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VCE Biology ¾
AOS 1 Revision [1.0]
SAC 3

45 Marks. 5 Minutes Reading. 60 Minutes Writing.

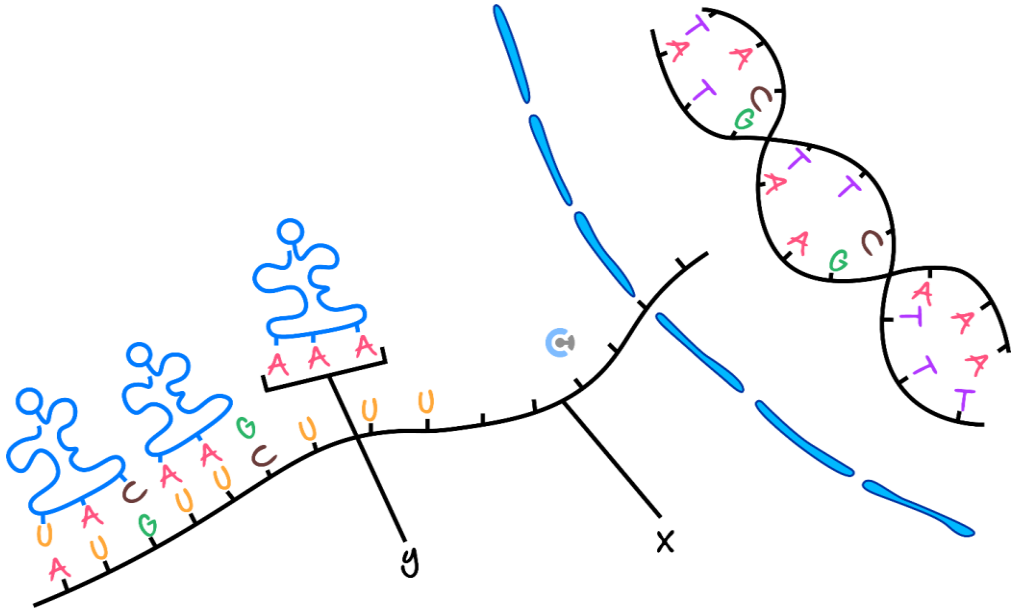
Section A: SAC Questions (45 Marks)

The Production of Human Insulin:

- In 1928, researchers discovered that insulin is a polypeptide, a finding that was further refined in 1952 when the precise amino acid sequence was determined. This hormone, critical for regulating metabolism, consists of two distinct protein chains: Chain A, which is made up of 21 amino acids, and Chain B, longer with 30 amino acids. Insulin's primary role is to facilitate anabolic processes, including the synthesis of proteins, which is essential for the growth and repair of skeletal muscle tissue. It plays a pivotal role in the body's management of nutrients, particularly by promoting the uptake of glucose into cells.

Question 1 (8 marks)

Insulin, like any other protein, is synthesised through transcription and translation of the genetic instructions that encode for it.



a. Name the complex responsible for the final step in the production of insulin polypeptides. (1 mark)

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b. What is label *Y*? (1 mark)

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c. Outline the role of the complex named in **part a.** in the production of an insulin polypeptide. (3 marks)

The diagram above displays gene expression in eukaryotes.

d. What process is missing from this diagram? Name 2 events that occur during this process. (3 marks)

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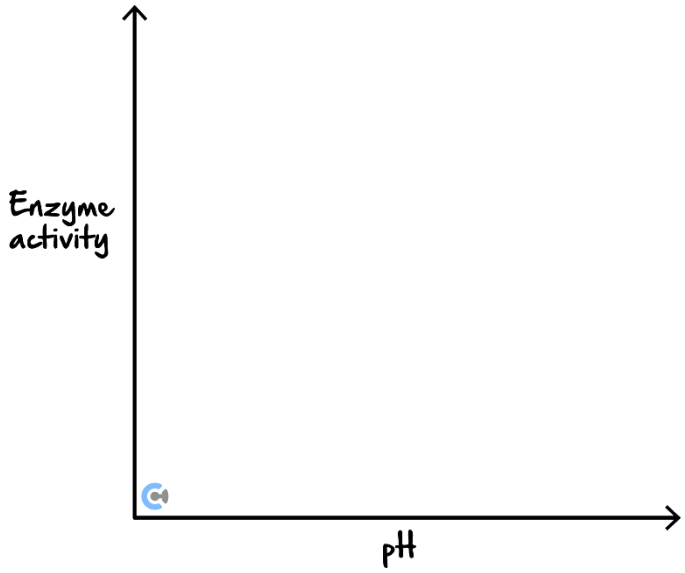
Definition

Insulin Production:

- The global surge in diabetes prevalence and advanced insulin administration methods, which often necessitate increased dosages, are anticipated to boost the demand for recombinant insulin. Existing production methods may struggle to meet this need affordably due to capacity constraints and elevated costs. To manufacture therapeutic recombinant insulin, a host organism is necessary–one capable of proper protein folding and post-translational modifications. Escherichia coli (E. coli) is commonly employed for producing recombinant human insulin for therapeutic use.

Question 2 (5 marks)

a. Sketch a graph showing enzyme activity against pH. (2 marks)



b. When bacterial growth is sufficient, scientists will often store bacteria at 4°C. Explain in terms of enzyme activity why scientists use this technique. (3 marks)

Question 3 (10 marks)

a. Name three types of enzymes required to create recombinant DNA. Explain the role of each enzyme in creating a recombinant plasmid containing the human insulin gene. (6 marks)

Type of enzyme	Role in creating recombinant plasmid

Plasmids must have a few key components in order to be considered as candidates for bacterial transformation.

b. Name ONE feature (excluding genes) that a plasmid must have to be used in the production of insulin, and describe their significance. (2 marks)

c. Explain the feature of the genetic code that allows for insulin production to be performed using bacteria. (1 mark)

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d. Explain why there is an additional step of purification and modification when producing human insulin using E. coli. (1 mark)

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Definition

Insulin Ethics:

- Previously, insulin was extracted from the pancreas of pigs and cows until 1979.
  This emerged as a cost-effective mechanism that did not involve the killing of animals. However, there are still some reservations regarding the use of antibiotics, which may lead to antibiotic resistance.

Question 4 (9 marks)

a. Using the ethical concepts and principles you have learned this year, discuss the ethical implications of using recombinant plasmids to produce insulin. (4 marks)

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b. Would E.coli be considered transgenic in this case? Explain. (2 marks)

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c. Suggest a method by which insulin could be produced with plasmids without using antibiotics. (3 marks)

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Definition

Insulin Ethics:

- Insulin, a hormone synthesised by pancreatic cells, plays a crucial role in the regulation of blood sugar. When one eats, blood sugar levels spike, prompting insulin release, which aids in glucose uptake by cells. Conversely, a drop in insulin prompts the liver to release glucose into the bloodstream. Inadequate insulin production or utilisation can precipitate diabetes mellitus.
  A healthy adult's pancreas typically harbours around 200 units of insulin, secreting 30 to 50 units daily. Insulin release is triggered by various physiological cues, with blood sugar levels being a primary regulator.

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Question 5 (6 marks)

a. The trp operon is one such example of gene regulation in E.coli.

Suggest 2 benefits of performing gene regulation. (2 marks)

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b. What is an operon? (1 mark)

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c. Explain how trp production in E.coli is regulated by repression of the trp operon. (3 marks)

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Question 6 (2 marks)

Outline the difference between a consequences-based and a duty/rule-based approach to resolving bioethical issues.

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Question 7 (1 mark)

Scientists are using blue-white screening to identify bacteria that have successfully taken up a recombinant plasmid containing the human insulin gene. In this experiment, the lacZ gene was intentionally interrupted by inserting the insulin gene within it. After transformation, bacterial colonies are grown on an agar plate containing X-gal.

Which of the following results would indicate that a bacterial colony has successfully taken up the recombinant plasmid containing the insulin gene?

A. A blue colony because the lacZ gene is still functional and able to break down X-gal.

B. A white colony because the inserted insulin gene has disrupted the lacZ gene, preventing β-galactosidase production.

C. No colony growth because the insulin gene prevents bacteria from dividing.

D. A fluorescent colony because insulin-producing bacteria glow under UV light.

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Question 8 (1 mark)

Which feature of a plasmid ensures that it is replicated and passed on to bacterial daughter cells after transformation?

A. The restriction enzyme recognition site, which ensures only transformed bacteria can grow.

B. The origin of replication (ORI), which allows the plasmid to be copied independently of the bacterial chromosome.

C. The selectable marker, which allows plasmids to be inherited through bacterial reproduction.

D. The promoter region, which initiates transcription and ensures plasmid replication.

Question 9 (1 mark)

During heat shock transformation, scientists treat bacterial cells with Ca^{2+} (calcium ions) before applying a brief heat pulse.

What is the main role of Ca^{2+} ions in this process?

A. Ca^{2+} strengthens the bacterial cell wall, preventing damage during heat shock.

B. Ca^{2+} activates heat shock proteins, which bind to plasmids and transport them into the bacterial cytoplasm.

C. Ca^{2+} neutralises the negative charge of the bacterial membrane and plasmid DNA, allowing DNA to enter the cell.

D. Ca^{2+} integrates into the plasmid DNA, making it more stable and easier to replicate.

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Question 10 (1 mark)

Scientists have inserted the human insulin gene into a plasmid and transformed E. coli cells. To confirm that the plasmid now contains the insulin gene, they use restriction enzymes to cut the plasmid and then run the DNA fragments through gel electrophoresis.

Which of the following results would confirm that the insulin gene was successfully inserted?

A. Two DNA bands: One corresponding to the plasmid backbone and one corresponding to the insulin gene.

B. A single DNA band matching the size of the original unmodified plasmid.

C. No DNA bands at all because recombinant plasmids do not migrate in a gel.

D. Multiple identical DNA bands across all lanes because all DNA fragments migrate the same distance in gel electrophoresis.

Question 11 (1 mark)

A biotechnology company is producing recombinant human insulin using E. coli. Initially, the bacteria are grown in large fermentation tanks, but the scientists notice that insulin production decreases significantly over time. They suspect that the plasmid containing the insulin gene is being lost from bacterial cells as they divide.

Which of the following would be the most effective strategy to ensure that bacterial cells retain the plasmid and continue producing insulin?

A. Growing the bacteria in a medium containing an antibiotic that selects for plasmid-containing cells.

B. Attaching the insulin gene to the bacterial chromosome instead of using a plasmid to ensure it is passed on during cell division.

C. Adding extra glucose to the medium to provide bacteria with more energy for plasmid replication.

D. Growing the bacteria in a medium containing X-gal that selects for plasmid-containing cells.

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