Name _____ Date _____

CHAPTER 1 QUESTIONS

1. Why would it be beneficial to use a micropipette to measure reagents in biotechnology rather than another measuring instrument?

2. What do the results of gel electrophoresis tell you about genetic material?

Name _____ Date _____

CLONE THAT GENE QUESTIONS

1. Which restriction enzyme did you choose? Why did you choose that one?

2. Where would you insert the insulin gene, and why?

3. Which antibiotic would you use to determine if the recombinant DNA was taken in?

Name _____ Date _____

CHAPTER 2/2A QUESTIONS

1. List in words or indicate in a drawing the important features of a plasmid vector that are required to clone a gene. Explain the purpose of each feature.

2. What role do restriction enzymes have in nature?

3. Using your understanding of evolution, why would bacteria retain a gene that gives them resistance to antibiotics? How is the existence of bacteria with antibiotic resistance affecting medicine today?

Name _____ Date _____

4. Bacteria, sea anemones, and humans seem, on the surface, to be very different organisms. How can a gene from humans or a sea anemone be expressed in bacteria to make a product never before made in bacteria?

5. Due to a mishap in the lab, bacteria carrying a plasmid with a kanamycin-resistant gene and bacteria carrying a plasmid with an ampicillin-resistance gene were accidentally mixed together. How would you design an experiment allowing you to sort out the two kinds of bacteria? (Hint: Make sure that you do not kill off one of the kinds of bacteria you are trying to sort out!)

Name _____ Date _____

CHAPTER 3 QUESTIONS

1. What role do DNA ligases have in nature?

2. What role do DNA ligases have in gene cloning?

3. What properties of the DNA restriction fragments produced in Laboratory 2 enable ligation of these fragments?

4. Could two rfp fragments join to form a plasmid during the ligation? If not, what would prevent that? If so, what would be the result?

5. During ligation, both hydrogen and covalent bonds form. Which bonds form first? Why do both types of bonds need to form?

Name _____ Date _____

CHAPTER 4 QUESTIONS

1. Why is it important to verify that you have the correct recombinant plasmid?

2. How did your actual gel results compare to your gel predictions?

3. Do you see any bands that were not expected? What could explain the origin of these unexpected bands?

4. Does the gel show that your restriction digest and ligation procedures were successful? Describe the evidence you used to make this assessment.

5. In the geK– and geA– lanes, do you see evidence of multiple configurations of plasmids? Explain your answer.

Name _____ Date _____

6. In the geK+ and geA+ lanes, do you see evidence of complete digestion? Explain your answer.

7. In which lane would you expect to find the *rfp* gene and the *ampR* gene in the gel photograph? Are you able to locate these two genes? Explain your answer.

8. Compare the lanes that have linear fragments with the lanes that have plasmids. Is there a difference in the shape of the bands between these two DNA forms?

9. In Laboratory 3, you described all the possible plasmids that you could make by ligating the digested fragments of the pKAN-R and the pARA plasmids. Two of the *rfp* gene fragments (807 bp each) may form a circularized fragment because each end of the fragments terminates in *Bam*HI and *Hin*dIII sticky ends. Is there evidence of a circularized 1,614 bp fragment in the geLIG tube lane? Explain your answer.

Name _____ Date _____

CHAPTER 5 QUESTIONS

1. Look at the results of your transformation. Do your actual results match your predicted results? If not, what differences do you see, and what are some explanations for these differences?

2. How many red colonies were present on your LB/amp/ara plate?

3. Why did the red colonies appear only on the LB/amp/ara plate and not the LB/amp plate?

Name _____ Date _____

4. Recombinant plasmids are engineered so that they can replicate in the cell independently of the chromosome replication. Why is it important to have multiple copies of a recombinant plasmid within a cell?

5. How is the information encoded in the *rfp* gene expressed as a trait? Be sure to use what you have previously learned about gene expression and the relationship between DNA, RNA, protein, and traits.

6. Why is it possible for bacteria to make a human protein, such as insulin, or a sea anemone protein, such as RFP?

7. The only bacteria that could produce the RFP in Laboratory 5 were bacteria that were transformed with the pARA-R plasmid. Why?

Name _____ Date _____

CHAPTER 6 QUESTIONS

1. Why is a protein's conformation important for carrying out its function?

2. What properties of the amino acids in a protein relate to protein folding?

3. Does the eluate containing your RFP appear less bright or brighter than it did in the cell lysate following centrifugation? If there is a noticeable difference in the intensity of the red color, what might account for that?

4. What characteristic of RFP is used as the basis for separation by column chromatography?

5. How might the column chromatography procedure be adjusted or modified to increase the purity of the RFP sample?