Spring 2005 Molecular Biology Exam #2 – Applying Lessons

There is no time limit on this test, though I have tried to design one that you should be able to complete within 5 hours, except for typing. You are not allowed to use your notes, any books, any electronic sources except those specified in the exam, nor are you allowed to discuss the test with anyone until all exams are submitted on Monday March 21. **EXAMS ARE DUE AT 9:30 ON MONDAY, MARCH 21**. You may use a calculator and/or ruler. The answers to the questions must be typed unless the question specifically says to hand write the answer in the space provided. If you do not write your answers on the appropriate pages, I may not find them unless you have indicated where the answers are. There are 4 pages to this exam, including this cover page.

For the figures, I took photographs from journals, so you may detect warps or angles that seem odd. This is due to my need to reduce glare and shoot form about a 45 degree angle. Do not take this odd perspective into consideration for your answers. Ignore the tilted angles of the figures.

-3 Pts if you do not follow this direction:

6 pts.

- 1. Tell me how to make the following two solutions. You must show your work to receive partial credit for wrong answers.
- a. Make 275 mL of a 1.5% SDS, 1X TBE solution.
- b. Make 450 mL of 2M NaCl, 500 mM Tris, 0.01 M EDTA.

FWs: NaCl = 58.5; EtBr = 394; EDTA = 416; Tris = 121; HCl = 36.5; agarose = 204. Other raw materials include SDS = stock solution of 20%; TBE = stock solution that is 10X;

8 pts.

2. Identify this protein: CMAVMAIHLILLTAGTALLLIQVLNL. Tell me the name of the protein, the species from which it came, and what method you used to reach your answer. This question is open use of the internet.

10 pts.

3. Calculate the molecular weights of the two bands in figure 1. To receive credit for this answer, you must show me the graph you used to generate your answer. You may use Excel for this question. Use the 2 dash marks at the top as the edge of the wells in the gel.

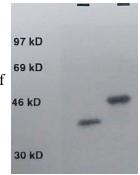
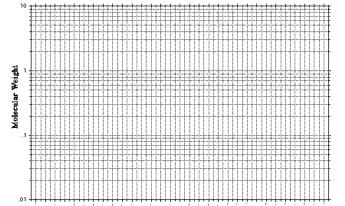


Figure 1.



16 pts.

- 4. A series of experiments were performed to understand what binds to the Tyro3 receptor on the surface of some cells. Left side: Cell expressing Tyro3 were bathed in different media contain various unknown components. When the receptor binds its ligand, it autophosphorylates. Right side: They engineered cells to secrete a soluble form of Tyro3 (i.e. no transmembrane domain; called sTyro3) or a soluble form of an unrelated receptor called TrkB (i.e. sTrkB).
- a. Describe the main point for the left side of this figure.
- b. What effect did sTyro3 have on phosphorylation of the normal Tyro3?
- c. What was the purpose of creating sTyro3?
- d. What effect did sTrkB have on phosphorylation of the normal Tyro3?
- e. What was the purpose of creating sTrkB?



12 pts.

5. Eventually, the investigators fond the protein that is the Tyro3 ligand and they very cleverly called it protein S. They cloned the protein S gene and transfected the protein S gene into COS cells. Describe the data in figure 3 and what the data tell us.

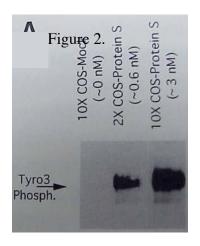


Figure 3.

10 pts.

6. Figure 4 shows two northern blots with the indicated probes. Assume that good RNA was loaded in every lane. Interpret these data taking into consideration the data from question 5. Do not describe every lane, just the main lesson or lessons from this figure. The abbreviations are: THYmus, SPLeen, LIVer, KIDney, LuNG, Skeletal Muscle, HeaRT, SKiN, INTestines, UTeRus, OVaRY, PLAcenta, TeSTes, Adult BRain. Support your conclusions by citing data.

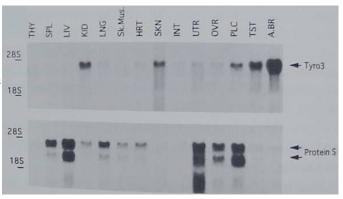


Figure 4.

12 pts.

7. Write down the two sequences that appear in figure 5, from 5' to 3'. After you have written down the two sequences on separate lines, Write the deleted base or bases missing in the mutant.

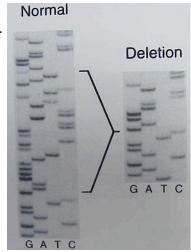
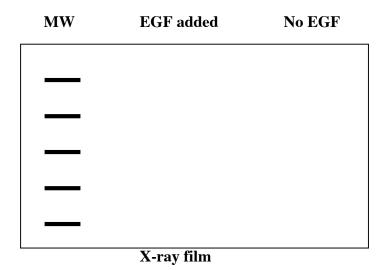


Figure 5.

16 pts.

8. Figure 6 shows what an investigator thinks the EGF receptor looks like when it has bound EGF (epidermal growth factor). When the ligand is bound, the heterotrimer of Grb2, SoS and Ras bind to EGFR.

Draw the results from an immunoprecipitation experiment if all the proteins were labeled with ³⁵S, separated by SDS-PAGE, and imaged via fluorography on X-ray film. Use the blank film below to draw (by hand or in Word) the data if the antibody used binds to Grb2.



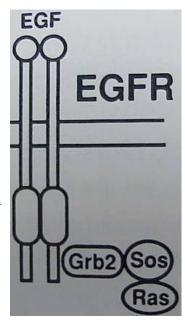


Figure 6.

10 pts.

9. A population of identical cells was measured by flow cytometry. The data are presented in Figure 7, with fluorescence intensity on the X-axis and number of cells on the Y-axis. Explain the data for the 5 surface proteins as labeled by fluorescent antibodies in 5 different experiments.

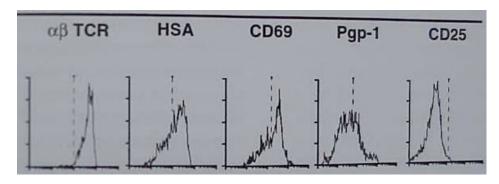


Figure 7.