Unit 7 (Gene Regulation & Biotechnology) Review Packet – ANSWER KEY

AP Biology

Topic #1: Gene Regulation and Development

With regard to the operon pictured to the right, the image on top shows the operon in its normal state, and the image on the bottom shows the operon in the presence of molecule #5 (looks like a + sign).

1. Identify the different parts of the picture. Your options are repressor, promoter, genes of the operon, operator, RNA polymerase, and inducer.

- 1. RNA polymerase
- 2. Repressor
- 3. Promoter
- 4. Operator
- 5. Inducer
- 6-8. Genes of the operon

2 What type of operon is shown—inducible or repressible—and how do you know?

This is an inducible operon. In its normal state it is turned off (RNA polymerase cannot bind to the promoter correctly to begin transcription of the genes of the operon). In the presence of the inducer molecule, the operon can be turned on.

3 What is the role of molecule #5 in regulating the operon shown above?

In the presence of the inducer molecule (#5), the repressor is inactivated and cannot bind to the operator, allowing RNA polymerase to bind correctly to the promoter and begin transcription of the genes of the operon.

4. Why is a catabolic operon (one that contains genes for enzymes used to break down molecules) usually an inducible operon?

It is usually turned off but can be turned on in the presence of a particular molecule in the environment that must be broken down (ex: lactose sugar). We don't need to break down the partiular molecule unless it's present.

5. Why is an anabolic operon (one that contaisn genes for enzymes used to build molecules) usually a repressible operon?

It is usually turned on because it is used to synthesize an essential molecule (ex: tryptophan), but it can be turned off if the essential molecule is present in high concentrations.

6. Let's say methyl groups are added to the DNA of the gene coding for human growth hormone. How will this affect the amount of human growth hormone produced?

Methyl groups cause DNA to coil tightly, preventing RNA polymerase from binding to the promoter to begin transcription of the gene. This will decrease the amount of gene expression and thus the amount of human growth hormone produced.

7. Let's say acetyl groups are added to the histone proteins that interact wth the DNA of the gene coding for human growth hormone. How will this affect the amount of human growth hormone produced?

Acetyl groups cause DNA to coil less tightly, allowing RNA polymerase to bind to the promoter to begin transcription of the gene. This will increase the amount of gene expression and thus the amount of human growth hormone produced.





8. How is the regulation of gene expression different for prokaryotic cells vs. eukaryotic cells?

Prokaryotic cells only regulate gene expression at the transcription level (either preventing or facilitating the binding of RNA polymerase to the promoter of an operon). Eukaryotic cells regulate gene expression at many different levels of protein synthesis—including DNA structure, transcription, post-transcription (mRNA processing), translation, and post-translation.

9. How can changes at the level of mRNA processing (after transcription) produce totally different proteins (ex: variations of the different antibody proteins that are targeted to attack specific bacteria or viruses?

Remember, this is considered Post-Transcriptional modification of gene expression. Splicing (i.e. cutting) out the introns in different ways / combinations can produce multiple proteins from the same gene. (Note: ALL of the introns are ALWAYS removed from Pre-mRNA, but some of the exons may be removed as well for the reason stated above).

10. Explain the role of homeotic genes in pattern formation during embryonic development.

Homeotic genes code for major transcription factors that regulate genes involved in the development of the basic body segments. Homeotic genes determine what body region develops where (head, limbs, abdomen, etc). Homeotic genes are found in many animals (ex: fruit flies, mice, humans), indicating they were found in a common ancestor.

11. Explain the role of apoptosis in morphogenesis during embryonic development.

Apoptosis (programmed cell death) is used to sculpt / shape the body structures during the second step of embryonic development—morphogenesis. During programmed cell death, enzymes are activated that break apart the structures and molecules of the cell. These broken-down cell "chunks" are swallowed and further digested by a macrophage (a "big eater" cell).

12. Explain the role of embryonic induction in cell differentiation during embryonic development.

In embryonic induction, cells of one tissue send a signal molecule to a nearby group of cells to induce them to become a particular cell type. (For example, cells of the eye socket signal nearby cells to become the lens of the eye.)

The Hedgehog protein (Hh) plays a critical role during a certain period of embryo development. As illustrated in the figure to the right, when Hedgehog is present, it binds to membrane proteins Ptch and Smo. Activated Smo interacts with a complex of proteins, which eventually results in the activation of a Gli transcription factor that stimulates the transcription of target developmental genes in the nucleus. The image on the left shows the signaling pathway when Hedgehog is not present.

13. Suppose a scientist injects a compound that prevents the activation of Smo in the presence of the hedgehog protein. How will this affect the amount of transcription of the genes in the nucleus?

If Smo is blocked, Gli will not be activated so it cannot be used as a transcription factor to stimulate transcription of the genes in the nucleus. Transcription of these genes will decrease or stop altogether.



Two different genes are known to be involved in the development of different body regions of *Drosophila* fruit flies. The diagram below shows the distributions and levels of mRNA transcribed from two different genes (bicoid and caudal) in different locations in a *Drosophila* egg immediately before fertilization. Note: *bcd* stands for bicoid mRNA and *cad* stands for caudal mRNA



The diagram below shows the distributions and levels of the two corresponding proteins along the body after fertilization. Note: BCD stands for bicoid protein and CAD stands for caudal protein.



14. How will the removal of caudal mRNA affect the distribution of bicoid protein in the fertilized egg?

Caudal mRNA and protein do not appear to affect the distribution of bicoid protein. If caudal is removed, bicoid protein should still be translated in the same location bicoid mRNA was found in the unfertilized egg (i.e. the anterior region). Bicoid protein, however, does affect the translation of caudal mRNA and prevents caudal mRNA from being translated in the anterior part of the egg (the same part of the egg as the bicoid).

15. The image to the right shows the regulation of transcription of the albumin gene in liver cells vs. brain cells. Based on the diagram, why is the albumin gene transcribed at a higher level in liver cells?

In liver cells (but not brain cells), specific activator molecules (called regulatory transcription factors in this image) are present that can bind to the enhancer region and increase transcription of the albumin gene.



Topic #2: Biotechnology

16. Explain the purpose of each of the three factors in polymerase chain reaction (PCR).

- 1. Heat: "Denatures" (i.e. separates) the two strands of the DNA double helix by breaking the hydrogen bonds between complementary nitrogen bases.
- 2. Primers: Short sequences of DNA nucleotides that anneal (stick) to the beginning of the target sequence and tell Taq polymerase where to begin replicating the target sequence.
- 3. Taq polymerase: Brings in free nucleotides to match up with the nucleotides of the target sequence to create copies of the target sequence.

17. Which person in the DNA fingerprint shown to the right—Bob, Sue, John, or Lisa—matches the blood stain DNA? How do you know?

John ; his pattern of restriction fragments (bands) matches with the blood stain pattern.

18. If the "wells" of the gel are located up at the top, which DNA fragment is larger—A or B? How do you know?

Fragment A is larger because it migrates more slowly towards the far end (the positive end) of the gel.



Below is a plasmid with restriction sites for BamHI and EcoRI. Several restriction digests were done using these two enzymes either alone or in combination. Use the figure to answer questions 13-15



Hint: Begin by determining the number and size of the fragments produced with each enzyme. "kb" stands for kilobases, or thousands of base pairs.

19. Which lane shows a digest with BamHI only? Explain your answer.

If only BamHI is able to make "cuts" on the plasmid, it will produce fragments of the following sizes – 2 Kb, 6 Kb, and 12 Kb. Therefore, Lane III shows a digest with BamHI only.

20. Which lane shows a digest with EcoRI only? Explain your answer.

If only EcoRI is able to make "cuts" on the plasmid, it will produce a single fragment with a length of 20 kb. Therefore, Lane II shows a digest with EcoRI only.

21. Which lane shows a digest with both BamHI and EcoRI? Explain your answer.

If both BamHI and EcoRI are able to make "cuts" on the plasmid, it will produce fragments of the following sizes – 2 Kb, 6 Kb, 8 Kb, and 4 Kb. Therefore, Lane IV shows a digest with both BamHI and EcoRI.

In a lab experiment that WORKED, scientists transformed *E. coli* bacteria with a plasmid containing the gene for ampicillin resistance (ampR) and the gene to enable the bacterium to glow (pGlo). The pGlo gene is typically turned off but can be turned on in the presence of the sugar arabinose (ara). The scientists attempted to grow cultures of this transformed bacteria in three conditions—plain LB agar (bacteria food), LB / amp, and LB / amp / ara. They then attempted to grow cultures of untransformed bacteria (lacking the plasmid) in the same three conditions. The table below summarizes all the treatment groups.

	LB	LB / amp	LB / amp / ara
<i>E. coli</i> with plasmid	1	2	3
-			
E. coli without plasmid plasmid	4	5	6

22. For each plate, state whether there will be bacterial growth, the type of growth (lawn or colonies), and whether the bacteria will glow. Provide an explanation for your answers.

Plate	Growth?	Lawn or	Glow?	Explanation
	(yes or no)	Colonies?	(yes or no)	
1	Yes	Lawn	No	Bacteria will have uninhibited growth on this plate because it contains plain LB agar (bacteria food)
2	Yes	Colonies	No	Only successfully transformed bacteria will grow on a plate with ampicillin. Bacteria that have not been successfully transformed will die (because they do not have the plasmid / gene for ampicillin resistance), which explains the blanks spots around the colonies.
3	Yes	Colonies	Yes	Only successfully transformed bacteria will grow on a plate with ampicillin. Bacteria that have not been successfully transformed will die (because they do not have the plasmid / gene for ampicillin resistance), which explains the blanks spots around the colonies. In the presence of the sugar arabinose, the bacteria in the colonies will glow.
4	Yes	Lawn	No	Bacteria will have uninhibited growth on this plate because it contains plain LB agar
5	No	N/A	N/A	The bacteria cannot grow on the plate because it contains ampicillin, and the bacteria have not been transformed with the plasmid containing the gene for ampicillin resistance.
6	No	N/A	N/A	The bacteria cannot grow on the plate because it contains ampicillin, and the bacteria have not been transformed with the plasmid containing the gene for ampicillin resistance.

23. List the steps involved in creating human insulin protein through using recombinant DNA.

- 1. Remove the bacterial plasmid from the bacteria.
- 2. Cut the human insulin gene and a bacterial plasmid with the same restriction enzyme
- 3. Allow the sticky ends of the human insulin gene and the bacterial plasmid to hydrogen bond to each other.
- 4. Seal the sticky ends of the human insulin gene and the bacterial plasmid together with DNA ligase
- 5. Induce a bacterium to take in the recombinant plasmid ("transform it") by putting it in a CaCl₂ solution and heatshocking it.
- 6. Allow the bacterium to transcribe and translate its plasmid DNA as well as the human insulin gene. Extract the human insulin protein that is created.

24.Golden rice is a transgenic plant, meaning it contains a gene from another organism. In this case, it has been given the gene for the creation of beta carotene (vitamin A). How can a bacterium be used as a "vector" to insert the beta carotene gene into the golden rice plant cells?

- 1. Perform steps 1-5 above using the gene for the creation of beta carotene (vitamin A) and a plasmid from a bacterium that infects plants (ex: Agrobacterium tumefaciens).
- 2. Allow the bacterium to infect a plant cell (in this case a rice cell), and insert its plasmid DNA and the beta carotene synthesis gene into the plant to be incorporated into the plant's genome and then be transcribed and translated by the plant. This allows rice to be able to produce and therefore contain beta carotene when it didn't before.