Dr. Reichler's Bio 325 University Extension TTh 7^{:30}-9pm Print Name: Exam #2 October 23, 2008

Read each question carefully and don't hesitate to ask if a question seems unclear. If possible, answer each question in the space provided, but if needed, continue on the back. If you use a drawing as part of your answer, be sure to also include a written explanation. These questions have specific answers, although for some, more than one answer is possible. To receive full credit you must clearly and fully answer the question being asked. This exam is worth 103 points with the points for each question noted in parentheses.

1. You are investigating the molecular basis for the differences in prairie and montane vole reproductive behavior. You determine that both species contain the same amount of oxytocin receptor mRNA. Given this data, what might explain the difference in the two species' behavior? (10 pts) *The difference must be post-transcriptional. One species may have a protein or miRNA that inhibits translation. The receptor protein may be inactivated or have a short half-life.*

2. You are able to determine that the DNA from one bone marrow cell has a greater mass than another bone marrow cell. Both cells have the same number of nucleotides. Which one is more likely to be totipotent? Why? (8 pts)

The cell with lighter DNA. The loss of totipotency is marked by an increase in DNA mathylation that would add mass to the DNA.

3. In a eukaryotic cell, how could expression of a gene decrease the amount of gene product for another gene? In other words how could the expression of gene T cause a decrease in the quantity of the gene Q product? (8 pts)

It could code for a protein or miRNA that inhibits translation. OR It could code for a protein that inhibits or inactivates a transcription factor. OR It could code for a protein that methylates the DNA or deacetylates the nearby histones.

4. How could the proteins produced from a single gene have more than one final localization in the cell? (8 pts)

Alternate splicing could produce different versions each with a different mRNA or protein localization signal.

5. You transform a human cell with a reporter gene attached to the promoter for α -hemoglobin gene. Under conditions that you know induce expression of α -hemoglobin, you cannot see any of the reporter protein. Why is this surprising and why has it occurred? (8 pts)

The reporter gene has the α -hemoglobin promoter, and it should be expressed when the α -hemoglobin is expressed. When the cells were transformed with the reporter gene, it may have been inserted into the DNA in a region that is tightly packaged, and therefore nor expressed even in the presence of the appropriate transcription factors.

6. An mRNA is localized to a specific region of a cell. Design an experiment to eliminate **one** of the three hypotheses for how mRNA can be localized to a specific location in a cell. (8 pts) *mRNA is localized by its transport via the cytoskeleton. Disable cytoskeleton transport and see if mRNA distribution is affected. OR mRNA is localized by random diffusion and localized degradation. Disable mRNA degradation, and see if mRNA distribution is affected.*

7. Humans and chimpanzees have approximately the same number of nucleotides in their DNA.

Would you expect to find more pseudogenes in humans or chimpanzees? Why?(8 pts) *Humans have more transposons and more active transposons. The movement of a transposon into a gene could cause it to no longer function and lead to the formation of a pseudogene.*

8. You want to genetically engineer bacteria to produce one of the human *hox* proteins. What human cells would you use as your source material, why would you use these cells, what technique would you use to prepare the human gene for expression in bacteria, and why do you need to perform this technique to express the *hox* protein in bacteria? (8 pts)

Early embryoss. They express thehox genes so you can get hox mRNA for use in reverse transcription that will produce cDNA, which is the coding region sequence without introns. Bacteria cannot splice introns.

9. Are the genes that determine whorl identity in flowers the first developmental genes to be expressed during flower formation? Why or why not? (8 pts) *No, other genes must determine the number of whorls first.*

10. We saw that many *hox* proteins share similar sequences having to do with DNA binding. What other similarity would you expect to find in *hox* proteins? Why? (8 pts) *A nuclear localization signal. Transcription factors must be in the nucleus to function.*/

11. You hope you have inserted a gene into a plasmid, but you **cannot** use blue/white screening to check. How would you test whether your plasmid had an inserted gene or not? (10 pts) *Any of: PCR with primers specific for the inserted gene; successful amplification will show the presence of the inserted gene. Sequence the plasmid and see if the inserted gene is present. Cut the plasmid with restriction enzymes and see if a band the size of your inserted gene is produced.*

12. You perform PCR with primers for a gene and run the results on a DNA gel, but instead of amplifying one band of DNA, you get two. Why? (see illustration)

You <u>cannot</u> see the few longer than desired PCR products. There are only a few of them compared to billions of correctly sized bands. (8 pts)

expected result actual result

The primers were not specific and bound to more than one region of DNA amplifying multiple regions of the DNA.

Bonus: You cut a region of two people's DNA with the same restriction enzyme and get different patterns. Later you are able to get the extract the DNA sequence of this region, and you find that there is no difference between these individuals in the restriction enzyme recognition sequence. Why did their DNA produce two different patterns? What might have caused this difference in their DNA? (3 pts)

The number of nucleotides between the restriction sites was different. This could be due to a transposon or other DNA rearrangement.