

2002 Molecular Biology Exam #3 – Final Exam

There is no time limit on this test, though I have tried to design one that you should be able to complete within 4 hours, except for typing. You are not allowed to use your notes, any books, or electronic sources, nor are you allowed to discuss the test with anyone until Monday May 13, 2002. **EXAMS ARE DUE AT 9 AM ON MONDAY, May 13.** You may use a calculator and/or ruler. The answers to the questions must be typed.

Once again, I took photos of figures, so do not interpret any slant or distortion of the figures. Poor quality of figures is due to my photography and the need to avoid glare. You may type directly in this file and print your answers, or you may type your answers in a separate file. Just keep your answers in order and numbered. I want a hard copy of your test answers.

-3 points if you do not follow this direction:

Please do not write or type your name on any page other than this cover page. This test is 8 pages long, including this cover sheet.

Name (please print here):

Write out the full pledge and sign:

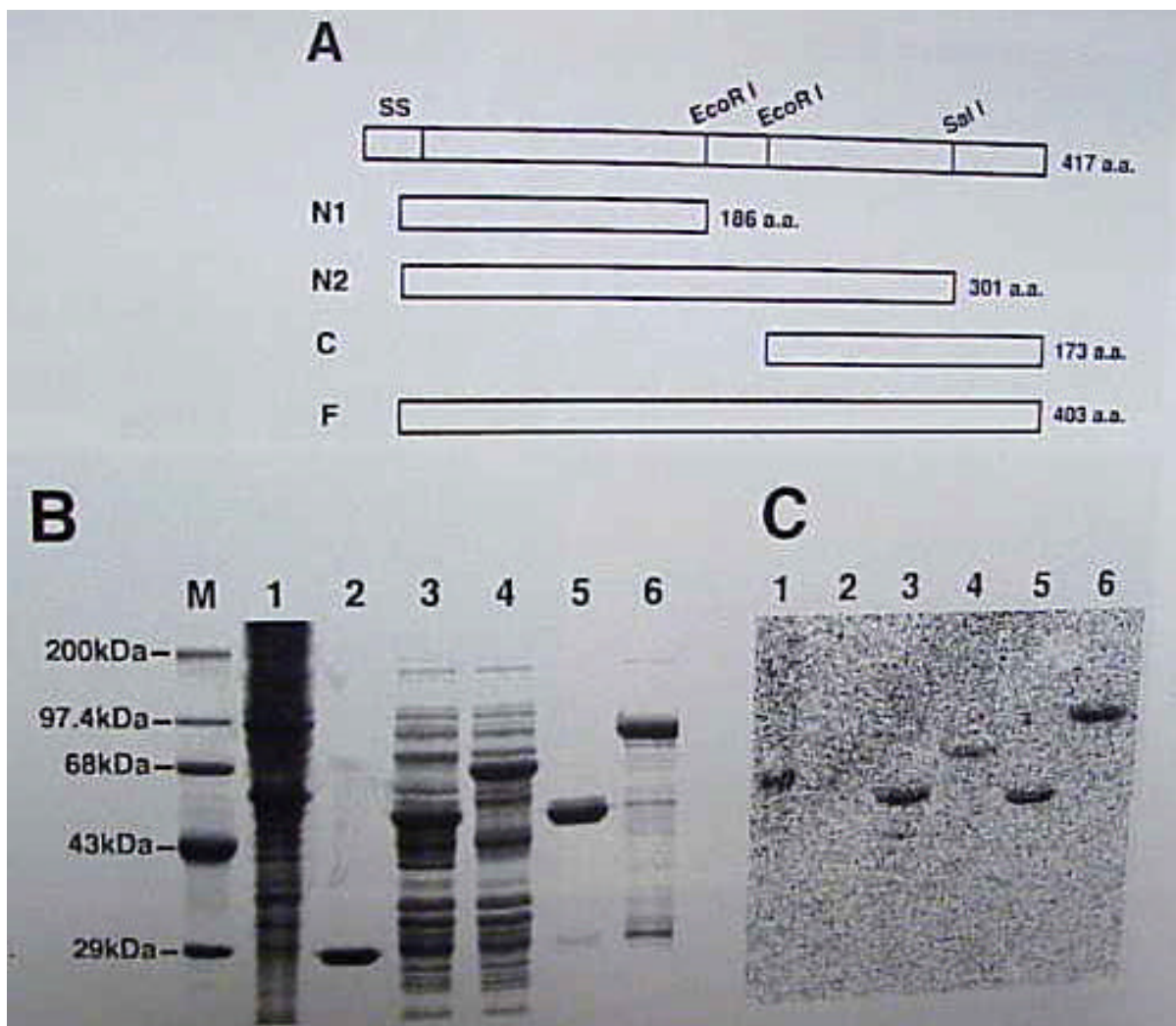
How long did this exam take you to complete (excluding typing)?

As it turns out, very little is what it seems to have been. For example, the original eggplant produced a white fruit and was later selected for a fruit that produced fewer seeds. As a side effect of fewer seeds, the fruit turned purple, but the name of eggplant remained. Another example is Craig Venter who had claimed to sequence DNA isolated from several people but in fact, he only used his own DNA.

The following questions appear to be very difficult, but in fact they are not. Please provide me with perfect answers so I do not have to buy a new red pen.

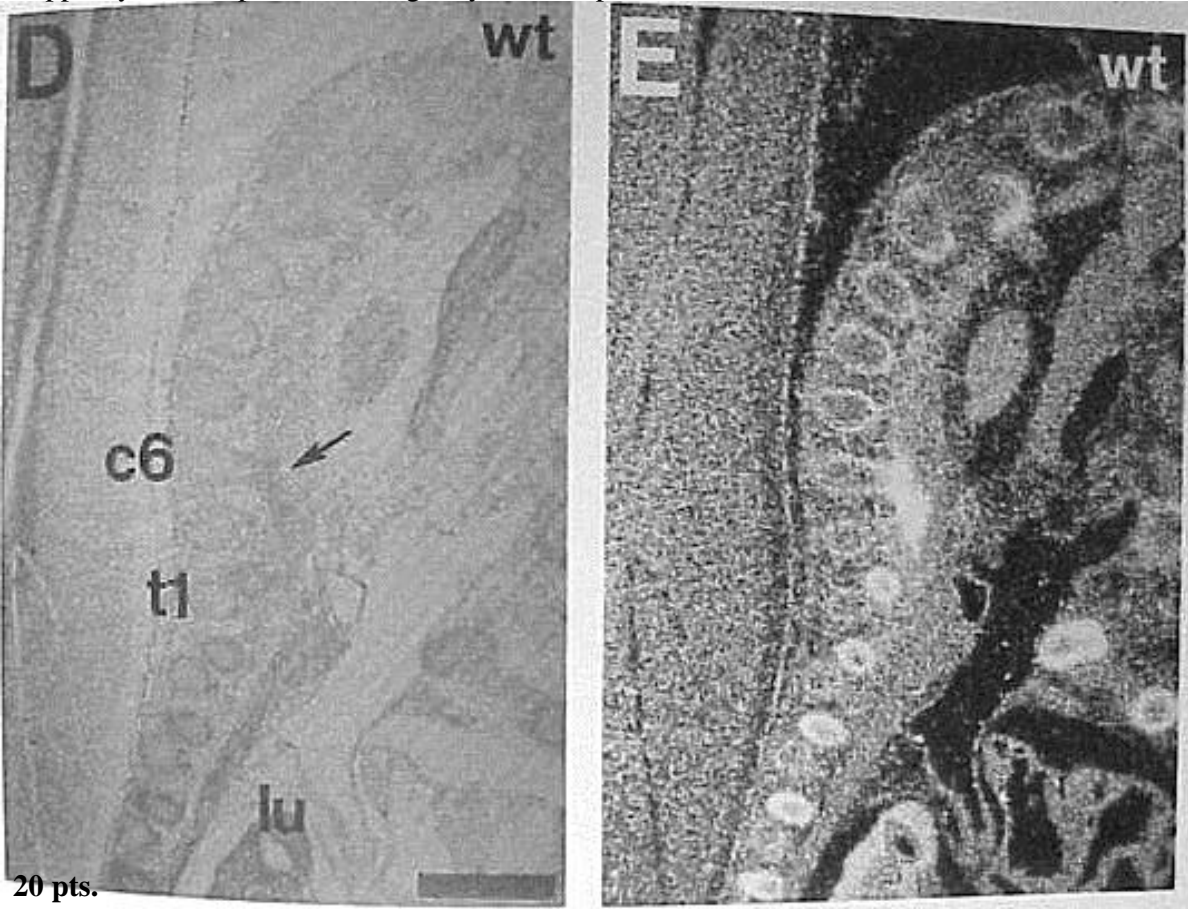
10 pts.

1. Figure 1 examines the calcium binding properties of the protein called calsequestrin. Panel A shows the full length protein of 417 amino acids. Below this are 4 truncated proteins that were produced in *E. coli* fused to an epitope tag called GST. Panel B is an SDS-PAGE stained for all protein with a Coomassie Blue dye. Panel C is an autoradiogram of the same gel that had been incubated with radioactive ⁴⁵Ca. The lanes in the gel are: M=MW, 1= rat heart SR, 2=GST protein, 3= GST+N1 construct, 4=GST+N2 construct, 5=GST+C, and 6=GST+F. Interpret this figure with special attention paid to panel C.



10 pts.

2. Figure 2 shows two micrographs from a developing mouse embryos. The section shows the developing vertebrae, some ribs, etc. Panel D is a bright field shot and E is a dark field shot of the same section. The investigators performed an *in situ* hybridization on this section (BMP5 anti-sense probe was used in panel E). The point these authors wanted to make was that the gene BMP5 is expressed near the region denoted by the arrow in panel D. Do you believe them? Support your interpretation using only the data presented here.

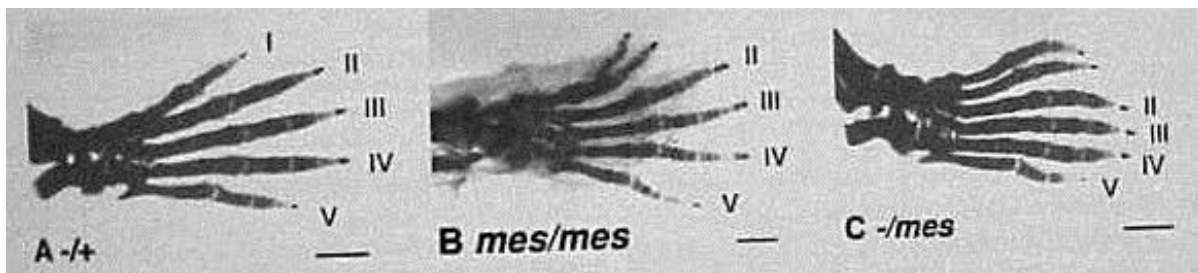


20 pts.

3. Below is a figure that should dispel the misconception that the number of toes or fingers is determined by the number of cells. These are the front paws of 3 (not blind) mice with genotypes shown below each photo. a) What type of mutation is *mes*?

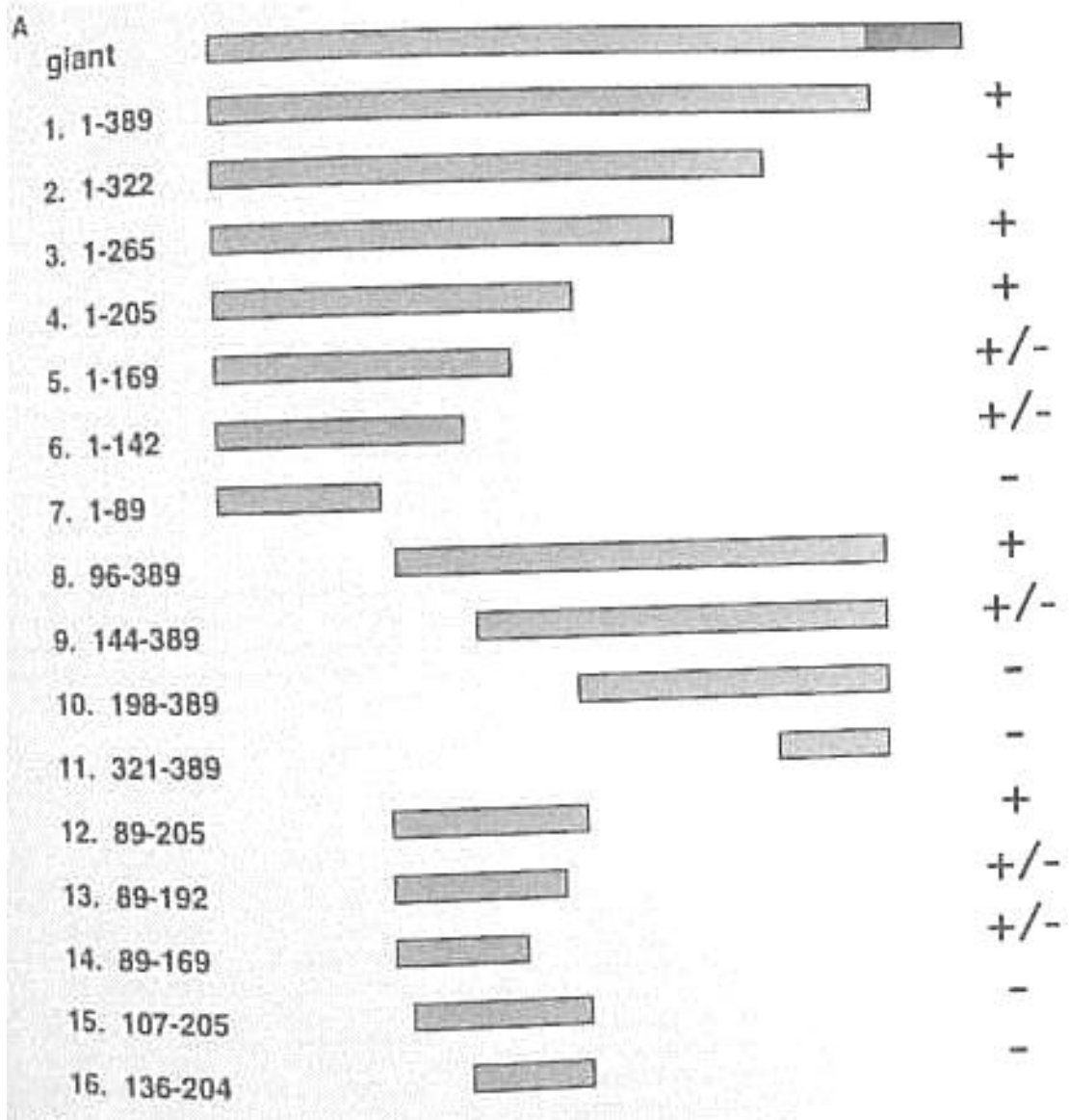
b) Propose a mechanism to explain this genotype/phenotype relationship (i.e. how can this mutation produce this phenotype?).

c) Design an experiment to test your hypothesis.



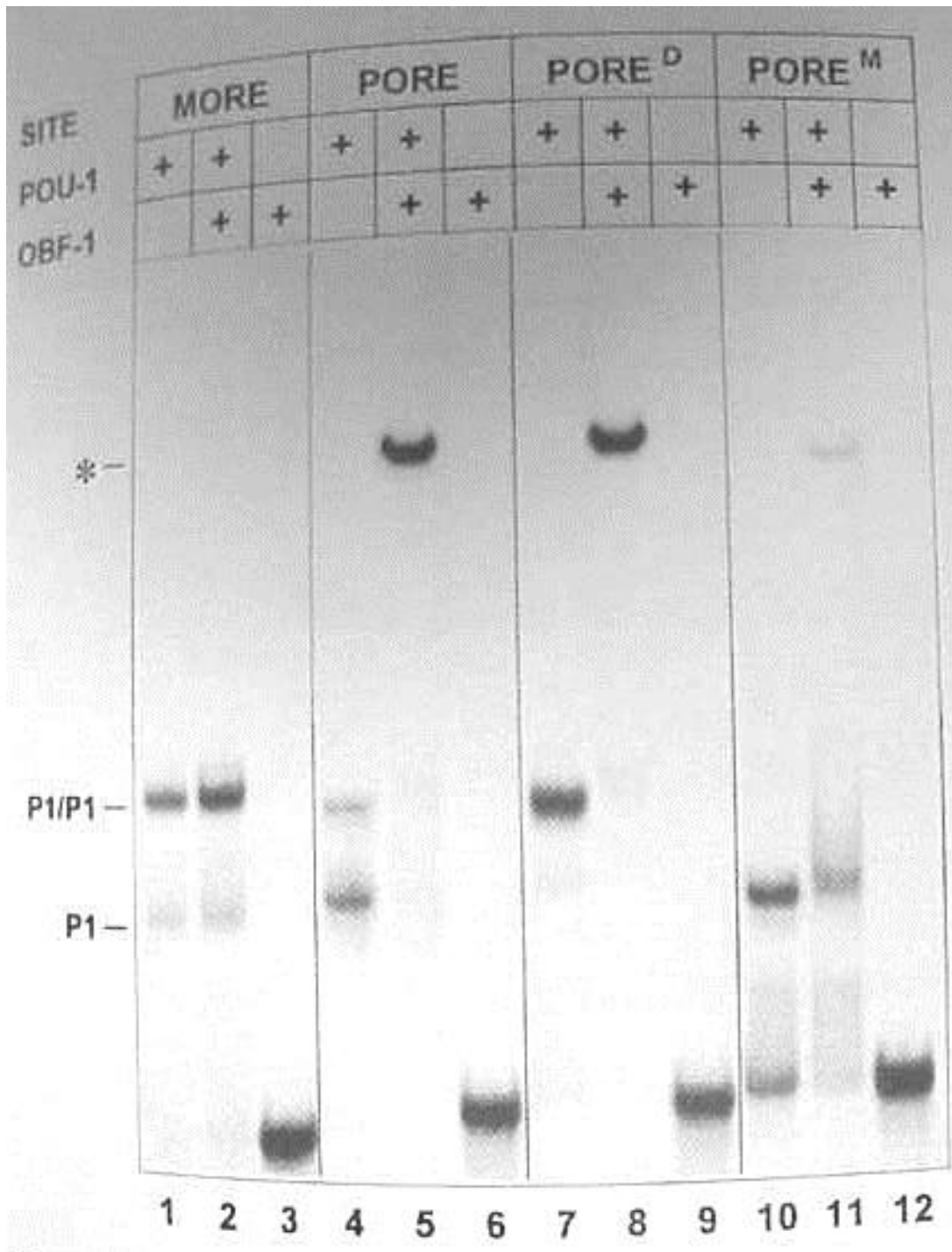
10 pts.

4. Below is a series of cartoons showing mutant forms of the fly “giant” protein. In the far right column, + indicates normal function and – indicates lack of normal function. The numbers in the far left column indicate which amino acids are present in each construct. Interpret this figure as fully as you can. You do NOT need to interpret each construct, just summarize the main conclusion or conclusions that can be made from this figure.

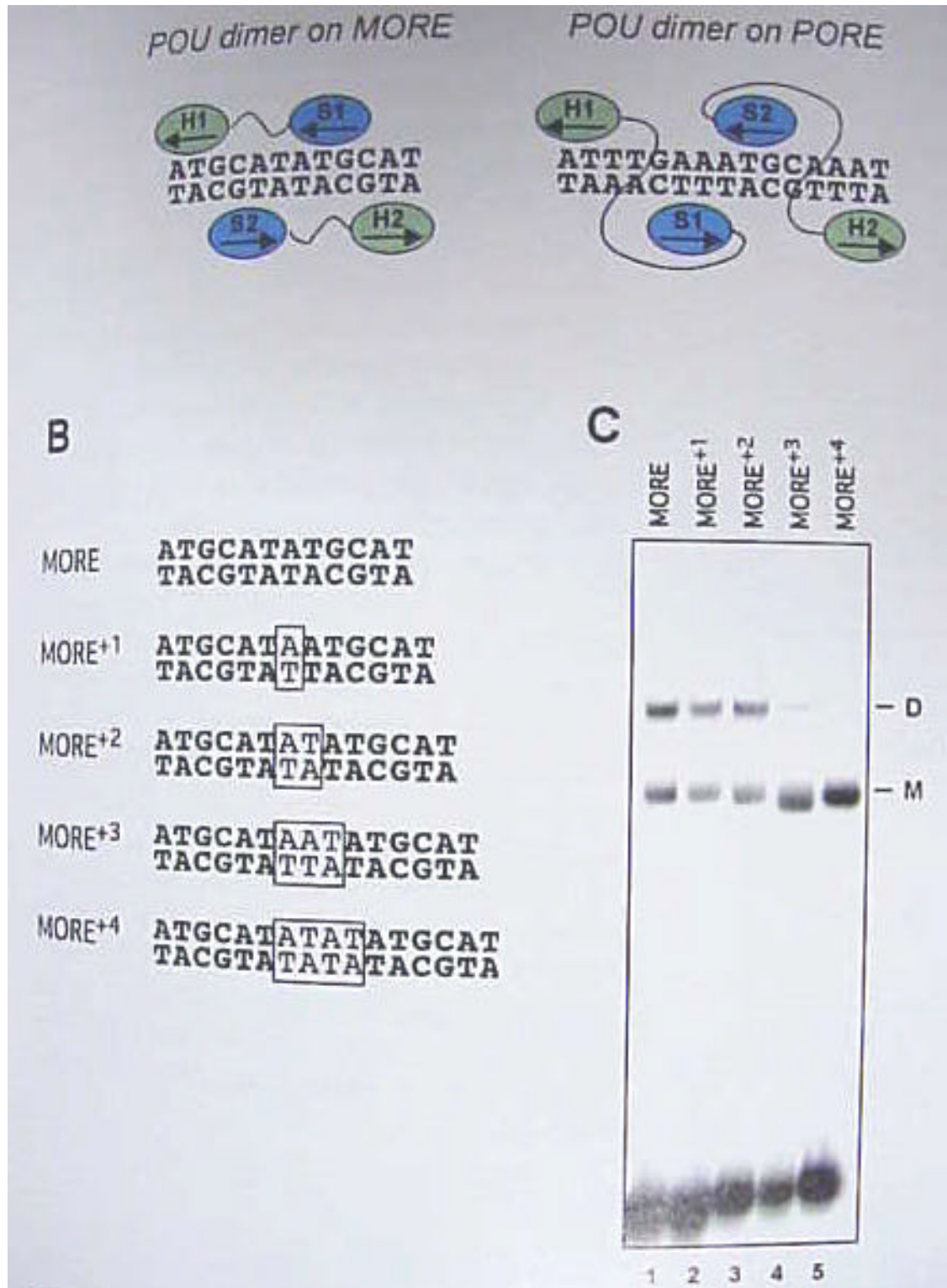


30 pts.

5. This question has two figures. The first figure shows a series of band shift assays using the MORE and PORE DNA sequences. POU-1 is a transcription factor, as is OBF-1.



In this second figure, we can see the binding sites for POU-1 on MORE and PORE DNA sequences. In panel B, we see the *wt* MORE sequence plus 4 mutants of MORE. Panel C shows another series of band shift results. Incubated with each DNA was POU-1 and a mixture of other transcription factors. The D and M in this figure are unrelated to the D and M in the previous figure.

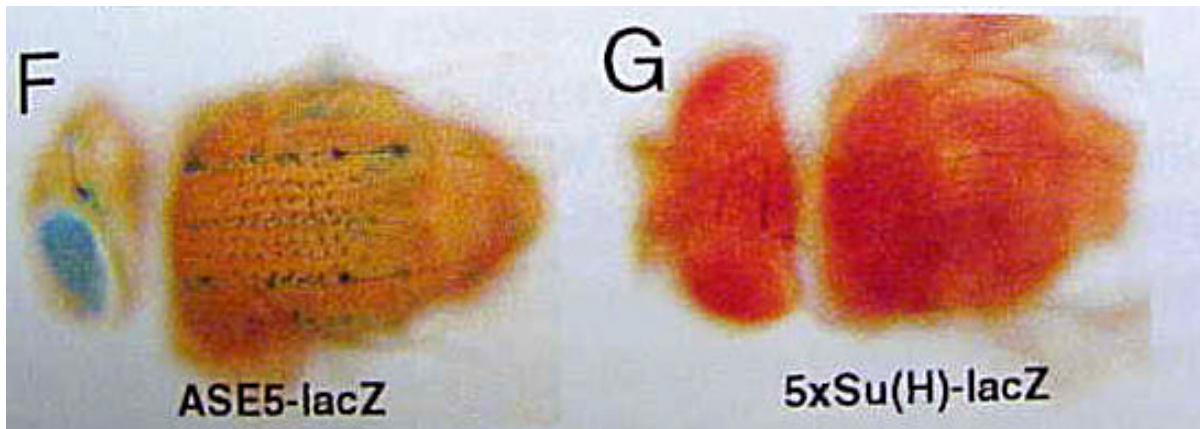


- Summarize the conclusions for the first band shift assay.
- Summarize the conclusion for panel C in the second band shift experiment.
- Given the model of POU-1 binding to MORE and PORE, explain what is happening in panel C.
- Design an experiment to test your hypothesis.

15 pts.

6. Below are two color photographs of two different transgenic flies. On the left, the promoter and enhancer (called ASE5) of Su(H) (suppressor of hairy) was placed upstream of the *lacZ* gene. On the right is another transgenic fly that contains a truncated version of ASE5 (called 5xSu(H) upstream of *lacZ*. Su(H) binds to ASE5 in 5 different places. 5xSu(H) retains all 5 binding sites.

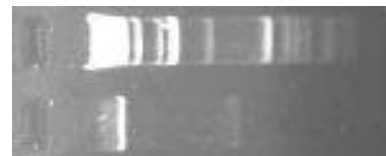
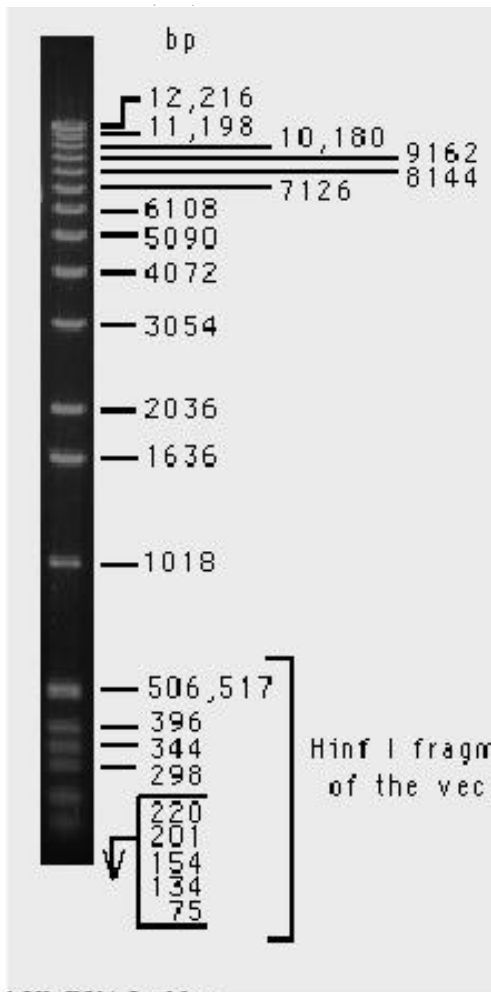
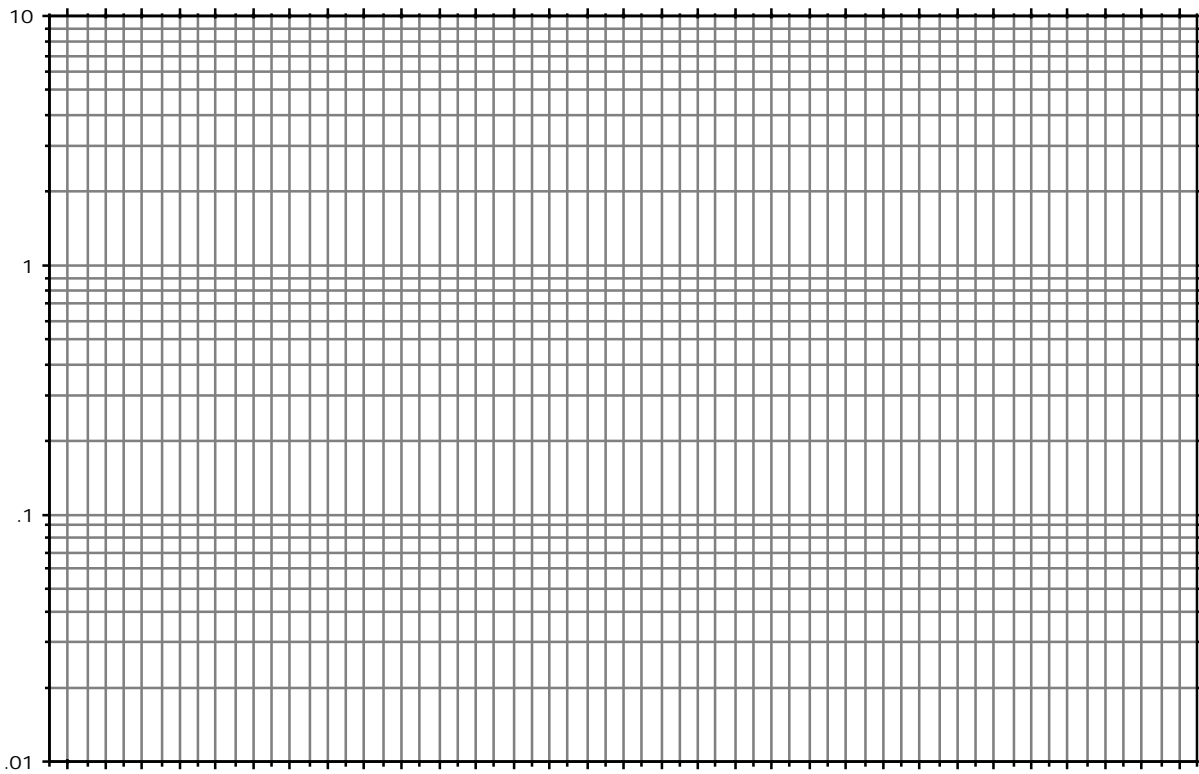
- What effect does Su(H) have on *wt* ASE5?
- Where is Su(H) expressed normally?
- What can you conclude from panel G if you know that Su(H) is still able to bind to all 5 of its sites in 5xSu(H)?



5 pts.

7. Determine the molecular weight of the band in the gel next to the “*”. To get any credit for this question, you must provide a graph to show how you got your answer. You may use the graph provided or you may create one in Excel.

Molecular Weight



1 Kb DNA Ladder
0.5 µg/lane
0.9% agarose gel
stained with ethidium bromide