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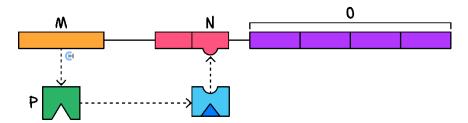
VCE Biology ¾
Gene Expression & the trp Operon II [0.4]
Workshop Solutions



Section A: Multiple Choice Questions (20 Marks)

Question 1 (1 mark)

The diagram below shows the process of gene regulation in the trp operon. Structures M-P are involved in the process.



Which one of the following identifies structures M-P?

	Structure M		Structure N	Structure O	Structure P
A.	Regulatory gene		Operator	Repressor	Structural genes
В.	Structural	D is correct. I	The regulatory gene (structure M) j		
C.	Structural	shape to conform to the amino acid tryptophan. The repressor protein then has a shape that can bind to the operator (structure N) so that the structural genes (structure O) cannot be transcribed. A, B and C are incorrect. These options do not correctly identify structures M-P.			
D.	Regulatory	gene	Operator	Structural genes	Repressor

Question 2 (1 mark)

When there is a high concentration of tryptophan,

- **A.** structure P will change shape and bind to structure N, which stops gene expression.
- **B.** structure *N* will change shape to conform with the tryptophan, preventing the RNA polymerase from moving past structure *N*.
- **C.** structure *M* will be prevented from producing structure *P*.
- **D.** structure N will change shape to conform with the tryptophan, preventing structure P from binding to it.

Question 6 A A is correct. When there is a high concentration of tryptophan, it is a waste of energy for bacteria to form tryptophan via a metabolic pathway. This means that the repressor protein (structure P) will bind to the operator (structure N) and prevent transcription of the trp structural genes (structure O). B and D are incorrect. The operator (structure N) does not change shape in the presence of tryptophan. C is incorrect. The presence of tryptophan does not affect the production of the repressor protein (structure P); it only prevents the transcription of the structural genes (structure O).



Question 3 (1 mark)

Identify the correct function of X, Y, and Z of the repressed trp operon as shown in the figure below.

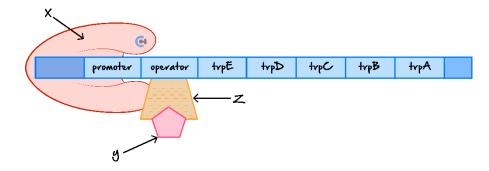


Figure 1: The gene structure of the trp operon

	X	Y	Z
A.	Repressor	Tryptophan	RNA polymerase
В.	Tryptophan	mRNA	Repressor
C.	RNA polymerase	Tryptophan	Repressor
D.	mRNA	Tryptophan	RNA polymerase

Question 4 (1 mark)

Gene expression is often suppressed by the end product

A. acting as a regulator.

B. acting as a promotor.

Question 4

C. not being expressed.

The protein products of gene expression can be used to regulate the gene, as once it is in excess, it switches off the gene acting as a regulatory protein.

D. being overexpressed.



Question 5 (1 mark)

When a prokaryote needs to produce tryptophan,

- **A.** RNA polymerase will bind to the tryptophan and mRNA will be produced.
- **B.** tryptophan will be released from the repressor and mRNA will be produced.
- C. tryptophan will detach from the promoter region and mRNA will be produced.
- **D.** RNA polymerase will detach from the operator region and mRNA will be produced.

Question 6 (1 mark)

The trp operon gives a model of gene regulation in bacteria. It involves the activation of a metabolic pathway that allows the synthesis of tryptophan in a low-tryptophan environment.

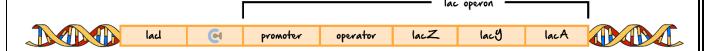
Which one of the following occurs when tryptophan is being produced by the trp operon?

- **A.** A repressor is bound to the operator section of the gene, allowing RNA polymerase to bind to the promoter for the subsequent expression of the gene.
- **B.** The shape of the repressor prevents it from binding to the promotor, allowing RNA polymerase to bind to the operator for the subsequent expression of the gene.
- **C.** None of the structural genes from the trp operon are expressed.
- **D.** The structural genes are expressed as RNA polymerase that was once bound to the promoter moves unimpeded along the trp operon.



Question 7 (1 mark)

Structural genes can be switched off and turned on by transcriptional factors expressed by regulatory genes. In prokaryotes, a group of genes associated with the breakdown of lactose is grouped together in a single operon called the lac operon. The diagram shows the position of the genes on the prokaryotic chromosome.



Transcription of the structural genes within the lac operon will occur when

- **A.** a repressor molecule is attached to the operator.
- **B.** RNA polymerase is attached to the promoter.
- C. lactose is absent from the prokaryotic cell.
- **D.** transcription of the lacl gene is optimal.

Question 8 (1 mark)

Transcription of the structural genes within the lac operon results in the production of molecules of

- A. a transcription factor.
- **B.** a repressor protein.
- C. lactose.
- **D.** mRNA.

Question 9 (1 mark)

In the context of the trp operon, what is attenuation?

- **A.** A process that increases the transcription of the trp operon.
- **B.** The addition of tryptophan to the growth medium of bacteria.
- C. A regulatory mechanism that uses the ribosome to terminate transcription prematurely.
- **D.** The degradation of tryptophan by bacterial enzymes.



Question 10 (1 mark)

In the absence of tryptophan, which structural formation in the trp operon's mRNA leader region prevents attenuation?

- **A.** The formation of a hairpin followed by a series of uracils.
- **B.** The formation of a hairpin that disrupts the ribosome.
- C. The formation of an anti-terminator hairpin structure.
- **D.** The binding of a repressor to the terminator sequence.

Question 11 (1 mark)

How does the ribosome's speed during translation affect attenuation in the trp operon?

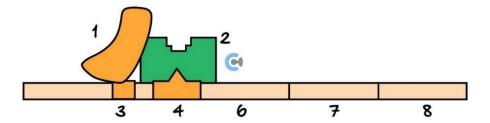
- **A.** A slow-moving ribosome during translation allows the formation of a terminator structure in the mRNA, leading to attenuation.
- **B.** A fast-moving ribosome prevents the formation of the stem-loop structure necessary for attenuation.
- **C.** The speed of the ribosome has no effect on attenuation.
- **D.** A slow-moving ribosome prevents attenuation by allowing anti-terminator structure formation.





The following information applies to the two questions that follow.

A repressed lac operon is represented by the following diagram:



Source: https://qph.ec.quoracdn.net

Question 12 (1 mark)

The promotor region is represented by:

- **A.** 1
- **B.** 2

- C 1.RNA polymerase, 2.Repressor Protein, 3.Promotor Region,
 - 4.Operator.

C. 3

D. 4

Question 13 (1 mark)

Transcription would begin in the presence of which of the following molecules?

A



B.



C



D



A

The shape that binds with the repressor protein (2) to remove it is only matched by A.



Question 14 (1 mark)

Repressor proteins regulate the production of protein by binding to the

- **A.** promotor region of the gene preventing RNA polymerase from functioning.
- **B.** operator region of the gene preventing RNA polymerase from functioning.
- C. RNA polymerase preventing it from binding to the promotor region of the gene.
- **D.** DNA polymerase preventing it from binding to the promotor region of the gene.

Question 1

Repressor proteins stop the gene from being expressed, hence they bind to the promotor site to stop RNA polymerase from being able to traverse along the gene.

Question 15 (1 mark)

In *E. coli*, the lac genes are expressed when lactose is present as

- **A.** RNA polymerase binds to the region.
- **B.** the repressor binds to the promotor region.
- **C.** lactose binds to the repressor protein.
- **D.** DNA polymerase binds to the promotor region.

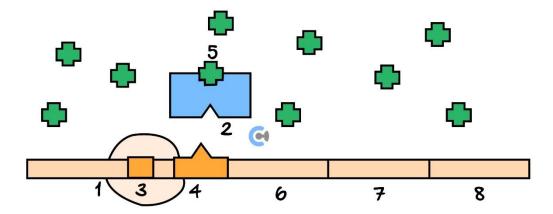
Question 3

Lactose binds to the repressor protein changing the shape of the repressor protein so it can no longer bind to the promotor region of the gene.



Question 16 (1 mark)

The lac operon shows how gene expression is controlled at the level of transcription. The model illustrates how bacteria, in the presence of lactose, are able to activate a gene that leads to the digestion of lactose. Once the lactose concentration is low the gene switches off. The diagram below illustrates this form of gene control when the lactose levels are high.



Components 1-8 are respectively:

- A. 1-Lactose; 2-RNA Polymerase; 3-Repressor; 4-Promoter; 5-Operator; 6,7,8-Genes
- **B.** 1-Lactose; 2-RNA Polymerase; 3- Promoter; 4-Repressor; 5-Operator; 6,7,8-Genes
- C. 1-RNA Polymerase; 2-Repressor; 3-Promoter; 4-Operator; 5-Lactose; 6,7,8-Genes
- **D.** 1-RNA Polymerase; 2-Lactose; 3-Repressor; 4-Promoter; 5-Operator; 6,7,8-Genes

Question 26

Space for Persona

Students should be able to understand the diagram; even though the lac operon is not required knowledge, gene regulation is an important part of the course. Numbers 6, 7 and 8 are clearly the genes to be activated. This means that 1 is likely to be RNA polymerase (making either \mathbf{C} or \mathbf{D} correct). Due to the lock and key shape of 2 and 4, iIt is likely that 2, if bound to 4, would stop transcription. Therefore 2 would act as a repressor protein. To detach the repressor, lactose needs to be present and so 5 must be the lactose.

Note: Knowledge of promoters and operators is not expected and even though this is part of the question, it is not important in determining the answer.



Question 17 (1 mark)

A study has found that there is a perfect linear relationship between the number of genes and the total sequence length of DNA in prokaryotic cells. However, this relationship is not as perfect in eukaryotic cells.

The best explanation for this difference is that

- **A.** prokaryotic cells are smaller than eukaryotic cells.
- **B.** eukaryotic DNA includes introns, which are absent in prokaryotic DNA.
- C. eukaryotic DNA codes for more proteins than prokaryotic DNA.
- **D.** prokaryotic DNA has different monomers than eukaryotic DNA.

Question 18 (1 mark)

DNA, mRNA, and tRNA all play important roles in protein synthesis. During the production of a protein, which of the following would occur during translation?

- **A.** The anticodon TAG on a tRNA molecule attached to an amino acid binds to the codon ATG on an mRNA molecule at a ribosome.
- **B.** The codon UAC on a tRNA molecule attached to an amino acid binds to the anticodon AUG on an mRNA molecule at the ribosome.
- C. The anticodon GGA on a tRNA molecule carrying an amino acid binds to the codon CCU on an mRNA molecule at a ribosome.
- **D.** The anticodon UAC on an mRNA molecule carrying an amino acid binds to the codon AUG on a tRNA molecule at a ribosome.

Question 19 (1 mark)

Which of the following statements refers to the degenerate nature of the DNA code?

- **A.** The codons UCU and UCC both code for the amino acid serine.
- **B.** DNA is found in all living things.
- **C.** The same gene will encode the same protein in different organisms.
- **D.** The codon CCG will always code for the amino acid proline, regardless of the organism or species it occurs in.



Question	20	(1	mark	١
Question	40	(I	mai K	J

The sequence of a particular gene in a DNA strand contains 640 triplet codes. How many codons would the corresponding mRNA strand that leaves the nucleus have?

- A. More than 640 codons
- **B.** 640 codons
- **C.** 638 codons
- **D.** Fewer than 640 codons

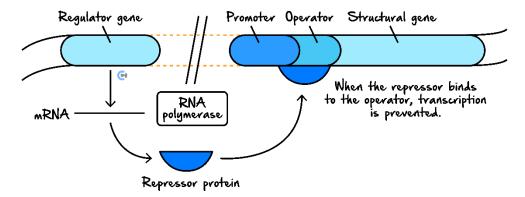
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Section B: Short Answer Questions (59 Marks)

Question 21 (6 marks)

In eukaryotic cells, many genes are inactive and need to be activated to be expressed. This is a way of ensuring cellular specificity. Certain signals are required to activate genes. The diagram below shows a simplistic view of gene regulation at the transcriptional level.



a. Why is it better for genes to be regulated at the transcriptional level? (1 mark)

If all genes were transcribed in each cell, there would be a lot of energy wasted if those mRNA strands were then stopped from being translated.

The following events occur in a particular sequence and show how a particular gene can be inactive:

- 1. The active repressor binds to the operator, blocking the expression of the structural genes.
- **2.** The structural gene is off.
- **3.** The regulator gene is transcribed, producing mRNA.
- **4.** RNA polymerase is unable to bind to the promoter as the repressor is bound to the operator.
- **5.** Regulator mRNA translates into an active repressor protein.
- **b.** Correctly order the events 1 to 5. (1 mark)

3, 5, 1, 4, 2

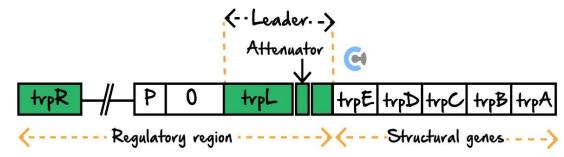


c.	To switch the structural genes on, the repressor needs to be removed from the operator. Describe how the repressor could be removed from the operator. (1 mark)
	The regulator gene could be inactivated, which would mean the repressor was no longer produced.
d.	Once transcription occurs there is further processing of the formed pre-mRNA. Discuss the importance of the removal of introns to the production of mRNA. (1 mark)
	Introns are transcribed but removed prior to translation. The exons bind to form the final mRNA product which carries the correct order of nucleotides for the protein to be formed.
e.	Describe the events of translation that lead to a specific structural protein. (2 marks)
	mRNA moves through the ribosome and is read in groups of 3 nucleotides (codons). tRNA carry an anticodon which binds to the codon in position. It also carries a specific amino acid which is put in the correct position in the growing polypeptide. 1 mark



Question 22 (6 marks)

The synthesis of tryptophan in prokaryotic cells such as *Escherichia coli* is regulated. The positions of both the regulatory region and the structural genes are shown in the diagram below.



Source: adapted from Trpoperon, https://en.wikipedia.org/wifi/File:Trpoperon.svg licensed CC-BY SA 3.0>

E. coli has two mechanisms that regulate the expression of the structural genes for the synthesis of tryptophan: repression and attenuation.

Compare the two mechanisms of regulation. Refer to the diagram in your answer.	

Students were required to compare the similarities and differences between the processes of repression and attenuation. However, students could also have answered this question by outlining the processes of both repression and attenuation as their way of comparing the two processes.

Key points included:

In repression, when tryptophan (*trp*) levels are high, two *trp* amino acids bind to the repressor, causing a conformational shape <u>change</u> in the repressor, enabling it to bind to the operator. This inhibits RNA polymerase from binding to the promoter to transcribe the structural genes.

In attenuation, when <u>trp</u> levels are high, RNA polymerase begins transcription of the leader or attenuator sequence. Within this sequence, there are two <u>trp</u> codons. As the ribosome does not stall here, a hairpin termination loop forms, resulting in the detachment of both RNA polymerase and the ribosome.

Both mechanisms occur to regulate trp levels, conserving energy in prokaryotic cells.

As both repression and attenuation occur when trp levels are high, students were not required to discuss these mechanisms in low trp environments.



Question 23 (13 marks)

In the depths of the ocean, the marine bacterium *Vibrio fischeri* exhibits bioluminescence, a result of gene expression from the lux operon. This operon consists of a sequence of genes critical for light production.

Recent studies have isolated a specific DNA sequence within the regulatory region of the lux operon:

Promoter sequence: 5' - TATAATGCGTCA - 3'

Operator sequence: 5' - GGACAATTTGCG - 3'

Initial coding sequence of lux gene: 5' - ATGGAAACCTTACCTGAC - 3'

The expression of the lux operon is regulated by the density of the bacterial population. At low densities, bioluminescence is not observed. However, as the population density increases, the concentration of a signalling molecule, the autoinducer, also increases. Once a threshold concentration of 20 nm is reached, the autoinducer binds to a regulatory protein, initiating transcription of the lux operon.

Experimental data show that at an autoinducer concentration of 5 nm, there is minimal transcription activity.

However, as the concentration reaches 20 nm and above, there is a significant increase in the rate of transcription, as measured by the amount of mRNA produced.

a.	Explain how environmental factors may control the expression of bioluminescence through transcription factors. (2 marks)						
		The marine bacterium will be able to detect population densities. When high, the autoinducer cause a regulatory protein to bind and initiate transcription.					



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	Transcription and translation occur simultaneously – this allows for regulation of the	
	trp operon via attenuation	
	There is no RNA processing in prokaryotes.	
iestion	24 (2 marks)	
nat are	operons and why might they be useful, particularly in gene regulation?	
	Operons are where multiple structural genes are under the control of a single promotor and operator, and this may be useful in gen regulation as many genes can be regulated	
	simultaneously, conserving regulatory resources as compared to having separate repressors and activators for each individual gene.	
	Allows whole pathway to be controlled efficiently.	
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Question 25 (13 marks)

The trp operon is found naturally in *E. coli* bacteria, and demonstrates multiple structural genes under the control of a single promoter, regulating the production of trp within the cell.

a. Using your knowledge, describe how the process of repression regulates the trp operon in *E. coli*. (4 marks)

The regulatory gene upstream of the operon will code for the production of the trp repressor protein.

When there is a sufficient level of trp in the cell, it will bind to the repressor protein. This causes a conformational shape change in the repressor, therefore allowing it to bind to the operator region of the operon.

This blocks RNA polymerase form transcribing the genes, resulting in the enzymes responsible for trp synthesis not being produced.

When there is a shortage of trp however, the repressor will not have trp binding to it, and will therefore not undergo the conformational shape change required to bind to the operator. In this case,

RNA polymerase can bind to the promotor and travel along the gene unimpeded, allowing transcription to occur. Hence, the enzymes required for trp synthesis will be produced, allowing the cell to produce trp on its own.

In prokaryotes, transcription and translation can occur simultaneously due to a lack of processing and a nucleus.
In between the operator and the structural genes, there is a leader sequence.
This leader sequence contains the code for two trp residues. If there are low levels of tryptophan, then the ribosome will be stalled while translating this sequence.
Following the leader sequence there is an attenuator – if the ribosome is stalled than this attenuator will from a anti terminator loop and allow transcription to occur.
 If there are high levels of trp then the ribosome will not be stalled – thus resulting in a terminator loop to be formed, preventing full transcription of the structural genes.



c.	Why	Why isn't the process described in part a. permanent? (2 marks)			
		This ensures that the cell can be responsive to the current levels of trp present in determining its regulation			
		The repressor itself doesn't permanently attach to the operator even with trp present, after a while it detaches spontaneously. This means that a new repressor will have to bind trp to prevent expression. Hence, this process is NOT permanent.			
d.		ify and describe some of the differences between gene expression in prokaryotes and eukaryotes, and in how this is relevant to the trp operon. (2 marks)			
		Transcription and translation occur simultaneously – this allows for regulation of the trp operon via attenuation			
		There is no RNA processing in prokaryotes.			
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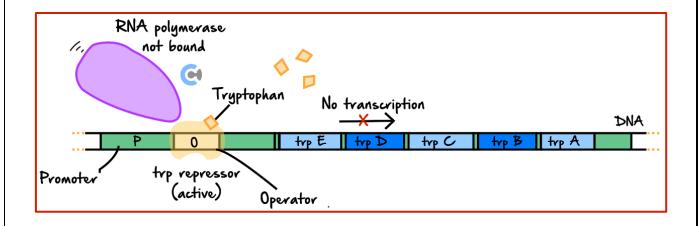


Question 26 (3 marks)

Unlike humans, the bacterium *Escherichia coli* does not need to rely on consuming tryptophan from the environment as these bacteria can synthesise their own. The trp operon regulates the production of tryptophan in *E. coli*. The diagram below shows the position of the five genes in the trp operon.

Annotate the diagram below to show how *E. coli* regulates the expression of genes through repression when tryptophan levels are high in the cell.

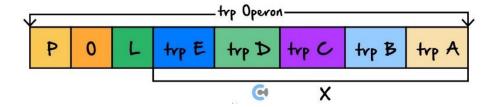






Question 27 (8 marks)

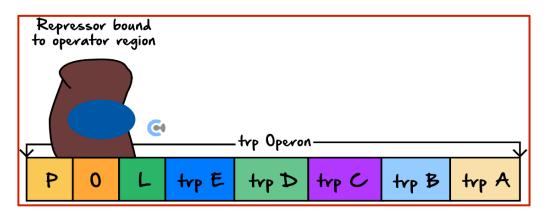
The trp operon is found in *Escherichia coli* bacteria and functions to provide these bacteria with the ability to produce the essential amino acid tryptophan when it is absent from their environment or to repress its production when it is high in their environment. The figure below shows a pictorial representation of the trp operon.



a. Using the information outlined in the figure above, complete the table below, identifying parts of the trp operon using labels, and names and describing the function performed. (6 marks)

	Letter	Structure	Function	
Letter	P	Promoter region	Site of binding of	n
P			RNA polymerase	
	0	Operator region	Site of binding of	
			repressor protein	
	X	Structural genes	Code for enzymes	
			involved in	involved in
			tryptophan synthesis	nthesis
L			(6 marks)	

b. In the figure below, draw where a repressor protein would bind when tryptophan levels are high in the bacterial cell. (1 mark)



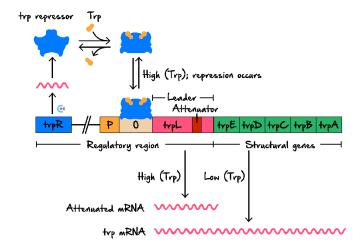
c. Explain why bacteria regulate the gene expression of enzymes involved in the synthesis of tryptophan. (1 mark)

Regulation of gene expression of enzymes involved in tryptophan synthesis enables the bacteria to conserve energy/ATP when tryptophan levels in the environment are high (1 mark).



Question 28 (8 marks)

The trp operon contains five structural genes, trp E, trp D, trp C, trp B and trp A, which encode the enzymes needed to synthesise tryptophan. Regulation of the trp operon is accomplished by repression and attenuation.



a. Explain how hairpin loops in the leader section assist in regulating the expression of the trp operon by the process of attenuation. (6 marks)

Worked solution

The leader segment has 2 adjacent *trp* codons, which when transcribed have regions that undergo base pairing to form hairpin loops. There are 2 types of loops: one that stalls the ribosome and the other that does not. When tryptophan levels are high, the ribosome does not pause at the *trp* codons in the leader section. The hairpin loop formed acts as a signal to terminate transcription. A shorter (attenuated) strand of mRNA detaches from the ribosome. This shorter strand does not encode for the five structural genes, so they are not expressed. Therefore, the production of tryptophan ceases until the concentration of tryptophan in the cell is low enough so that more needs to be synthesised.

Mark allocation: 6 marks

- 1 mark for identifying that there are 2 different types of hairpin loop: one stalls the ribosome, and the other does not.
- 1 mark for stating that the ribosome does not stall/translates quickly when tryptophan levels are high.
- 1 mark for stating that the hairpin loop acts as a transcription termination signal.
- 1 mark for stating that an attenuated strand detaches from the ribosome, or the ribosome detaches itself.
- 1 mark for stating the structural genes that code for the synthesis of tryptophan are not expressed.
- 1 mark for stating that tryptophan is not synthesised, until sufficient quantities of the tryptophan already present have been used up.



Worked solution	
Possible answers in	clude:
Repression involves	the action of a repressor protein, whereas attenuation does not.
	s the synthesis of mRNA from occurring (prevents mRNA synthesis), a alters the process of transcription after it has already started.
	in is produced upstream of the promoter region, whereas attenuation section downstream from the promoter region.
	s the synthesis of mRNA from occurring, whereas attenuation enables RNA to be synthesised.
Any other valid answ	ver
Mark allocation: 2 i	marks
 1 mark for any 	correct comparison between the mechanisms of repression and a maximum of 2 marks.



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