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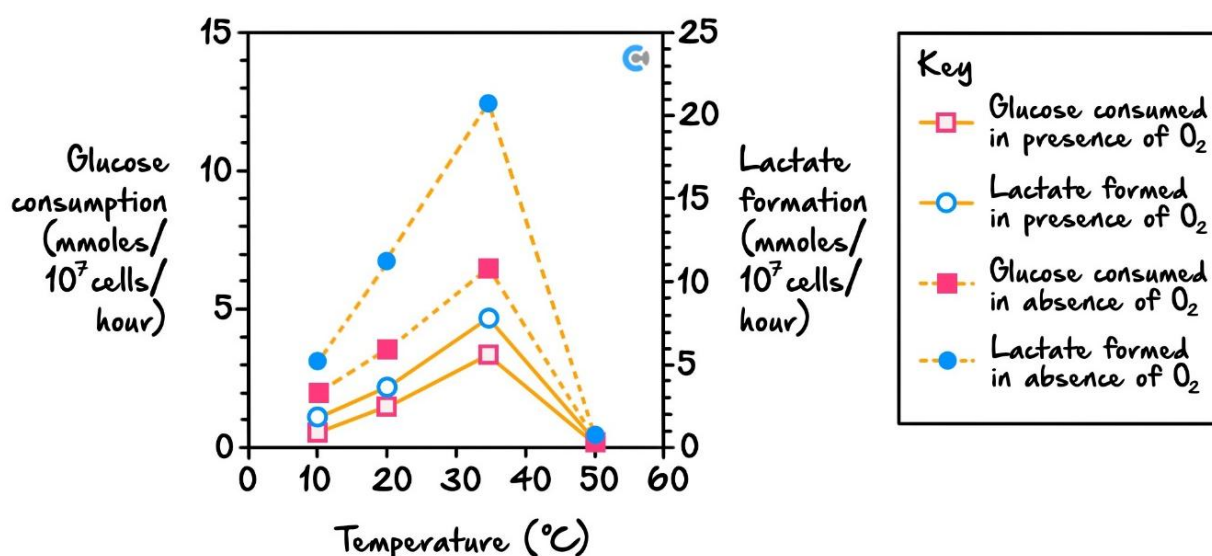
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VCE Biology $\frac{3}{4}$
Cellular Respiration & Anaerobic Fermentation [0.10]
Workshop Solutions

Section A: Multiple Choice Questions (21 Marks)

Question 1 (1 mark)

The graph below shows the effect of temperature and oxygen on the rate of consumption of glucose and the corresponding rate of production of lactate in white blood cells at four different temperatures (10°C, 20°C, 35°C, and 50°C).



Under which of the following conditions is the rate of lactate formation highest?

- A. 10°C in the absence of oxygen.
- B. 20°C in the absence of oxygen.**
- C. 35°C in the presence of oxygen.
- D. 50°C in the presence of oxygen.

Question 2 (1 mark)

Why is the rate of consumption of glucose higher in the absence of oxygen than in its presence at 35°C?

- A. Oxygen stimulates lactate formation.
- B. Oxygen inhibits the uptake of glucose by the cell.
- C. Glycolysis reactions require the presence of oxygen.
- D. Less ATP is formed from glucose in the absence of oxygen.**

Question 3 (1 mark)

Why are the rates of consumption of glucose and formation of lactate at 50°C the same irrespective of oxygen levels?

A. Most of the enzymes involved in glycolysis have become denatured.

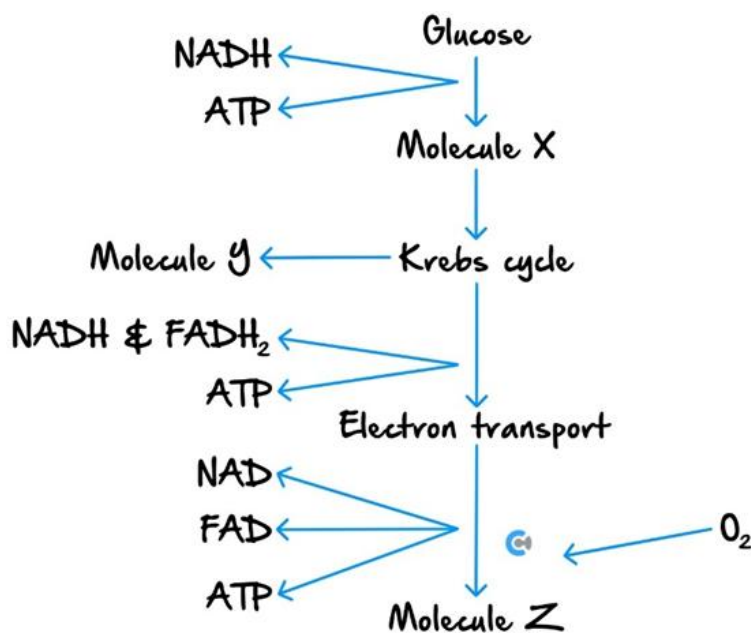
B. Glucose is converted to ATP by a single chemical reaction.

C. At 50°C the cell uses heat as a source of energy.

D. At 50°C metabolic pathways are very efficient.

The following information applies to the three questions that follow.

The flowchart shows the process of cellular respiration in an eukaryotic cell.



Question 4 (1 mark)

Molecules X, Y and Z respectfully are:

A. Carbon dioxide, pyruvate and water.

B. Pyruvate, water and carbon dioxide.

C. Pyruvate, carbon dioxide and water.

D. Water, carbon dioxide and pyruvate.

Question 5 (1 mark)

Molecule *X* is produced in the:

- A. Cristae of the mitochondria.
- B. Grana.
- C. Cytosol.
- D. Mitochondrial matrix.

Question 6 (1 mark)

Inputs of the electron transport stage include:

- A. NADH, FADH₂, ADP + Pi, oxygen.
- B. NADH, FADH₂, ATP, water.
- C. NAD⁺, FAD⁺, ADP + Pi, ATP.
- D. NADP, FADH₂, ADP + Pi, oxygen.

Question 7 (1 mark)

The build-up of lactic acid in the muscle cells of weight-lifters is the result of anaerobic conditions and:

- A. Rapid ATP production during the Krebs cycle.
- B. Rapid ATP production during glycolysis.
- C. Slow ATP production during electron transport.
- D. Slow ATP production during cellular respiration.

Space for Personal Notes

Question 8 (1 mark)

A change in which of the following would **not** affect the rate of anaerobic cellular respiration?

- A. Oxygen concentration.**
- B. Glucose concentration.
- C. ATP production.
- D. Temperature.

Question 9 (1 mark)

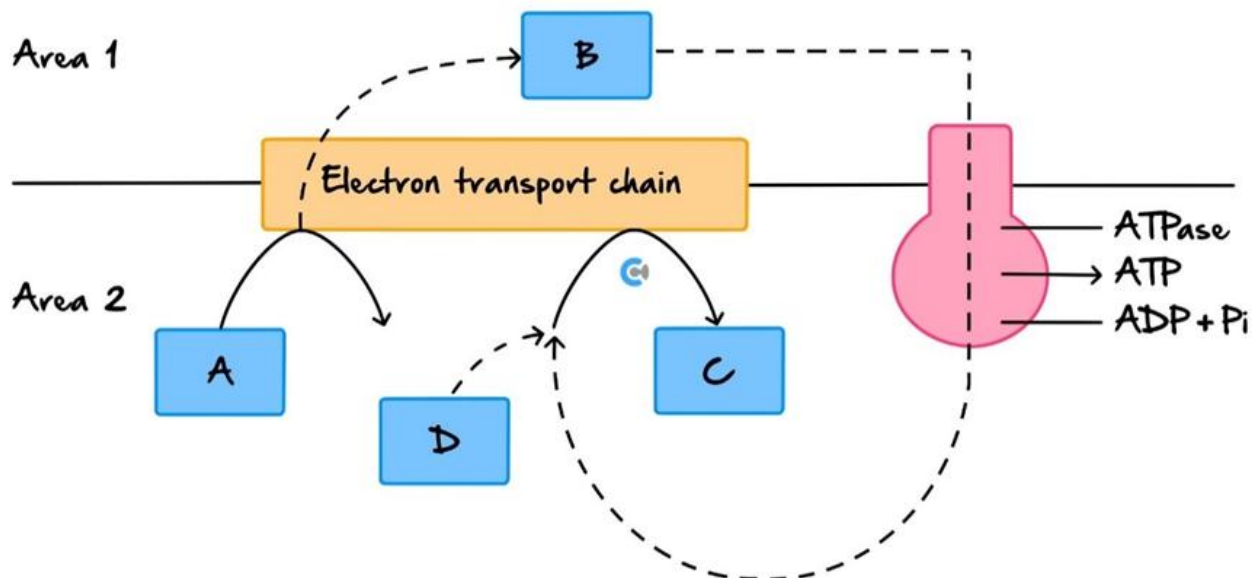
Some species of bacteria can be added to biomass to produce bioethanol, a renewable source. The production of bioethanol involves the breakdown of starch:

- A. By glycolysis followed by alcoholic fermentation.**
- B. Via aerobic respiration followed by carbon dioxide production.
- C. Through the Krebs cycle followed by lactic acid fermentation.
- D. In the presence of oxygen followed by fermentation energy.

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Question 10 (1 mark)

The diagram below shows a reaction that occurs in most eukaryotic cells. Chemicals A-D are involved in the process.



Which one of the following identifies chemicals A-D?

	Chemical A	Chemical B	Chemical C	Chemical D
A.	NADPH	Hydrogen	Oxygen	Water
B.	Oxygen	NADH	Water	Hydrogen
C.	Water	Carbon dioxide	Oxygen	Hydrogen
D.	NADH	Hydrogen	Water	Oxygen

Question 11 (1 mark)

The role of oxygen in aerobic cellular respiration is:

- A. As a reactant in glycolysis.
- B. As a waste product in the Krebs cycle.
- C. As a reactant to produce acetyl-CoA.
- D. As an electron acceptor in the electron transport chain.

Question 12 (1 mark)

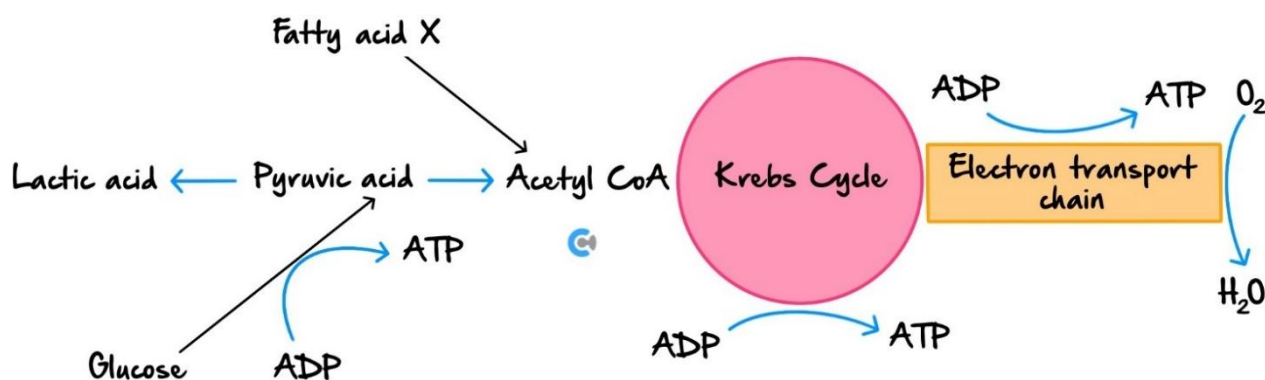
Identify the correct statement below.

- A. The Krebs cycle produces NADPH.
- B. Glycolysis produces 1 ATP molecule.
- C. The electron transport chain produces water.
- D. More ADPs are used in glycolysis than in other stages.

Question 13 (1 mark)

If there is insufficient glucose for cellular respiration, fatty acids can be changed to acetyl CoA. Each fatty acid *X* molecule produces eight molecules of acetyl CoA.

The diagram below summarises the pathways for the breakdown of fatty acid *X* and glucose. The number of molecules produced in each step is not shown.



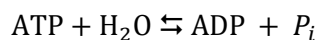
Referring to the information above and your knowledge of cellular respiration, which one of the following conclusions can be made?

- A. Most of the ATP is made in the Krebs Cycle.
- B. Pyruvic acid is converted to acetyl CoA under anaerobic conditions.
- C. No ATP can be formed from the breakdown of glucose under anaerobic conditions.
- D. One fatty acid *X* molecule produces more ATP in aerobic conditions than one glucose molecule does.

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Question 14 (1 mark)

The following reaction represents the ATP cycle:



A reasonable conclusion to make about the ATP cycle would be:

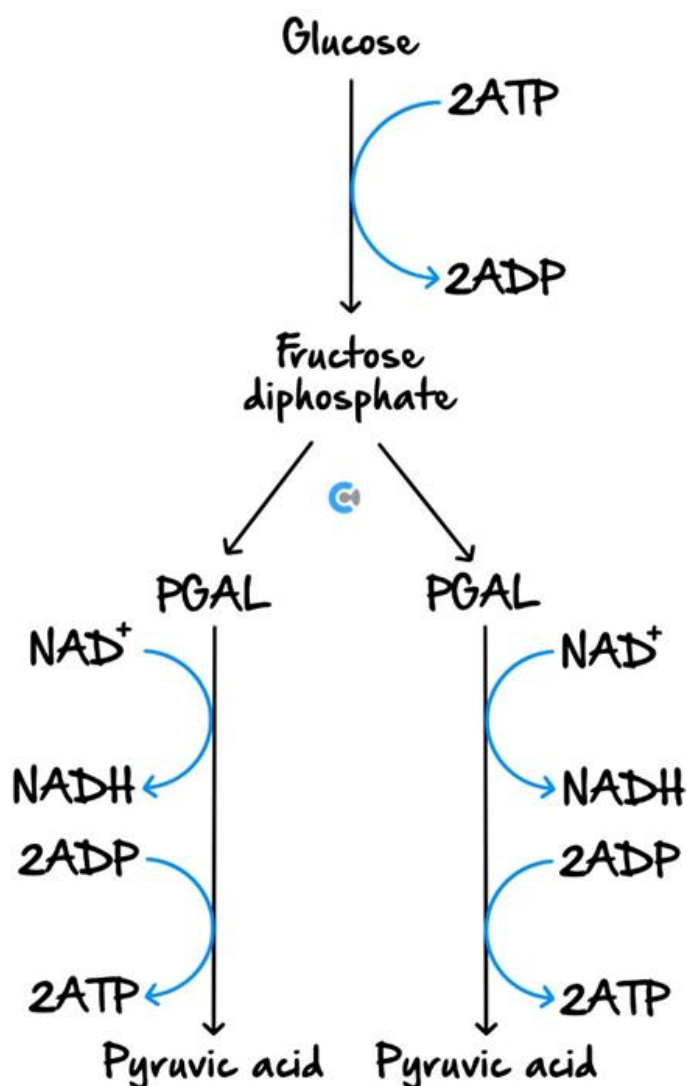
- A.** The formation of ATP is a catabolic reaction requiring an input of energy.
- B.** The formation of ADP from ATP would occur while digesting fats.
- C.** The formation of ATP from ADP and the formation of ADP from ATP would occur in the same cellular location.
- D.** The formation of ATP is an example of a condensation reaction that is anabolic and endergonic.

Spa

The ATP cycle is a two-way reaction. One way is the catabolic/exergonic/exothermic/hydrolysis reaction of converting ATP into ADP and using the energy for whatever reason is necessary. The other way is the anabolic/endergonic/endothermic/condensation reaction converting ADP into ATP. The energy to do this generally comes from cellular respiration.

The following information applies to the two questions that follow.

The following chemical pathway proceeds in virtually all cells.



Question 15 (1 mark)

The name of the above chemical pathway is:

- A. The Krebs cycle.
- B. The electron transport chain.
- C. The link reaction.
- D. Glycolysis.**

Glycolysis is the breakdown of glucose into pyruvic acid. Once the pyruvic acid is formed, if oxygen is present, it moves into the mitochondria. Initially it is converted into acetyl CoA (via the link reaction), which is inserted into the Krebs cycle where carbon dioxide and ATP are formed. The H⁺ is picked up by a hydrogen carrier, and the hydrogen becomes part of the electron transport chain. This produces the majority of the ATP generated via aerobic respiration.

Question 16 (1 mark)

The products and by-products of this reaction are:

A. Pyruvic acid, ATP, ADP and NADH.

B. 2 pyruvic acid, 2 ATP and 2 NADH.

C. 2 pyruvic acid.

D. 2 ATP and 2 NADH.

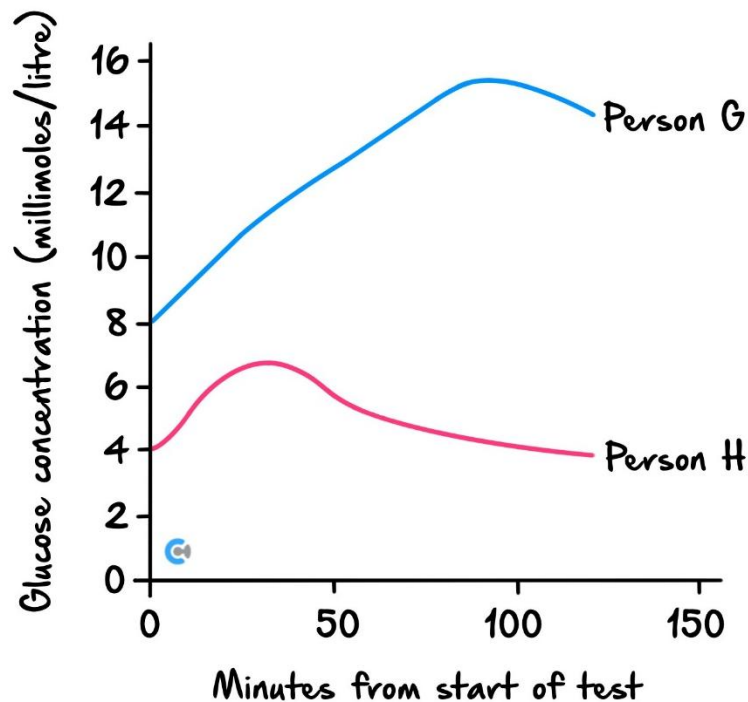
It can be seen clearly that there are two pyruvic acid molecules produced per glucose molecule. More obscure are the other products. There is a net 2 ATP produced in total (two used and four produced). There are also two NADH formed.

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Question 17 (1 mark)

Gestational diabetes mellitus (GDM) is a condition that is diagnosed when higher-than-normal blood glucose levels first appear during pregnancy. In Australia, 8-10% of pregnant women will develop GDM around the 24th to 28th week of pregnancy, which is why routine screening occurs at this time. Women take a glucose challenge test that involves drinking a glucose solution and testing the amount of glucose that remains in the blood after one hour.

The graph shows the results of GDM glucose challenge tests that were taken by two women.



From the graph, it is possible to conclude that:

- A. Person *G* does not have diabetes as the amount of glucose in her blood begins to decline after around 100 minutes after drinking the glucose solution.
- B. Person *H* does not have diabetes as the level of glucose in her bloodstream peaks for a short time and then returns to its original level.
- C. Both patients have diabetes.
- D. Neither patient has diabetes.

A is incorrect - Person *G* has diabetes, her blood glucose level does not return to its original level within the duration of the test.
 B is correct - Person *H* does not have diabetes
 C is incorrect - Person *G* has diabetes, Person *H* does not.
 D is incorrect - Person *G* has diabetes, Person *H* does not.

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Question 18 (1 mark)

Which one of the following gives a biochemical reaction correctly linked with its corresponding cellular location?

- A. The light-independent reaction occurs within the lumen of the thylakoid.
- B. ATP hydrolysis occurs at the ribosomes in the cytosol.**
- C. Glycolysis occurs within the matrix of the mitochondria.
- D. The Krebs cycle occurs within the stroma of the chloroplast.

To solve this question, a thorough understanding of the metabolic processes within cells is needed.

A is incorrect because it is the light-dependent reaction, not the light-independent reaction, occurring in the grana. C is incorrect because glycolysis occurs in the cytosol, not the mitochondria. D is incorrect because the Krebs cycle occurs in the matrix of the mitochondria, not the stroma of the chloroplast. B is correct because ATP hydrolysis (to ADP and P_i) occurs at the ribosome during protein synthesis, as this is an endergonic reaction.

Question 19 (1 mark)

A patient is diagnosed with a rare disease that impairs the function of Complex I of the electron transport chain in mitochondria.

What is the most likely consequence of this dysfunction on cellular respiration?

- A. Increased efficiency of glycolysis.
- B. Decreased production of ATP during oxidative phosphorylation.**
- C. Enhanced activity of the Krebs cycle.
- D. Increased lactic acid production in the absence of oxygen.

Question 20 (1 mark)

Which of the following correctly identifies which loaded carriers are produced during the Krebs cycle?

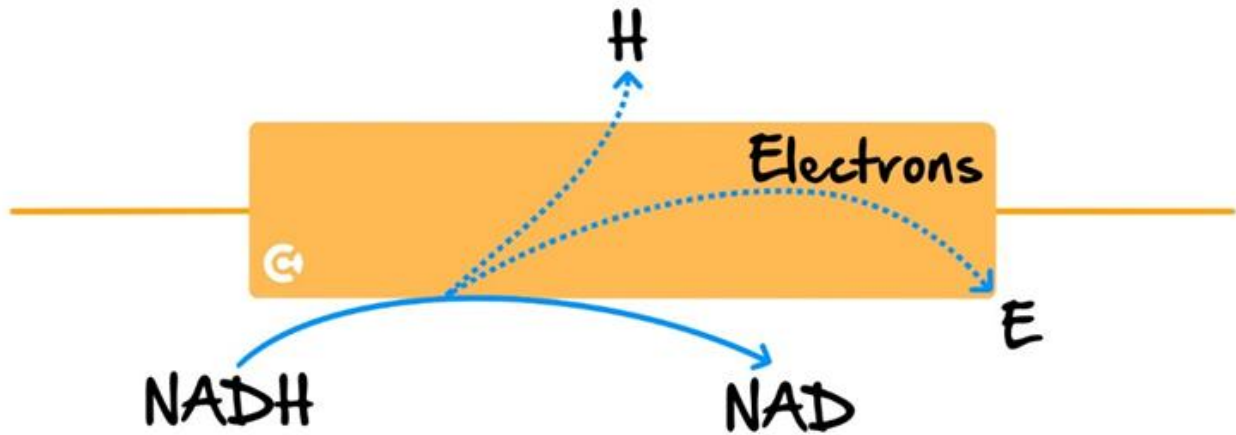
- A. NADH only.
- B. $FADH_2$ only.
- C. NADH and $FADH_2$.**
- D. NADPH and $FADH_2$.

Both NADH and $FADH_2$ are produced during the Krebs cycle.

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Question 21 (1 mark)

The diagram below simplifies what occurs to NADH at the cristae of the mitochondria. The electrons (E) are available for oxygen to eventually form water. The hydrogen (H) forms a concentration gradient across the mitochondrial membrane.



Once the NADH offloads its hydrogen and electrons, it would:

- A.** Collect more hydrogen from the breakdown of molecules in the Krebs cycle.
- B.** Collect more hydrogen from the breakdown of water within the grana.
- C.** Digest the NAD into raw materials so it can be recycled.
- D.** Move to the cytosol to collect more hydrogen from the condensation of amino acids at the ribosome.

NAD is a cofactor involved in the collection of hydrogen from glucose during cellular respiration. Once it collects hydrogen, it becomes NADH. The NADH moves into the mitochondria (or it is already there) to be an input into the electron transport chain (ETC). It offloads the hydrogen, then is able to collect more hydrogen from the Krebs cycle. The hydrogen carrier involved in the transferral of hydrogen during photosynthesis is NADP (making B incorrect).

Section B: Short Answer Questions (105 Marks)

Question 22 (9 marks)

The process of cellular respiration is a biochemical pathway with a few key steps.

Fill in the tables with inputs, outputs, and locations of each step.

a. Glycolysis. (3 marks)

<u>Inputs</u>	<u>Outputs</u>	<u>Location</u>
Glucose ($C_6H_{12}O_6$)	2 pyruvate	Cytosol of Cell
2 ADP + Pi	2 ATP	
2NAD +	2 NADH	

b. Krebs Cycle. (3 marks)

<u>Inputs</u>	<u>Outputs</u>	<u>Location</u>
2 Acetyl CoA	4 CO ₂	Mitochondrial Matrix
2 ADP + Pi	2 ATP	
6 NAD ⁺ + 6H ⁺	6 NADH	
2 FAD + 4H ⁺	2 FADH ₂	

c. Electron Transport Chain. (3 marks)

<u>Inputs</u>	<u>Outputs</u>	<u>Location</u>
O ₂	H ₂ O	Cristae of Mitochondria
26/28 ADP + Pi	26/28 ATP	
10 NADH	10 NAD +	

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Question 23 (6 marks)

Scientists measured the metabolic activity of mammalian cells by measuring the uptake of glucose into the cells. The cells were maintained at 37°C with a pH of 7.4 and suspended in a nutrient solution containing glucose. The uptake of glucose into the cells was recorded for the next 30 minutes.

- a. Explain why the uptake of glucose into the cells could be used to measure the metabolic activity of the cells. (2 marks)

Glucose is the substrate for aerobic cellular respiration (a metabolic process), and hence the levels of glucose uptake into the cell can show its rate. For example, the more glucose being taken into the cell can indicate a higher rate of respiration.

- b. The scientists repeated the experiment. They kept all conditions the same as for the first experiment, except that the cells were kept in low-oxygen conditions.

Would the uptake of glucose into the cells be expected to be higher, lower, or the same as for the first experiment? Justify your response. (4 marks)

Higher uptake

- With less oxygen, the cell would not respire aerobically, producing less ATP for each glucose molecule.
- The cell would take up more glucose to get the same amount of energy.

OR

Lower uptake

- With less oxygen, the cell would respire anaerobically and produce toxic products that could cause damage to cells.

OR

Same uptake

- Oxygen is not required for the breakdown of glucose, so glycolysis still occurs and produces less ATP and less energy for the cell.

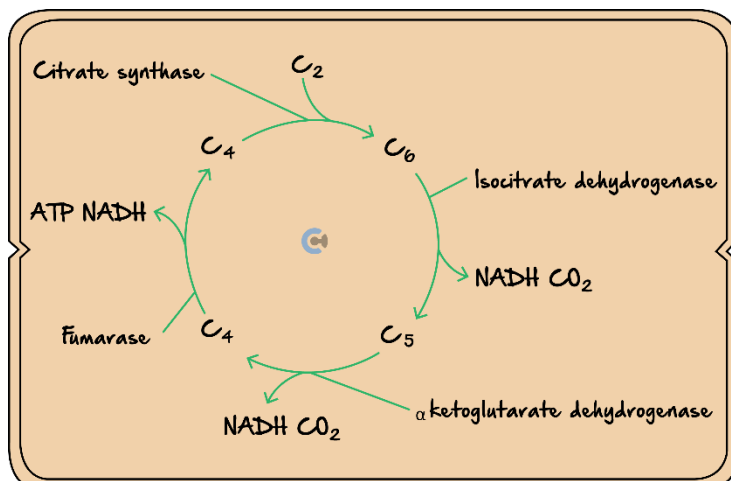
The marking scheme is weird for this one - as long as you elaborate and expand on one of the options, it should be fine.

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Question 24 (6 marks)

A simplified version of a stage of cellular respiration that occurs in the matrix of the mitochondria is shown in the diagram below.

Note: C_2 , C_4 , C_5 , and C_6 are different-sized carbon molecules (C_2 represents two carbon atoms in the molecule, C_4 represents four carbon atoms, and so on).



a.

- i. Name the stage of cellular respiration illustrated in the diagram. (1 mark)

The Krebs cycle

- ii. State the outputs from the simplified diagram of this stage of cellular respiration. (1 mark)

3NADH ATP 2 CO_2 $FADH_2$

b. Describe the function of citrate synthase. (2 marks)

Enzyme that catalyses a chemical reaction in this biochemical pathway.
Specifically, one that combines a 4-carbon molecule with a 2-carbon molecule.

- c. Fumarase deficiency is a very rare genetic disorder that causes individuals with the condition to have extremely low levels of fumarase.

Use the information provided with the simplified version of cellular respiration to describe **two** effects of fumarase deficiency. (2 marks)

Any two of:

- No ATP formed.
- A build-up of the first C_4 compound in the cycle.
- A lack of C_4 substrate for the citrate synthase reaction.
- A slowing down (or stopping) of the whole cycle.
- Less CO_2 and NADH.

Question 25 (11 mark)

Anaerobic Respiration is one mechanism by which glucose can be metabolised in animals, plants, and bacteria.

- a. What is meant by the phrase 'anaerobic'? (1 mark)

In the absence of oxygen.

- b. Other than ethanol, what is the product of this process (anaerobic respiration)? (1 mark)

Carbon dioxide

A limitation of using this process to manufacture bioethanol is the toxicity of ethanol to the cell, which causes death in most cases. A proposed method to solve this limitation is to increase the activity of alcohol dehydrogenase (which metabolises ethanol) by genetic editing via CRISPR-Cas9.

- c. Describe the process by which the scientist can edit the gene coding for alcohol dehydrogenase using CRISPR-Cas9. (3 marks)

Identify the gene coding for this – the target sequence.

Alter Cas9 and develop a guide RNA sequence complementary to this to make the edit.

Deliver this to the cells in the hope of increasing the rate of degradation of the ethanol.

- d. Evaluate the suitability of using this method for optimising bioethanol production. (2 marks)

Whilst this method would decrease toxicity to *S. cerevisiae* due to lower ethanol exposure, this would occur because plants are degrading the bioethanol itself, which would greatly decrease its yield. This renders this particular method counterproductive and therefore unsuitable.

Another method proposed by which ethanol production can be optimised is by introducing a gene coding for a synthetic cellulase into either sugarcane or yeast cells and causing the gene to be expressed in large quantities. Cellulase breaks down cellulose, a major component of plant cell walls, into glucose.

- e. Explain, with reference to cellulase's function, how the overexpression of the cellulase gene would lead to increased rates of bioethanol production. (2 marks)

Cellulase overexpression would lead to a high rate of glucose production from cellulose; the high concentrations of glucose substrate increase the rate of the enzyme-catalysed reaction pathway of anaerobic respiration, leading to a high rate of bioethanol production.

- f. Explain whether it is more suitable to introduce the cellulase transgene into sugarcane cells or yeast cells in aiming to optimise bioethanol production. (2 marks)

Yeast cells. Plant expression (and overexpression) of cellulase would be incompatible with plant life, as it would lead to the breakdown of its cell wall; the plant would then be unable to produce further cellulase. Yeast cells would not be subject to this restriction.

Question 26 (10 marks)

A scientist isolated mitochondria from a muscle cell and placed them inside an appropriate solution with a suitable substrate, ADP and inorganic phosphate, as well as other relevant coenzymes. Oxygen was bubbled through the solution. The scientist decided against using glucose as a substrate for this experiment. Each solution would be placed at a different temperature.

- a. Why is glucose not suitable as a substrate? (1 mark)

Glucose is not able to be a substrate for the Krebs cycle or the link reaction, which occur in the mitochondria.
Substrate of glycolysis, which occurs in the cytosol and hence not in any isolated mitochondria.

- b. How would you expect the oxygen levels to change around the mitochondria? Explain. (2 marks)

They should decrease around the mitochondria.

It will be performing aerobic respiration, which requires oxygen as an electron acceptor, and hence, it will be absorbed from the environment.

- c. Why is it important for an electron acceptor to be present in aerobic cellular respiration, and why does the Krebs cycle also not occur in the absence of one? (3 marks)

The high-energy electrons and protons that are left over from the electron transport chain can cause damage to cellular membranes, which could result in cell death.
The Krebs cycle doesn't occur as it produces these high-energy electrons and protons from the breakdown of Acetyl CoA. We want to stop it at the source; otherwise, there will be a build-up of these high-energy electrons and protons in NADH and FADH₂, which, as mentioned, could be damaging.

- d. What would you expect the differences in the oxygen level changes to be between the different solutions? Explain. (2 marks)

At higher temperatures, there should be a greater decrease in the level of oxygen surrounding the mitochondria – due to the increase activity of enzymes involved in aerobic cellular respiration increasing its rate.

- e. What other factors would need to be maintained if this experiment is to be valid? Name 2. (2 marks)

The pH of the solutions.

The level of oxygen bubbled.

The level of the chosen substrate (to keep yourself safe say this otherwise you might lose a mark twice if you chose the wrong substrate earlier.)

Any 2 of above – if other answers then check with aaliyan.

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Question 27 (8 marks)

In most mammalian cells, the ATP produced from the complete breakdown of glucose occurs by a series of chemical reactions. Coenzymes also play an important role in glucose metabolism.

a. Describe the roles of coenzymes in the formation of ATP. (3 marks)

Coenzymes are organic molecules that bind to the active site of enzymes, involved in the formation of ATP via aerobic + anaerobic pathways, improving efficiency, such as of the catalyst of reactions involved in ATP production. "NAD + and FAD Further, they can act as electron + proton carriers received from the breakdown of glucose to be involved in ATP production at the ETC.

A student measured the effect of oxygen on the metabolism of glucose in red blood cells at 37°C and observed the results shown in the table below.

Effect of oxygen availability on the metabolism of glucose by red blood cells

Oxygen	Glucose consumed (n moles/ 10^7 cells/hour)	Lactate produced (n moles/ 10^7 cells/hour)
Absent	1.6	3.2
Present	1.6	3.2

b. Why are the results the same irrespective of the presence of oxygen? Explain your answer. (3 marks)

Red blood cells have no nucleus + other organelles, including mitochondria. Hence, they are UNABLE to perform aerobic cellular respiration and Lactic acid) term always obtain energy through anaerobic. Therefore, there is no difference in lactate production in the presence or absence of oxygen.

During outbreaks of citrus canker, the infected trees and fruit are removed and burned. Recently it has been proposed that instead of being burnt, the trees could be used to produce biofuel.

The first stage in the production of biofuel from infected trees could involve a technique known as steam explosion. This involves using very hot steam at high pressures inside a pressure chamber to break open the cells from the trees. An investigation was conducted to determine the relationship between temperature and the percentage of cells broken open during the steam explosion technique. The results are shown below.

Temperature inside pressure chamber (°C)	Percentage of cells broken open
20	36
80	47
150	57
200	85
250	94

- c. Scientists decided to use the steam explosion technique at 250°C. The temperature was then lowered to 35°C and yeast was added to the ruptured cell mixture.

Why did the scientists decide to do each of the following?

- i. Use 250°C. (1 mark)

_____ Ensures the highest percentage of cells (94%).
broken open _____

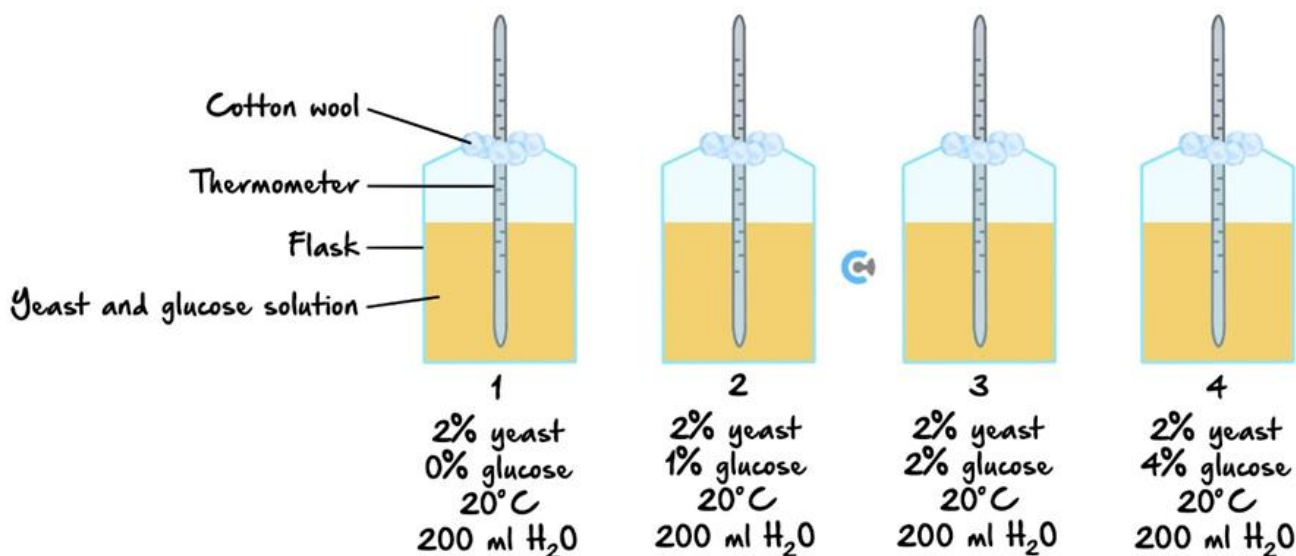
- ii. Lower to 35°C and add yeast. (1 mark)

_____ Optimal temperature of yeast and allow.
the yeast to produce ethanol via anaerobic fermentation! _____

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Question 28 (9 marks)

Four insulated flasks containing yeast (*Saccharomyces cerevisiae*), glucose, and a variety of other conditions were set up during a Biology class. The setup is illustrated in the diagram below.



- a. What was the purpose of the cotton wool in this experiment? (1 mark)

The purpose of the cotton wool in this experiment is to prevent contamination while allowing gases to escape.

- b. The temperature of each flask was recorded every 30 minutes for 24 hours. The maximum temperature increase of each flask was recorded in the table below.

Flask number	1	2	3	4
Maximum temperature increase (°C)	1	10	13	15

- i. Explain why there was an increase in temperature in flask 1. (2 marks)

The yeast in flask 1 already had organic molecules within it, such as glucose. Glucose was broken down by respiration (which is a catabolic process), and heat was produced, which caused the flask to increase in temperature.

- ii. Predict the maximum temperature increase that would be observed if there was a fifth flask that contained 8% glucose (all other variables are the same as above). (1 mark)

15°C ± 1°C (if a graph was plotted, the temperature would be seen to level off.)

- c. The water that was used throughout the series of experiments had been aerated. However, if the water had been boiled prior to the experiment, the yeast would have been exposed to anaerobic conditions.

- i. Discuss the difference/s that would be apparent in flask 4 during the 24-hour experiment if the conditions were anaerobic compared to aerobic. (1 mark)

Presence of ethanol

Note: Whilst fewer yeast cells, lower temperature increase and carbon dioxide could be accepted as answers, students must include ethanol.

- ii. Discuss the benefit to the yeast of aerobic conditions rather than anaerobic conditions. (1 mark)

Aerobic respiration is more efficient at providing energy (36 ATP) compared to anaerobic respiration, which provides less energy (2 ATP).

Respiration is a complex process that occurs in different cellular locations and involves three main stages.

- d. Discuss the main events of glycolysis. Include in your answer the cellular location, the inputs and the outputs of the process. (3 marks)

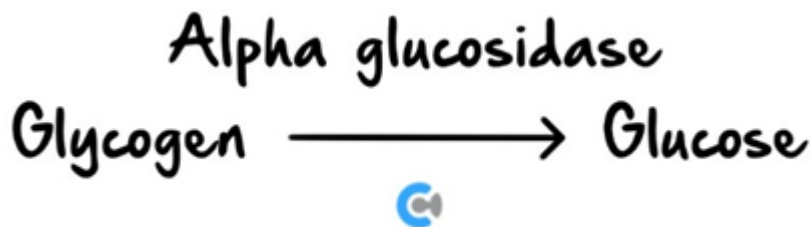
Glycolysis occurs in the cytosol of the cell.

The input is glucose, which is broken down by enzymes. Outputs include pyruvate, ATP and NADH.

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Question 29 (10 marks)

Infantile-onset Pompe disease (IOPD) is an inherited metabolic disorder that results from a mutation in the GAA gene. Symptoms include an accumulation of glycogen in the heart, liver, and skeletal muscles of the body. This can cause damage to the cells in the heart and liver, muscle weakness, and death. A functioning form of this gene produces the enzyme acid alpha-glucosidase (GAA), an enzyme responsible for the breakdown of glycogen in the body.



There are a number of mutations in the GAA gene that can result in a non-functioning enzyme.

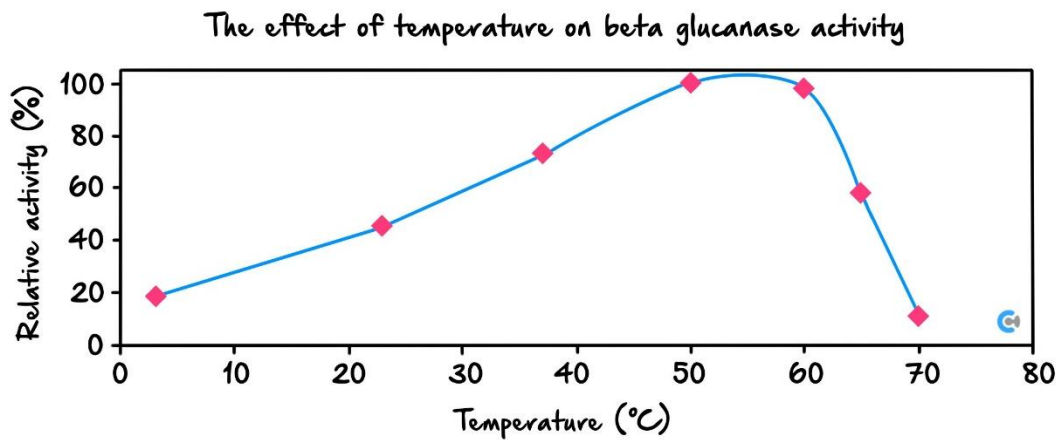
Normal	Gly	Pro	Arg	Asn	Ala	Gln
	GGG	CCC	CGG	GAT	GCC	CAG

Mutation	Gly	Pro	Arg	Asn	Ala	Gln
	GGG	CCC	CGG	GAT	GCC	CAG

- a. Explain why the inability to break down glycogen can cause damage to heart and liver cells. (4 marks)

Glucose is the main energy source for aerobic cellular respiration.
 No glucose means less ATP is formed.
 No energy for chemical processes.
 → cells will die.

To ensure that the enzyme is able to work effectively, it is important that the temperature of *Saccharomyces cerevisiae* is kept at a constant level.



- b. Use the graph above to identify the optimum temperature for *Saccharomyces cerevisiae* in the brewing process. (1 mark)

50-60°C

- c. Explain why a temperature of 10°C is not used in the brewing process. (2 marks)

10°C is too low temperature, molecules move slower decreasing collisions between enzyme and substrate, slowing the rate of ethanol production.

At the end of fermentation, portions of the yeast can be removed from the solution for the next brew. The top part of the solution will contain younger yeast, while the lower part of the solution will contain 'aged' yeast. Brewers claim that aged yeast cultures produce a 'better' beer with an increased ability to use the sugars to produce a high yield of ethanol and other flavours, as well as an increased flocculation potential (frothing when poured).

- d. Identify the inputs and outputs of the fermentation process. (2 marks)

Inputs	Outputs
inputs: glucose + ADP + Pi (1)	outputs: ethanol, CO ₂ , 2 ATP (1)

- e. Identify a coenzyme that is part of the process of fermentation. (1 mark)

NAD or ADP

Question 30 (4 marks)

The emerging Australian biofuels industry hopes to use farming waste such as plant stubble for profit within the next decade. Bioethanol is a liquid fuel that is chemically derived from cellulose-rich plant stubble, making it economically attractive. Bioethanol burns much more cleanly than fossil fuels, which reduces pollution and originates from renewable resources, which is a crucial factor for a sustainable future.

- a. Bioethanol is one example of a biofuel. Identify **one** other example of a biofuel. (1 mark)

One other example of a biofuel is biodiesel.

- b. How is bioethanol produced from cellulose-rich plant stubble? (2 marks)

When the cellulose in the plant stubble is exposed to certain conditions, such as a specific temperature and pH, enzymes convert the cellulose into glucose.

The glucose is then exposed to anaerobic conditions in a cellular environment, such as yeast or algae cells, to form bioethanol.

- c. Explain why biofuels are regarded as renewable energy resources. (1 mark)

Biofuels are renewable energy resources because they are produced using natural resources that constantly replace themselves and thus never run out.

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Question 31 (2 marks)

Plant materials containing cellulose and other polysaccharides are reacted with acids to break them down to produce glucose. This glucose is then used by yeast cells for fermentation.

a. Why is fermentation important for yeast cells? (1 mark)

To provide energy or ATP.

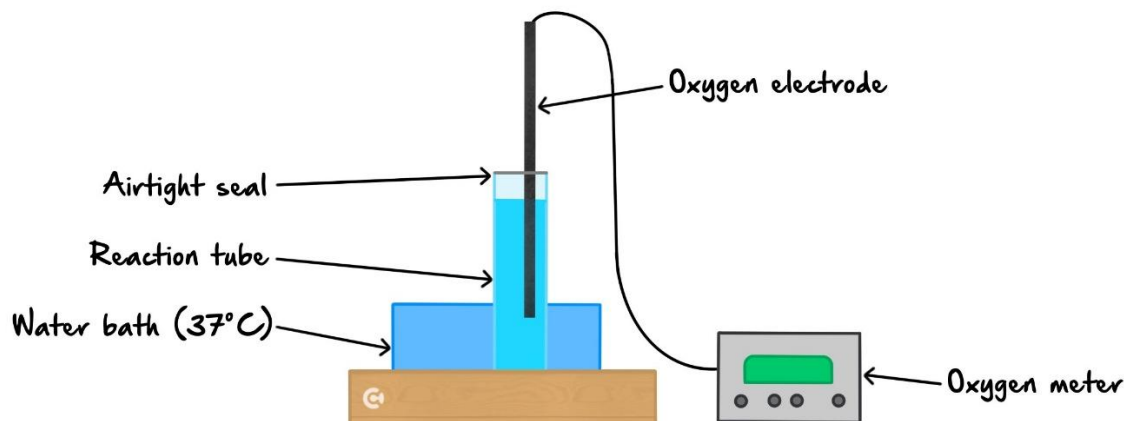
b. What are the products of fermentation in yeast cells? (1 mark)

ethanol/alcohol
carbon dioxide
ATP

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Question 32 (6 marks)

The apparatus shown below was used in a series of experiments to study aerobic respiration.



In three different experiments, the reaction tube initially contained the following:

- Suspension of mitochondria.
- Cytosol of cells from which the mitochondria had been removed.
- Suspension of mitochondria and cytosol of cells.

The temperatures and pH of the mixtures within the reaction tubes were carefully controlled so as not to damage the mitochondrial structure or any of the enzymes.

In each experiment, a solution containing glucose was first added to the mixture in the reaction tube, and the oxygen concentration was measured for three minutes. Then, a pyruvate solution was added, and the oxygen concentration was measured again for three minutes.

Using your knowledge and understanding of aerobic respiration and mitochondria, complete the tables below with your prediction of the change in oxygen concentration of the mixture in the reaction tube after the addition of each substance and give a reason for your prediction.

Experiment 1 - Suspension of mitochondria

Substance added	Change in oxygen concentration (increase/decrease/no change)	Reason
glucose	no change	Glucose is not metabolised (broken down) by the mitochondria.
pyruvate	decrease	Pyruvate is a substrate of the Krebs

Experiment 2 - Cytosol of cells from which the mitochondria had been removed

Substance added	Change in oxygen concentration (increase/decrease/no change)	Reason
glucose	no change	Glycolysis is anaerobic (or glucose converted to pyruvate but no mitochondria so oxygen is not used).
pyruvate	no change	No aerobic breakdown of pyruvate.

Experiment 3 - Suspension of mitochondria and cytosol of cells

Substance added	Change in oxygen concentration (increase/decrease/no change)	Reason
glucose	decrease	Glucose is converted into pyruvate, which is metabolised by the mitochondria using oxygen.
pyruvate	decrease	Pyruvate is metabolised by the mitochondria in a process that uses oxygen.

Question 33 (3 marks)

Briefly summarise the process of making bioethanol from sugarcane.

Sugarcane is harvested and processed before being fermented with yeast, undergoes alcoholic fermentation to produce ethanol and CO₂, in anaerobic conditions. This is then collected and purified before being used as bioethanol.

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Question 34 (9 marks)

A study investigated the effect of a drug called **Metabozone**, which inhibits the function of the enzyme citrate synthase, critical for the Krebs cycle. Researchers analysed glucose metabolism in actively growing muscle cells in two groups. One group was treated with 3 micromoles of **Metabozone**, while the other was not. The results are shown below:

Metabozone (micromoles)	Glucose Consumed (micromoles/ 10^6 cells)	Lactate Produced (micromoles/ 10^6 cells)
0	15.20	6.12
3	30.45	55.80

- a. Explain why both glucose consumption and lactate production change when cells are treated with 3 micromoles of **Metabozone**. (2 marks)

In the presence of the drug **Metabozone**, citrate synthase is inhibited, preventing pyruvate from entering the Krebs cycle. As a result:

- The cell consumes more glucose to meet its energy demands via glycolysis, which is less efficient than aerobic respiration.
- Lactate production increases because the cell relies on anaerobic fermentation to regenerate NAD⁺ for glycolysis.

- b. For cells that were not treated with **Metabozone**, from which metabolic pathways would they be obtaining most of their energy, and where do these pathways occur in the cell? (2 marks)

For cells not treated with **Metabozone**, energy is primarily obtained through:

- **Metabolic Pathways:** Glycolysis, Link Reaction, Krebs Cycle, and Electron Transport Chain.
- **Locations:**
 - ⚙ Glycolysis: Cytoplasm.
 - ⚙ Link Reaction, Krebs Cycle, and Electron Transport Chain: Mitochondria (matrix for the first two and cristae for ETC).

- c. **2-Deoxy-ATP (2-DATP)** inhibits the activity of the enzyme **phosphofructokinase (PFK)**, a key enzyme in glycolysis that converts fructose-6-phosphate to fructose-1,6-bisphosphate. You can measure the formation of fructose-1,6-bisphosphate in muscle cells.

Design a controlled experiment to determine whether **2-DATP** is a competitive or non-competitive inhibitor. In your response, include the setup, controls, and expected results for each type of inhibitor. (5 marks)

Experiment Design for 2-DATP:

- To determine if 2-DATP is a competitive or non-competitive inhibitor for phosphofructokinase (PFK), set up the following experiment:

Steps:

1. Prepare multiple test tubes with varying concentrations of fructose-6-phosphate (substrate).
2. Add the same concentration/volume of 2-DATP to each test tube.
3. Include a control test tube with no 2-DATP.
4. Maintain constant conditions (e.g., temperature, pH).
5. Measure the amount of fructose-1,6-bisphosphate formed under each condition.

Results Interpretation:

➤ **Competitive Inhibition:**

- Increasing the substrate (fructose-6-phosphate) concentration will overcome inhibition.
- At high substrate concentrations, the rate of fructose-1,6-bisphosphate formation will approach the same level as the uninhibited control.

➤ **Non-competitive Inhibition:**

- Increasing the substrate concentration will not restore the enzyme activity.
- The formation of fructose-1,6-bisphosphate will remain lower than the control across all substrate concentrations, as the inhibitor affects the enzyme's active site or functionality irreversibly.

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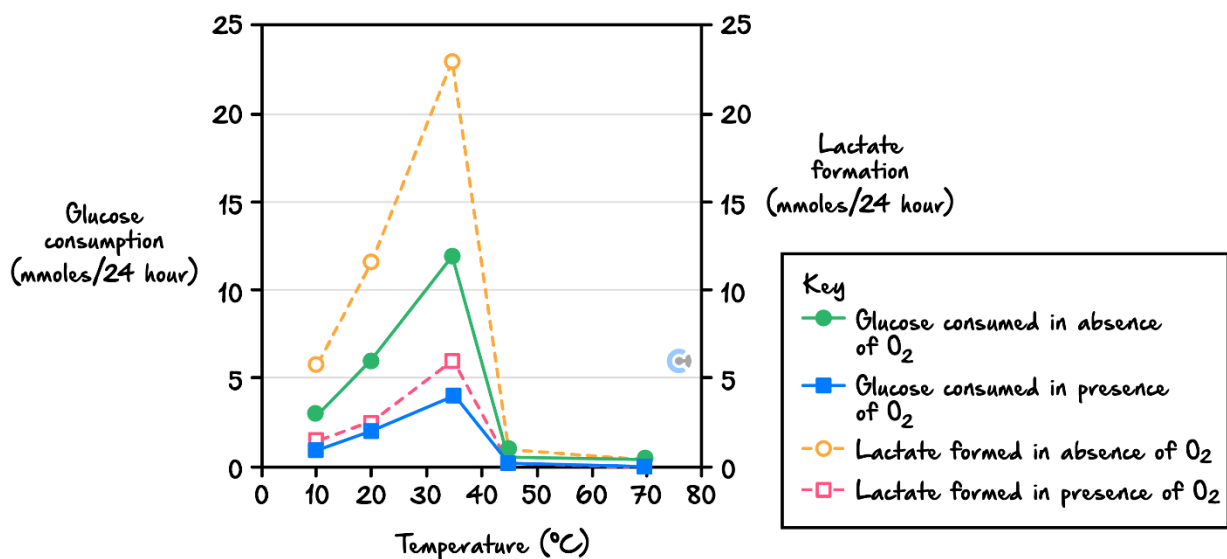
Question 35 (12 marks)

- a. Metabolic pathways possess a number of key regulatory enzymes. If one of these enzymes were inhibited using a non-competitive inhibitor, explain how this inhibitor would affect the activity of this enzyme. (2 marks)

A non-competitive inhibitor binds to a site on the enzyme away from the active site, causing a change to the active site. This results in the substrate no longer binding to the enzyme, or the rate of product formation is lower, or the enzyme cannot catalyse the reaction as efficiently.

A student was conducting an experiment using cultured human kidney cells. The student added the same number of cells suspended in the same volume of nutrient solution to 10 identical containers. Each container was then placed into its own incubator.

Ten incubators were set at one of five temperatures, 10°C, 20°C, 35°C, 45°C, or 70°C. At each temperature setting there were two incubators. One incubator at each temperature, setting was supplied with oxygen while the other was not. After 24 hours, the student measured the amount of glucose consumed and the amount of lactate produced in each cell population. The results they obtained were plotted below.



- b.
- i. What were the values for glucose consumption and lactate production at 35°C for the cells grown in the presence of oxygen? (2 marks)

Glucose consumption: Glucose consumption at 35°C is 4 moles/24 hours.

Lactate production: Lactate production at 35°C is 6 moles/24 hours.

- ii. Explain why the consumption of glucose and the production of lactate by these cells at 35°C was different if they were grown in the presence or absence of oxygen. (5 marks)

Students were required to recognise that glucose is consumed in both the presence and absence of oxygen and were then required to account for the results both in terms of the presence and absence of oxygen.

When there is no oxygen, glucose/pyruvate is converted into only lactate. Double the amount of lactate is produced for every one glucose consumed through glycolysis and fermentation, or more glucose will need to be consumed as less ATP/energy is produced. When oxygen is present, glucose/pyruvate can also be converted into water and carbon dioxide. This leads to a smaller ratio of lactate formed compared to glucose consumed, or a greater amount of ATP/energy is produced through aerobic cellular respiration to meet its energy requirements.

- c. Explain why both glucose consumption and lactate production in the cells grown at 45°C were lower than those grown at 35°C. (3 marks)

At 45°C, enzymes involved in biochemical processes begin to denature, resulting in a decreased rate of reaction compared to when there is optimal glucose consumption and lactate production at 35°C.

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