein Y using a rectangle to designate transmembrane alpha-helices. (10 pts)
- B)** Label the cytosolic and extracellular spaces on the diagram above and briefly explain below your reasoning for choosing this orientation. (4 pts)
- C)** Assuming all N-linked glycosylation and lipidation sites on this protein are fully recognized, circle and label all sites of modification on the protein sequence above. (6 pts)

- D)** Unprocessed Protein Y is 128 amino acids long how many amino acids long would the fully processed and modified protein be? (show work and briefly explain; 4 pts)
- E)** If the Arg at position 2 was altered to a Gly and all available modification sites are still occupied what would you expect to happen to the protein in a co-translational translocation and modification assay? (6 pts)
- F)** What is the first thing that you would do to get an idea about the possible function of Protein Y in the cell? (4 pts)
- G)** Expression of this mammalian protein in *E. coli* results in its misfolding, aggregation and accumulation in inclusion bodies. Explain why you should not be surprised by this result. (6 pts)

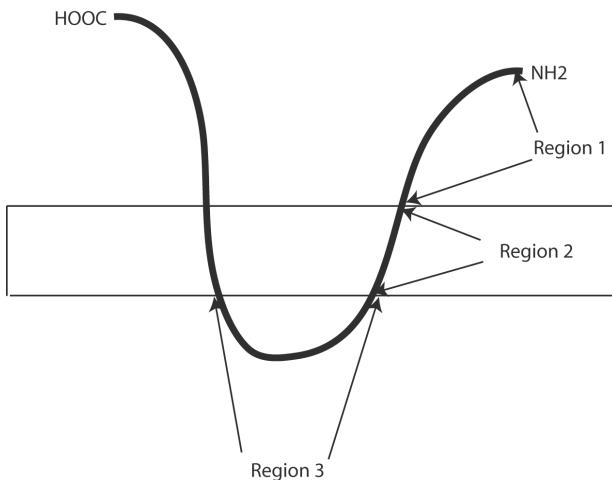
3. Using the following cartoon for *influenza* viral protein hemagglutinin (HA), estimate the length of the protein diagramed if the diagram is accurately drawn to scale? (Briefly explain your answer showing the work; 9 pts)



4. (10 pts) Maintaining the permeability barrier in the ER membrane is essential during protein translocation. For a polytopic protein this can be a complicated process. Two models for maintaining the permeability barrier were discussed in class.

A) Choice the model that you believe is most likely to be accurate and support your choice with a brief explanation as to why you believe this is the most accurate model. (4 pts)

B) Describe how this permeability barrier is maintained for the polytopic protein diagramed below when the following region just outside the ribosome and being delivered to its proper location. Both protein termini are localized within the cytoplasm in its final orientation (6 pts).



Region 1:

Region 2:

Region 3:

5) (20 pts) Define the following terms or answer the short question:

A) Detergent aggregation number.

B) Proteoglycan

C) Two methods for identifying the translocation of a secretory protein using the *in vitro* translocation assay established by Blobel.

D) UDP-Glucose: glycoprotein glucosyltransferase

E) SNARE hypothesis