2010 'A' Level
H2 Biology
Mark Scheme

PAPER 1 (MCQ)

1	**C**
2	**A**
3	**C**
4	**C**
5	**D**

6	**D**
7	**C**
8	**D**
9	**C**
10	**B**

11	**B**
12	**A**
13	**A**
14	**B**
15	**C**

16	**D**
17	**D**
18	**B**
19	**D**
20	**B**

21	**C**
22	**C**
23	**B**
24	**D**
25	**C**

26	**D**
27	**C**
28	**B**
29	**C**
30	**C**

31	**A**
32	**D**
33	**B**
34	**B**
35	**D**

36	**A**
37	**A**
38	**B**
39	**B**
40	**D**

PAPER 2 (CORE)

QUESTION 1

(a)
A – Mitochondrion;;
1 Presence of cristae;;
2 Double membrane ;;
Reject rod-shaped, reference to cisternae

B – Rough endoplasmic reticulum;;
1 Presence of ribosomes;;
2 Parallel stacks of cisternae;;
Reject references to sac-like or tube-like structures as not visible on the diagram

Examiner's comments: The features of mitochondria were well known, although some talked about rod-shapes or confused cristae with cisternae. Candidates must ensure that when asked for 'visible features' that they only state features that they can see on the electron micrograph. Some listed sacs or tubes as features of the rough endoplasmic reticulum. These were not clearly visible on the diagram and reflect what the candidate had learnt rather than what they could see.

(b)
(i)
A – Synthesis of ATP during aerobic respiration;;
B – Protein synthesis (by the ribosomes attached to it) / transport of protein / vesicle formation;;

(ii)
1 ATP synthesized by A is hydrolyzed to release energy for protein synthesis during translation e.g. amino acid activation;;
2 Ribosomes on B synthesizes respiratory enzymes/proteins needed for aerobic respiration;;

Examiner's comments: Majority of candidates explained how the products of A were useful in B, rather than vice versa

(c)
1 Compartmentalisation so that different conditions are established to maintain optimum conditions for enzyme reactions; ;
2 Increase surface area for metabolic reactions e.g. attachment of more electron carriers and ATP synthase for oxidative phosphorylation;;

QUESTION 2

(a)

1 DNA polymerase works only in one direction, from 5' to 3', adding new nucleotides to the **available** 3'-OH end of a pre-existing polynucleotide chain;;
2 DNA strands are antiparallel
/DNA strands run in the 5'→ 3' and 3'→ 5' directions ;;

(b)

1 The parental DNA molecule is a double helix consisting of **two strands**, each strand serves as a **template** for the synthesis of a new complementary daughter strand;;
2 each of the **two new DNA molecules** contains **one new and one original/parental strand** of DNA;;

Examiner's comments: Care must be taken when using the terms like 'strand' when describing a DNA molecule. Weaker candidates referred to the newly synthesized DNA molecule as being 'a strand'.

(c)

1 Helicase;;
2 (DNA) Primase;;
3 RNA primer ;;
OVP
4 ATP;;
5 Single-stranded binding proteins;;
6 Deoxyribonucleotides;;
7 DNA ligase;;

Examiner's comments: Candidates must be encouraged to tailor the length of their answers to the space provided. They must also be precise with terminology used. Imprecise answers, such as 'DNA primer' and 'RNA primase' were not allowed.

(d)

1 During DNA replication, complementary base pairing may result in one or more incorrectly paired bases which is not corrected by proof reading of DNA polymerase;;
2 Resulting in a substitution where one nucleotide in the gene sequence is being replaced by another nucleotide;;

Examiner's comments: Candidates need to use the marks allocated as a guideline to the number of points or detail that they include. Most candidates confined their answers to describing one form of mutation, such as substitution, but did not give clear enough additional detail for full credit to be awarded.

(e)

1 In sickle cell anemia, substitution of **thymine for adenine** at the 17th nucleotide of the **gene coding for β-globin chain of hemoglobin** results in a change in (mRNA) **codon** from GAA to GUA and subsequent change of the 6th **amino acid** from **glutamic acid to valine** ;;

2 Glutamic acid is hydrophilic whereas valine is hydrophobic, the tertiary structure of the molecule changes due to the **change in R groups** of the amino acid;;

3 Hb S stick to each other via their hydrophobic regions and polymerize into long fibres inside the red blood cells, deforming them into sickle shape;;

4 Due to the sickle shape, red blood cells clump and clog small capillaries, obstructing other cells from moving through the capillaries, leading to other symptoms such as physical weakness, pain, or organ damage
/ Sickle-shaped red blood cells have a shorter lifespan compared to normal cells and hemolyse readily resulting in anemia and also making them ineffective in transporting oxygen gas;;

Examiner's comments: Candidates must ensure that they describe the whole process and not miss the initial effects in an attempt to include all of the detail they have learnt about a given mutation. Many candidates began their responses beyond the immediate effect of a mutation, namely the alteration in the base sequence or codon and the subsequent change in amino acid.

QUESTION 3

(a)
A – Capsid head
B – Contractile sheath / Tail sheath
C – Base plate

(b)
1 Adsorption of phage through binding of **attachment sites of tail fibres** to **receptor sites on bacterium;;**
2 Release of lysozyme which degrades host bacterial cell wall;;
3 Tail sheath contracts to drive a hollow tube into bacterium cell membrane; Phage DNA injected into cytoplasm of bacterium;

Examiner's comments: Candidates need to be clear of the difference between the bacterial cell wall and bacterial cell membrane

(c)

	Lytic cycle	Lysogenic cycle
1	Arrest of bacterium host gene expression when phage enters	No arrest of bacterium host gene expression when phage enters
2	Bacterial chromosome hydrolysed to provide nucleotides for phage genome replication	No bacterial chromosome hydrolysis
3	Viral genome does not integrate into host genome	Viral genome integrates into host genome, known as prophage
4	No latency period	Latency period where bacteria genes are not expressed in bacterium host
5	Bacterium host is lysed	No lysis of bacterium host (before induction phase)

(d)
(i)
1 Genome enclosed by protein capsid;;
2 Rod-shaped TMV similar to rod-shaped tail sheath;;

(ii)
1 RNA genome in TMV vs DNA genome in T4 phage;;
2 No tail fibres in TMV vs presence of tail fibres in T4 phage;;
3 No tail sheath / collar in TMV vs presence of tail sheath in T4 phage;;
[Any 2]

QUESTION 4

(a)

A Nucleosome;;
B DNA;; Reject: Linker DNA

(b)

1 To make long DNA molecule more compact to fit in the nucleus;;
2 To prevent breaking or damage to DNA;;
3 Allows for regulation of gene expression / transcription (by interactions between DNA and histones);;

(c)

(i)

1 Histone methylation results in the binding of proteins which lead to tighter nucleosomes / more compact chromosomes;;
2 Prevents RNA polymerase and transcription factors from binding to promoter to initiate transcription;;

Examiner's comments: Candidates must study diagrams carefully to help with answering questions.

(ii)

1 many genes are not required in that one type of differentiated cells thus their gene expression is switched off;;
2 large size of genome (thus the need for making it compact to fit into nucleus);;
3 eukaryotic genome contains many non-coding regions (thus, need to make these regions compact to be packed into the nucleus and not interfere with areas where genes are actively expressed);;

Examiner's comments: Candidates need to carefully target the depth of their answers to the number of marks allocated. Many restricted their potential by repeating ideas that most genes are not required in differentiated cells without going on to **other ideas**.

(iii)

1 genes involved in the same function grouped into operons;;
2 expression of genes regulated by a single promoter and operator site;;
3 binding of repressor proteins to operator prevents RNA polymerase from binding to promoter / prevent access of structural genes by RNA polymerase (thus, preventing of transcription of structural genes);;

QUESTION 5

(a)

1 Continuous variation;;

(b)

1 phenotype is due to interactions; between polygenes/more than 3 genes;

2 there is an additive effect of the genes; but each gene alone has little overall effect on the phenotype;

3 environment affects the phenotype to varying degrees in different individual plants, causing a range of phenotypes;

4 independent assortment of genes during metaphase I and anaphase I of meiosis; ensures that each individual possess a range of genes from any polygenic complex;

Examiner's comments: Weak candidates tend to address the phenotype range for F1 and F2 generation and why its in the middle, referring the parents as homozygous, F1 being heterozygous/co-dominance of alleles. Question is just asking for reasons for having a range of phenotypes instead of discrete ones.

(c)

1 genotypes of F2 more varied than F1 as a result of gamete formation (in F1 heterozygotes) during meiosis;;

Processes that produce variations include:

2 independent assortment of homologous chromosomes/random assortment of genes;;

3 crossing over and recombination at the chiasmata at prophase I;;

4 random fusion of gametes during fertilization;;

OVP:

5 there may be presence of multiple alleles (which increases the variability of genotypes);;

6 different genotypes will respond to the same environment differently; leading to a wide range of phenotypes in F2;;

Note: F1 – variation is due to only environment; genotype is not considered as individuals are all genetically identical/ all heterozygous

QUESTION 6

(a) (i)
1 ref. reduced NAD and FAD;;
2 from glycolysis, link reaction, Krebs cycle;;

(ii)
1 electrons are accepted by oxygen (final electron acceptor);;
2 together with protons/H^+ to form water;;

(iii)
1 ref. high H^+ concentration constituting a proton-motive force;
potential energy released as H^+ ions diffuses from intermembrane space to matrix;
2 energy coupled to ATP synthase complex;
to form ATP from ADP and Pi;

(b)
1 ref. ETC consists of electron carriers arranged in decreasing energy levels;;
2 transfer of electron from an electron carrier of higher to lower energy level releases energy;;
3 to pump H^+ ions;
from matrix to the intermembrane space;

(b) [Any 3]

	Photophosphorylation	**Oxidative phosphorylation**
1	produces oxygen through photolysis	Oxygen is used as final electron acceptor
2	water is used for photolysis	Water is produced
3	Dependent on presence of light	Not dependent on light, dependent on presence of oxygen
4	Located in thylakoid membrane of chloroplasts	Located in inner membrane of mitochondria

QUESTION 7

(a)

1 The sea separating the islands led to geographical isolation / prevent gene flow between evolving populations;
Different selection pressures on the different islands ;

2 Genetic variations exist among the small flying ducks;
Natural selection is the outcome of differences in survival and reproduction among individuals of different phenotypes among the small flying ducks / ref. adaptive radiation;

(b)(i)

1 Phylogeny describes the evolutionary history of a species or a group of species from their common ancestors;;
2 Fossils shown are incomplete/damaged;
thus organisation of species according to particular characteristics cannot be done;

(ii)

1 Quantifiable – protein, nucleic acid sequence data are precise and accurate and easy to quantify;;
2 Objective – Molecular data can be easily described in an unambiguous manner, facilitating the objective assessment of evolutionary relationships;;

(c)

1 Hawaii had only formed recently so insufficient time for evolution of flightless birds;;
2 Flightless birds are unable to cross the sea separating the islands;;

QUESTION 8

(a) Describe how the prokaryote, *E.coli*, is able to respond to varying concentration of lactose. [6]

Presence of lactose

1. Lactose enters via the qualified leaky membrane and isomerizes to form allolactose, an inducer;;
2. allolactose binds to the allosteric region of the lac repressor ;
   alters its conformation at the DNA binding-site;
3. lac repressor is inactivated and is **no** longer able to bind to the operator (lacO) (operator is now exposed);;
4. RNA polymerase can access and transcribe the structural genes (lacZ , lacY and lacA);
   β-galactosidase, lac permease and lactose transacetylase are produced;

Presence of glucose:

5. in the presence glucose, even if lactose is present;
   transcription of the lac operon proceeds at a **low** level;
6. **as glucose concentration increases, cAMP levels decreases;**
   without cAMP, CAP is inactive does and does not bind to the CAP binding site;

Examiners' comment:
The question was clear in asking for responses to varying lactose concentrations, so no credit was awarded to answers describing events in the absence of lactose.

(b) Explain the role of glucagon in regulating blood glucose concentration in humans. [8]

1 produced by α-cells of the islets of Langerhans in the pancreas
2 glucagon released when blood glucose levels are below set point / norm of 70 mg per 100ml
3 travel via the bloodstream and bind to glucagon receptors on liver cells (**REJECT** muscle cells because glucagon has no effect on muscle glycogen; Reject Liver as a whole organ)

4 increase glycogenolysis / breakdown of glycogen to glucose
5 increase gluconeogenesis / use of other substrates (eg. fatty acids, lactic acid, proteins) instead of glucose as the main fuel in respiration.
6 decrease glycogenesis / formation of glycogen from glucose

7 blood glucose level is raised back to the norm
8 inhibits further secretion of glucagon

Examiners' comment:
Candidates should refer to 'liver cells' instead of simply 'liver'.

(c) Outline the main stages of cell signalling. [6]

1 ligand-receptor interaction;
ligands bind to specific receptors on target cells as they have complementary shapes to the ligand-binding sites present on receptor molecules;
2 binding of ligand triggers conformational changes in the receptor;
resulting in its activation;

3 signal transduction
/ relay of signals from the receptors at the cell membrane to target molecules within the cells;
second messengers are needed to relay the signals;
4 phosphorylation cascade;
sequence of phosphorylation reactions where the activated relay protein phosphorylates and activates a protein kinase which in turn phosphorylates and actives the subsequent protein kinase;
5 signal amplification;
presence of small quantities of ligand to elicit a large response from a target cell as the number of activated molecules at each consecutive step of the transduction pathway increases;

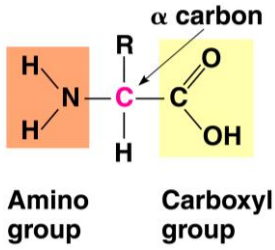
6 transduced signal triggers a specific cellular response;;

Examiners' comment:
There were some references to 'ligands', but the idea of 'specificity' was not commonly awarded. Candidates must be clear about where receptors are to gain credit.

QUESTION 9

(a) Describe the structure of an amino acid and how a peptide bond is formed with another amino acid [6]

Structure of an amino acid

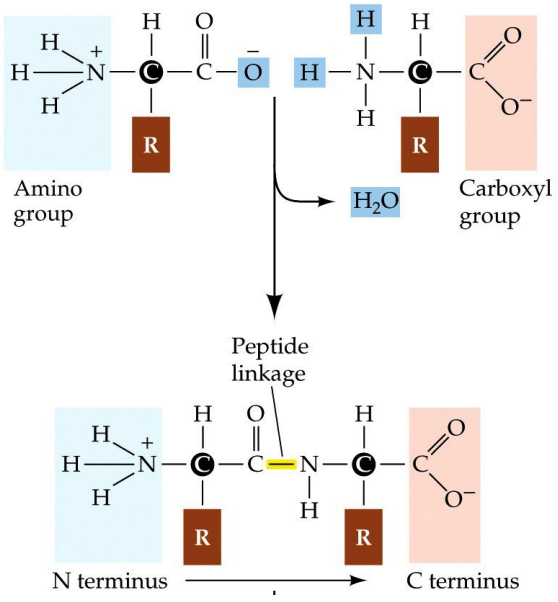


1 All amino acids have an amino (reject amine) group, a carboxyl group, a H atom attached to an α carbon;;
2 Variable R group in different amino acids ;;

Formation of peptide bond with another amino acid

3 Condensation reaction between two adjacent amino acids;
reaction involving loss of water;
4 Involving carboxyl group of first amino acid and amino group of following amino acid;;

5 Synthesis of covalent peptide bond occurs during **translation at the ribosomes;;**
6 Catalyzed by peptidyl transferase (found on the large ribosomal subunit);;



Examiner's comments: If candidates use a diagram, they must make sure that it is fully labeled and described to gain credit.

(b) Explain what is meant by primary, secondary, tertiary and quaternary structure of a named protein. [8]

Examiner's comments: When selecting a protein to describe, candidates need to ensure that they select one that will allow them to answer all parts of the question. Due to its lack of a tertiary structure, those **candidates that selected collagen struggled to gain full credit**.

Named protein: Haemoglobin

Primary structure

1 Refers to the number and sequence of amino acids making up each haemoglobin polypeptide (individual α and β subunits);
141aa for α subunit and 146 aa for β subunits;

2 Peptide bond involved in joining all amino acid monomers together;;

Secondary structure

3 Refers to the folding of the polypeptide into regular structures like the α helices and β pleated sheets;;

4 Regular structures held together by hydrogen bonds;
Formed between CO and NH groups of peptide bonds;

Tertiary structure [Max 2m]

5 Refers to the folding of the polypeptide chain into its unique 3-dimensional shape;
Amino acids far away in primary structure are brought close together (by R group interaction);

6 Non-polar/hydrophobic (side chains of) amino acids are buried in the interior;
Polar and charged/hydrophilic (side chains of) amino acids are on the surface;

7 Bonds involved in tertiary structure : hydrogen bonds, ionic bonds, hydrophobic interaction (between R groups);;

Quaternary structure

8 Refers to the arrangement of the polypeptide subunits within a protein that is made up of more than one polypeptide chain
/ spatial arrangement of more than one polypeptide chain ;
association of <u>2α and 2β subunits</u> to form functional haemoglobin molecule;

9 Bonds involved include hydrophobic interactions, hydrogen bonds and ionic bonds (between R groups of amino acids in the four subunits);;

(c) Outline how the structure of a named globular protein is related to its specific function. [4]

Examiner's comments: Care must be taken not to confuse haemoglobin with myoglobin.

[Any 4]

Molecular structure	Function
1. Globular and compact in shape	2. Allows maximum packing of haemoglobin into red blood cells for transport of oxygen.
3. Consists of four subunits, each capable of binding one oxygen molecule.	4. Facilitates transport of oxygen
5. Hydrophobic amino acid residues are buried in the interior of the globular structure while the hydrophilic amino acid residues are on the outside.	6. Soluble in an aqueous medium and hence a good transport protein for oxygen in blood.
7. Binding of first molecule of oxygen causes structural changes in other haem groups	8. Facilitates allosteric binding of oxygen to haemolgobin resulting in greater ease in loading / unloading oxygen
9. Presence of haem group	10. Fe^{2+} ion combines reversibly with oxygen and hence enhances the release of oxygen in metabolically active tissues.

PAPER 3 (APPLICATIONS)

QUESTION 1

(a)

(i)

1 Allows for independent replication of plasmid (and foreign gene);;
2 Multiple copies of plasmid (and foreign genes) within **one bacterium**;;

(ii)

1 Can be used to clone a wide range of foreign genes (due to a number of restriction sites for restriction enzymes);;
2 Allows for insertional inactivation of *lacZ* gene;
Which allows for subsequent selection of bacteria with recombinant plasmid;;

Examiner's comments: The most common (but wrong) response was to describe how a restriction enzyme would cut both the polylinker and the DNA flanking the foreign gene (but this does not explain the advantages).

(iii)

1 Allows for selection of transformed bacteria cells containing the plasmid;;
2 Bacteria cells without plasmid (and ampicillin resistance gene) cannot survive in ampicillin medium;;

Examiner's comments: Many candidates knew about the technique of selection by plating in ampicillin media but credit was not awarded as this knowledge was not applied to the context or the specific question asked.Weaker candidates confused bacteria that had been transformed and those that only contained the recombinant plasmid. The selection process involves the survival or death of bacteria not the plasmids.

(b)

(i)

1 Restriction enzymes used to cut <u>foreign DNA</u> ;;
2 to protect the bacterium; from attack by viruses / bacteriophages;

Examiner's comments: Candidates should be aware that they will not be asked for the same information in two different questions. Some candidates misread this question and repeated themselves in their answer to (b)(ii)

(b)(ii)

Similarity

1 Both require DNA ligase to produce the recombinant DNA molecule;;

Differences [Any 3]

SmaI	XmaI
1. Produces blunt-ended DNA ;	1. Produces sticky-ended DNA ;
2. Requires linkers to be added to blunt-ended DNA ;	2. No linkers need to be added;
3. Linkers need to be cut by another restriction enzyme to produce sticky ends;	3. No requirement for second restriction enzyme ;
4. Process requires longer time / more	4. Process requires lesser time / less

energy;	energy ;

Examiner's comments: (1) When asked for comparison, it is expected that candidates will write sentences containing information about each restriction enzyme, rather than sentences describe one and then sentences describing the other. This makes the comparison clear. (2) Tabulating answers to comparison questions such as this often makes the process easier and clearer.

QUESTION 2

(a)

(i)

1 tail cell is a somatic cell with diploid nucleus;
whereas the nucleus taken from an oocyte (a gamete cell) is haploid;
2 nucleus is obtained from cells of Rag2 mice;
ensures that embryonic stem cells generated are genetically identical to the cells of the Rag2 mice;
3 prevents Rag2 mouse's immune system from rejecting the injected embryonic stem cells;; (Reject: tissue rejection)

Examiner's comments: Candidates are reminded that terminology must be exact. It is not correct to say that there will be no tissue rejection, since only cells are involved.

(ii)

1 pluripotent: able to differentiate into almost any cell type to form any organ;
also multipotent but not totipotent;
2 capable of self-renewal;
by dividing continuously by mitosis to produce new stem cells;
3 Unspecialized and undifferentiated and thus able to differentiate into multiple specialized cell types during development into a fetus;
by differential gene expression;

(iii)

1 SCID is a recessive disease;
caused by simple loss-of-function mutation/possession of 2 recessive alleles;
2 Inserted normal allele is dominant;
one copy is enough to produce the functional gene product/protein (to mask effects of recessive mutation);

Examiner's comments: Candidates must ensure that they use gene and allele in the correct context.

(b)

1 random insertion of the transgene into host genome;

ref to e.g. inserted genes may not be expressed because they may be integrated into a highly condensed heterochromatic region
/ insertion of transgene disrupts the normal expression of cellular genes encoding proteins regulating cellular replication
/ cause the activation of an oncogene or it could inactivate a tumor suppressor gene or a gene involved in apoptosis;

2 limit on the size of the transgene;
(retroviruses) can only accept inserts of up to 8 kb of exogenous DNA ;
3 viral vectors can regain virulence;
generate immune responses, causing chronic inflammation and the rejection of the cells containing the transgene;
4 transient expression of the transgene;
if not integrated into host genome;

[Any 3]

(c)

Argument in favour of the use of 'cybrid' cells:

1 Ease of availability of cow or rabbit oocytes;;

2 Potential of human oocytes to develop into viable organism after fertilization, using such oocytes may be similar to murder;;

Argument against the use of 'cybrid' cells:

1 Infringe of animal rights / Act of animal cruelty;

2 Possibility of human with abnormal genomic makeup due to the influence of animal mitochondrial genes;;

Examiner's comments:
Candidates commit common errors based on the use of embryos, rather than oocytes. Vague statements such as "playing God" are not encouraged.

QUESTION 3

(a)

1 ref. **restriction digest** of the DNA samples **obtained from different test subjects** using same RE;;
2 ref. run gel electrophoresis by applying an electric current;
ref. agarose gel matrix as a molecular sieve;
3 ref. separation of DNA fragments based on molecular size;
smaller fragments travel faster towards the positively charged electrode; (Reject: travel further)

4 Gel is stained with ethidium bromide to reveal **different positions** of DNA fragments with varying number of repeats under UV light;;

Examiner's comments:
A generic description of electrophoresis would not, however, score full credit. Candidates were expected to use the information in the question and explain how you would obtain and use samples from different people and how they would behave under electrophoresis and therefore how they could be distinguished.

(b)

Denaturing:

1 Heat to 95ºC;;
2 DNA denaturation / Separation of DNA double helix into single strands;
ref. hydrogen bonds being broken ;

Annealing:

3 Temperature cooled to 55ºC;;
4 Annealing of (single-stranded) forward and reverse DNA primers;
to complementary sequences at the 3' ends of each single template strand;

Elongation:

5 Heat to 72ºC;;
6 Heat-stable/thermostable DNA polymerase catalyses the synthesis of a complementary DNA strand for each of the template strands;
extending primers in 5' → 3' direction / nucleotides added to 3' ends of both primers;

(c) (i)

Describe:

1 As the number of repeats of STR increases from 7 repeats to 10 repeats;
the temperature at which the DNA strands separates, increases linearly from 53.0ºC to 62.0 ºC;
2 As the number of repeats of STR increases further from 10 repeats to 12 repeats, the rate of temperature increase slows down, with temperature at which DNA strands separates, increasing from 62.0 ºC to 63.5ºC;;
(Note: ref to figures to support answer is mandatory)

Explanation:

3 Increasing number of repeats indicate a larger number of hydrogen bonds between the 2 strands of DNA;;
4 More heat energy needed to break bonds to separate double-stranded DNA (reject: higher temperatures needed);;

(c) (ii)

1 Temperature increase is not proportional to increase in number of repeats of STR / rate of temperature increase slows down as number of repeats increase; may not be possible to distinguish between the lengths of fragments with larger number of repeats as the temperatures at which DNA strands separates, for these fragments may be almost the same;

QUESTION 4 (SPA Planning)

4 Fig. 4.1 shows a biosensor that can be used to measure the concentration of glucose in a solution in mg/100 cm^3.

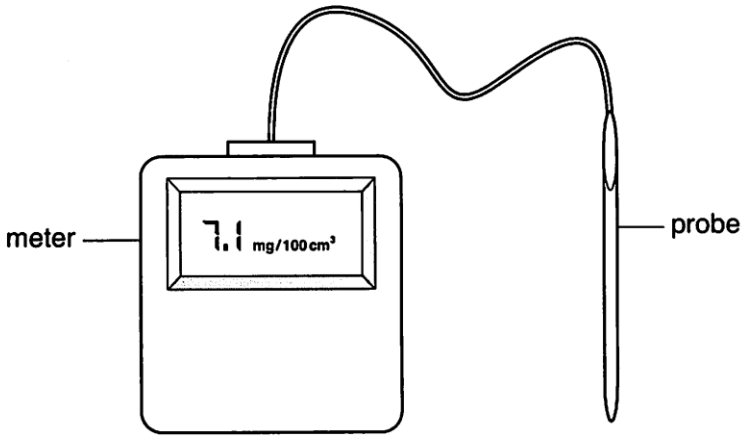


Fig. 4.1

An α-amylase obtained from a species of bacterium, *Bacillus licheniformis*, hydrolyses amylose to glucose.

Design an experiment, using a biosensor, to test the hypothesis that:

The rate of production of glucose is dependent on the concentration of α-amylase.

[Total: 12]

BACKGROUND/HYPOTHESIS

- When the **substrate** concentration is maintained at a **high** level and other conditions such as **temperature** and **pH** are kept **constant**, the **rate** of reaction is **directly proportional** to the **enzyme** concentration.

At low enzyme concentration

- At low enzyme concentration, adding more enzymes increases the rate of reaction.
- As **enzyme** concentration **increases**, the **frequency of successful collisions** between the enzyme and substrate molecules **increases**.
- With more enzyme presents, it is more likely that a substrate will bind to an empty active site on an enzyme.
- **More enzyme-substrate complexes** are formed
- Rate of reaction increases proportionally (**linearly**) with increasing enzyme concentration.

At very high enzyme concentration

- At very high enzyme concentration, the **substrate concentration** is the **limiting** factor.

- An increase in the enzyme concentration would not result in any further increase in the rate of reaction.
- Hypothesis: increasing the concentration of amylase will increase the rate of production of glucose from amylose and rate production of glucose will gradually plateau.

VARIABLES
Independent variable: enzyme (amylase) concentration; 5-6 different concentrations (20%, 40%, 60%, 80%, 100%); to be prepared by dilution from a stock enzyme solution.

Dependent variable: Rate of production of glucose per minute (mg $100cm^{-3}min^{-1}$), which is measured using the biosensor.

Variables to be fixed:
Temperature (to be fixed at 37^0C using a thermostatically controlled water bath)
pH (to be maintained at optimal level by using a suitable buffer)
Concentration and volume of substrate amylose: to be fixed at 1mol $^{dm-3}$ and$10cm^3$ respectively.

CONTROL
Replace the amylase with same volume of distilled water. This is to show that production of glucose is due to the effect of the enzyme.

PROCEDURE
1. Prepare $10cm^3$ of 5 different concentrations of amylase stock solution (100%) using the dilution table below.

Final concentration of enzyme solution/ %	Final volume of enzyme solution / cm^3	Volume of stock solution / cm^3	Volume of distilled water / cm^3
20	10.0	2.0	8.0
40	10.0	4.0	6.0
60	10.0	6.0	4.0
80	10.0	8.0	2.0
100	10.0	10.0	0.0

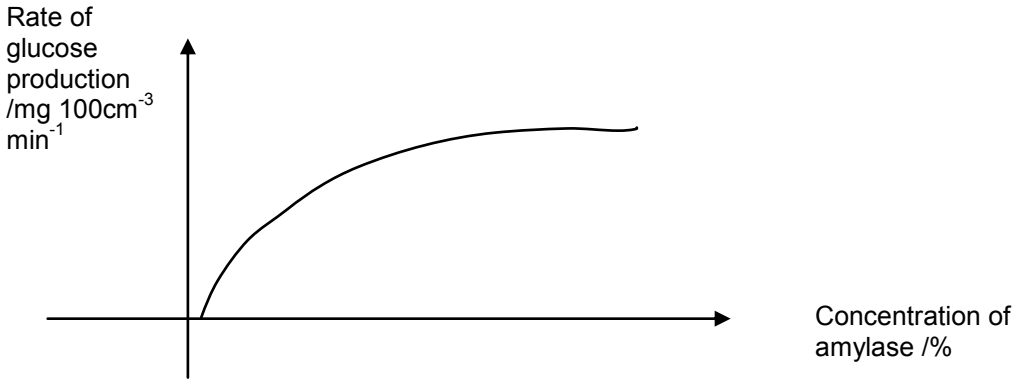
Formula: M1V1 = M2V2

2. Label 5 boiling tubes with the final concentration of each enzyme solution.
3. To the boiling tube labeled 100%, transfer $10cm^3$ of 1mol dm^{-3} amylose (substrate) and $1cm^3$ of buffer solution. Place the tube in 37^0C water bath and equilibrate for 5 min.
4. Equilibrate the 100% enzyme solution in the same temperature (water bath) for 5min. Add $1cm^3$ of 100% enzyme solution to the boiling tube.
5. After 1min, put the probe of the biosensor into the boiling tube and measure the concentration of glucose.
6. Repeat steps 2-5 for another 2 more sets of reading for 100% amylase.
7. Repeat steps 2-6 for 80%, 60%, 40%, 20% amylase.

RESULTS/CONCLUSIONS

Concentration of amylase solution / %	Concentration of glucose per min / mg $100cm^{-3}$ min^{-1}			Average concentration of glucose per min / mg $100cm^{-3}$ min^{-1}
	Reading 1	Reading 2	Reading 3	
20				
40				
60				
80				
100				

8. Plot a graph of Rate of glucose production /mg $100cm^{-3}$ min^{-1} against amylase concentration / %.



RISKS/PRECAUTIONS

-Broken glassware may result in cuts
-Ensure that all broken glass are cleared immediately with broom and dustpan.

QUESTION 5

(a) With reference to solving demand for food in the world, explain, with examples, the significance of genetically engineering crop plants to improve:

(i) their quality; [6]

Plants with improved nutritional qualities

1 E.g. Golden rice;;
2 produced by transplanting genes from daffodil and bacteria;;
3 that encode for proteins involved in synthesis of beta-carotene;;
4 help prevent Vitamin A deficiency which leads to blindness and susceptibility to disease;;

Plants with delayed ripening

5 Eg. Flavr-Savr tomato;;
6 Polygalacturonase is normally responsible for the ripening process and tomatoes which ripen too fast may result in spoilages and reduced shelf life;;
7 Tomato plants are genetically engineered to carry an antisense gene of the enzyme, which encodes for an antisense RNA complementary to the mRNA that codes for the enzyme;;
8 delay in fruit ripening / spoiling during transport / improved shelf life;;
9 larger and has greater flavour;;

~~**OR**~~

~~5 Introduce a gene whose gene product blocks the biosynthetic pathway for ethane;;~~
~~10 ethene is hormone responsible for fruit ripening and tomatoes which ripen too fast may result in spoilages and reduced shelf life;;~~
~~6 delay in fruit ripening / spoiling during transport / improved shelf life;;~~
~~7 larger and has greater flavour;;~~

Examiner's comments:
For both quality and yield, candidates were awarded credit for a named crop, the source of the gene, the gene product in the crop, the problem and the solution. Credit was also available for further detail.

(ii) their yield; [6]

Pest-resistant plants

1 Eg. Corn / potato / broccoli / tomato plants which can produce the Bt toxin;
2 Crops are genetically engineered to carry the Bt toxin gene from the bacteria *Bacillus thuringiensis*;;
3 which is expressed to produce the Bt toxin;;
4 that specifically kills insect pests;;
5 When an insect pest ingests Bt toxin, enzymes in the caterpillar's stomach convert it into an insect-specific toxin, causing paralysis and death;;
6 crop losses can be reduced, leading increased profits in agriculture;;

Herbicide resistant plants

7 E.g. Glyphosate-resistant soybean / tomato;;

8 glyphosate in herbicides works by inhibiting an enzyme EPSP synthetase, which plants require to make essential aromatic amino acids;;

9 Crops are genetically engineered to carry the EPSP synthase gene which is then expressed to produce high levels of EPSP synthase in the plant;;

10 This results in resistance towards glyphosate and genetically engineered crops are able to survive in the presence of glyphosate-containing herbicides while weeds are destroyed.

11 can spray herbicides to kill weeds without affecting crops;;
12 crop losses can be reduced, leading increased profits in agriculture;;

Examiner's comments:
For both quality and yield, candidates were awarded credit for a named crop, the source of the gene, the gene product in the crop, the problem and the solution. Credit was also available for further detail.

(b) Discuss the role that cloning in plants could play in such crop improvements. Include, in your discussion, any advantages or disadvantages of such cloning. [8]

Advantages

1 **genetically identical;**
possess the **desirable features of the stock plants;**

2 allows the **rapid** multiplication of plants;
giving rise to **bulk production** of genetically identical plants;

3 new plants are **disease-free**;
leading to higher **yield** / better **quality;**

4 **genetic modifications** possible (with help of protoplast cultures);
desirable traits can be introduced;

5 Take up **little space** when compared with plants growing in fields;
can be **grown intensively;**

6 Plantlets are light and small in **size;**
can be air-freighted and **transported** easily/cheaply/in large quantities, increasing international trade;

7 **Independent of climate** changes so plants can be produced continuously/at any time of year;
flexibility in meeting consumer demand;

8 possible to **standardise** the conditions for growth and obtain many batches of identical plants;
ensures product **uniformity;**

Disadvantages

1 lack of genetic diversity;
if a disease affects one plant, it will likely affect all the cloned plants / equal susceptibility to diseases ;

2 Aseptic conditions required as nutrients present in the culture media encourage the growth of microorganisms;
Possible contamination may result in huge losses;

3 Need trained personnel and costly equipment and facilities e.g. laminar flow hood to maintain aseptic conditions;;