H2 Biology • RI Prelim 2023

PAPER 1

1.	**B**	**7.**	**B**	**13.**	**D**	**19.**	**C**	**25.**	**D**
2.	**B**	**8.**	**A**	**14.**	**C**	**20.**	**A**	**26.**	**C**
3.	**B**	**9.**	**D**	**15.**	**D**	**21.**	**B**	**27.**	**A**
4.	**A**	**10.**	**A**	**16.**	**D**	**22.**	**A**	**28.**	**C**
5.	**C**	**11.**	**C**	**17.**	**D**	**23.**	**C**	**29.**	**C**
6.	**D**	**12.**	**D**	**18.**	**D**	**24.**	**C**	**30.**	**C**

PAPER 2

1(a) Name the structures **A**, **B** and **C**. [2]

A mitochondria;
B centrioles;
C lysosome/Golgi vesicle/ secretory vesicle; A: vesicle
2 marks if all correct, 1 mark if 2 correct

(b) Name one organelle that would also be present in a prokaryotic cell. [1]

Ribosome; R: 80S ribosome

(c) Describe the structure of **B** and its role. [4]

1. A pair of hollow cylinders made up of 9 triplets of microtubules (hollow tubes made of protein tubulin) each;
2. The two rod-like cylinders are positioned at right angles to each other;
3. Found in a region called centrosome which is microtubule organising centre (MTOC);

MAX 2m

4. During mitosis/ meiosis/ nuclear division, centrioles replicate and move to opposite poles of cells;
5. To determine polarity of the cell;
6. They help to organize microtubules to form spindle fibre;
7. To ensure proper separation of chromosomes during nuclear division;

MAX 2m

(d) Sodium ions cross cell surface membranes using facilitated diffusion or active transport.

Explain why sodium ions cross cell surface membranes by these mechanisms and not by simple diffusion. [3]

1. Sodium ions are charged and hydrophilic;
2. They cannot pass through the ***hydrophobic core**** of the phospholipid bilayer of membrane;
3. So must pass through transport proteins/carrier proteins with hydrophilic pore/channel embedded on cell surface membrane involved in facilitated diffusion or active transport.

[Total: 10]

2(a) Describe two differences in DNA behaviour during binary fission in a prokaryotic cell and mitosis in a eukaryotic cell. [2]

	Binary fission	Mitosis
1.	DNA replication of circular bacterial chromosome occurs during binary fission;	DNA replication of chromosomes occurs during the S phase of interphase prior to mitosis;
2.	Circular chromosome does not condense prior to separation;	All chromosomes in the nucleus will condense during ***prophase****;
3.	Chromosome does not attach to spindle fibres;	Chromosomes attach to spindle fibres via kinetochore proteins;
4.	Single chromosome does not line up across the equator of the cell;	Chromosomes line up in a single row at the metaphase plate;
5.	Chromosome does not separate into chromatids;	Chromosomes separate into sister chromatids during ***anaphase****;
6.	AVP	

(b) Suggest why the flasks were incubated at 35°C. [1]

This is the optimum temperature for the bacterial enzymes resulting in its fastest rate of growth/ division;

Fig. 2.2 is a graph which shows the results of the investigation.

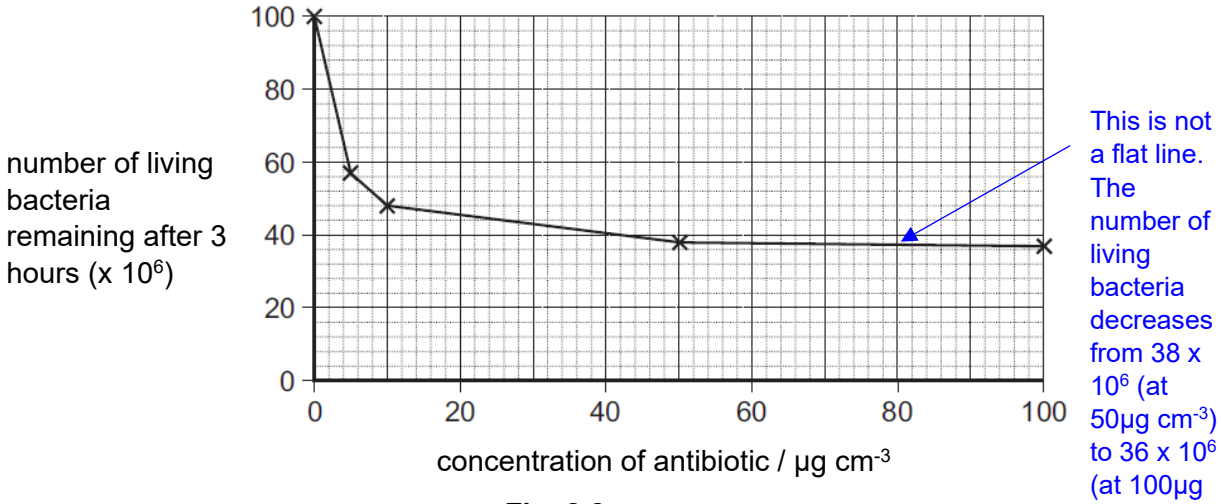


Fig. 2.2

(c) A student concluded from these results that an antibiotic dose equivalent to 50 μg cm^{-3} will effectively treat infections caused by *S. aureus*.

Evaluate his conclusion. [3]

Quoting of data (max 1 mark):

1. As concentration of antibiotic increase from 0 to 50 μg cm^{-3}, number of living bacteria decreases from 100 to 38 x 10^6; (other quoting of relevant data, where correct, will also be given credit)

Agree:

2. At 50 μgcm^{-3}, there was only 38 x 10^6 bacteria remaining and reduction of most of bacteria (62%) meant that 50 μgcm^{-3} was an effective dose;
   or
   When antibiotic concentration increased from 50 μgcm^{-3} to 100 μgcm^{-3}, there was only a very marginal decrease in bacteria (from 38 x 10^6 to 36 x 10^6) which was not significant. Hence 50 μgcm^{-3} was considered an effective dose without negative effects of higher antibiotic concentrations (beyond 50 μgcm^{-3});

Disagree:

3. When antibiotic concentration increased from 50 μgcm^{-3} to 100 μgcm^{-3}, there was a decrease in bacteria (from 38 x 10^6 to 36 x 10^6), and this means that 50 μgcm^{-3} was not as effective as 100 μgcm^{-3};
4. While the results are valid based on the experimental data, the effectiveness of the antibiotic was not studied in humans;
5. Number of living bacteria may be lower at higher antibiotic concentrations if incubated for longer than 3 hours;
6. Other concentrations can also be considered to treat the infection effectively as it causes a decrease in bacterial growth;
7. At 50 μgcm^{-3}, there is still 38 x 10^6 bacteria living, hence a large enough figure to cause disease, so ineffective;
   OR
   At 50 μgcm^{-3}, there is still 38 x 10^6 bacteria living, not all bacteria removed, hence ineffective;

(d) The antibiotic used in the experiment was penicillin. Penicillin acts as an inhibitor to an enzyme found in *S. aureus*.

(i) Name the enzyme inhibited by penicillin. [1]

Transpeptidase

(ii) Describe how penicillin inhibits this enzyme. [2]

1. Penicillin acts as a ***competitive inhibitor**** to transpeptidase;
2. It has a similar shape to substrate/complementary in shape to active site of the enzyme and will bind to ***active site**** of the enzyme;
3. Since penicillin binds and blocks ***active site****, the substrate is unable to bind to active site;

(iii) Explain how penicillin reduces bacterial growth as a result of this inhibition. [3]

1. Penicillin inhibits bacterial cell wall synthesis/ disrupts peptidoglycan synthesis;
2. By competitively binding to and inhibiting transpeptidase, it will inhibit formation of cross-links between adjacent chains/ peptidoglycan chains;
3. As a result, weakening the bacterial cell wall of dividing bacterial cells;
4. Because of high osmotic pressure inside the bacterium / when bacteria take in water by osmosis;
5. increased turgor pressure against weakened cell wall causes bacterium to swell and lyse;

[Total: 12]

3(a) **(i)** What are the unique features of stem cells that made them suitable to treat this disease? [2]

1. A stem cell is an ***undifferentiated / unspecialized**** cell capable of undergoing ***proliferation* and self-renewal****;
2. Ability to ***differentiate**** into specialized cells;
3. This is a result of differential switching on of genes which occurs when appropriate ***molecular signals****;

(ii) Explain why the mice in group SC-X needed to have their immune systems suppressed. [1]

1. To avoid stimulation of their immune system / prevent rejection of introduced cells;
2. because donor stem cells are recognised as foreign;

(b) **(i)** With reference to Tables 3.1 and 3.2, compare the effects of the different treatments given to the mice in groups SC-X and SC-S. [2]

1. Levels of expression of normal allele for *IDUA* is higher in group SC-X compared to group SC-S;
2. There is improvement in health of 2 mice in group SC-X but none in group SC-S;
3. indicating that high levels of expression (+++ at least) of normal allele of *IDUA* was required for improvement to health;
   A: reference to low expression (++ and below did not lead to improvement to health;
   R: merely copying table's data as +++ without interpreting this as "high"
4. Both showed gene expression;

(ii) Suggest why the expression of the normal allele of *IDUA* is different in the stem cells given to the mice in groups SC-X and SC-S. [2]

1. Stem cells from an unaffected donor in group SC-X likely to have 2 copies of normal functional allele *IDUA* in its genome, while transduced stem cells in group SC-S likely to have at most 1 copy of normal functional allele *IDUA* integrated into its genome resulting in a lower level of expression;
2. Normal functional allele *IDUA* in group SC-S may be linked to, and is regulated by a ***weaker promoter**** as compared to that in group SC-X;
3. Normal functional allele in transduced stem cells in SC-S may not be stably ***integrated**** into genome and is gradually lost with each subsequent cell division while stable integration is not an issue with stem cells from group SC-X;
4. Normal functional allele *IDUA* for stem cells in group SC-X was under influence of an ***enhancer**** . This ensures high level of expression of normal functional allele *IDUA* as compared to group SC-S where normal functional allele was integrated at a position not under influence of an enhancer;
5. Normal functional allele for stem cells in group SC-S was integrated at a position that put it under influence of a ***silencer**** . This result in a lower level of expression of normal functional allele *IDUA* as compared to group SC-X;

AVP: point of comparison with elaboration.

(c) Comment on the ethical aspects of using stem cells from a healthy person for the treatment of patients with Hurler syndrome. [2]

1. Adult stem cells are ***multipotent**** and cannot form a whole organism, hence no debate of whether or not a life is "sacrificed", as opposed to using embryonic or zygotic stem cells ;
2. Potential to cure patients completely so patients no longer have to be on lifetime medication;
3. Possibility of unforeseen consequences in treated patients/ recipients e.g. unexpected behaviour of cells, unknown long-term health effects, immunological reactions leading to patients on immunosuppressants for life;
   R: examples where outcome was certain
4. Issues of informed consent, understanding of aims and privacy for stem cell donors;

AVP: e.g.. Accessibility to treatment - where high cost would make the treatment inaccessible to people with limited income;

[Total: 9]

4 Studies have shown that genes such as cyclin dependent kinase inhibitor 3 (*CDKN3*) are highly overexpressed in cervical tumours.

Fig. 4.1 shows the gene expression of *CDKN3* in healthy and cancerous cervical tissue.

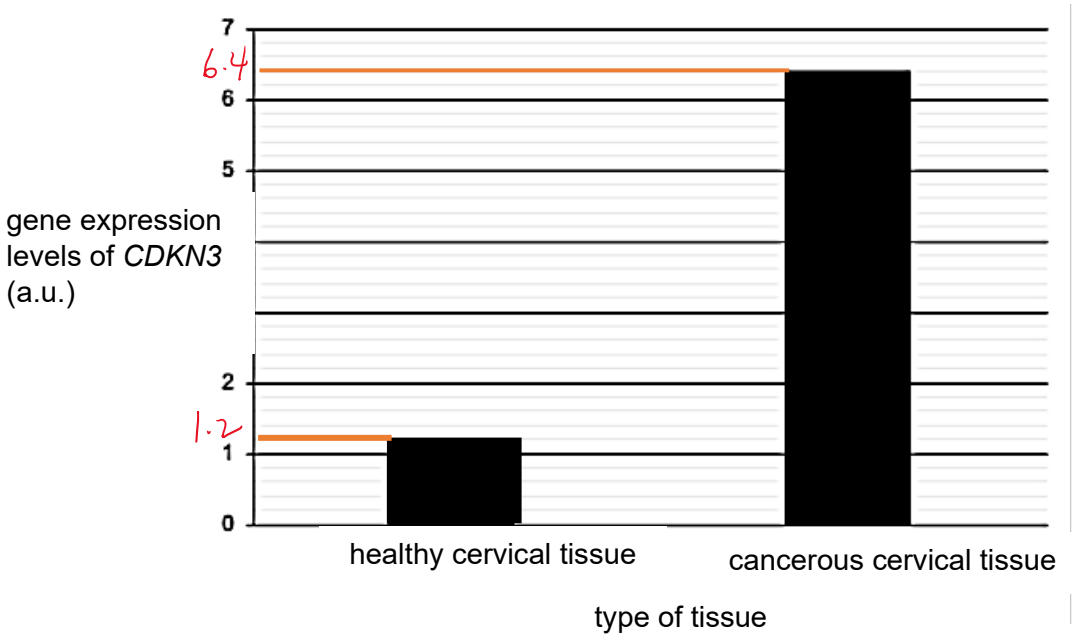


Fig. 4.1

(a) Calculate the percentage increase of *CDKN3* gene expression levels in cancerous cervical tissue compared to healthy cervical tissue.

Show your working clearly below. [1]

Percentage increase =
((Gene expression in cervical cancer tissue - gene expression in healthy cancer tissue) ÷ gene expression in healthy cancer tissue) x 100%

= ((6.4 - 1.2) ÷ 1.2) x 100%;
= 433%;
A: 2 dp or 2sf, 3 sf

(b) Overexpression of *CDKN3* in cervical tumours has been associated with decreased survival rates in cervical cancer patients and has been shown to trigger cells to exit mitosis and begin cytokinesis.

The data in Fig. 4.2 shows the survival rate of patients diagnosed with cervical cancer and their expression of *CDKN3* with time from the point of diagnosis.

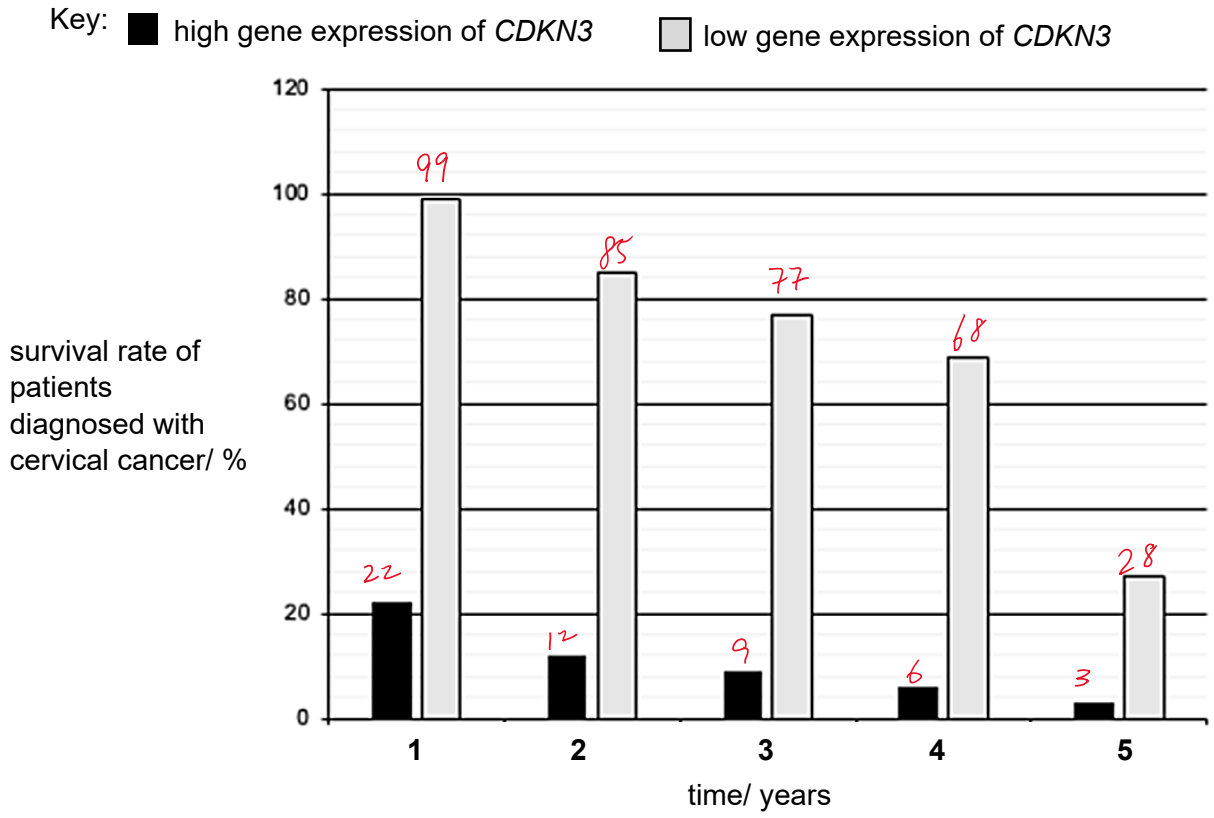


Fig. 4.2

(i) A hypothesis has been proposed that overexpression of *CDKN3* in cervical tumours results in an increased chance of patient death.

Evaluate the evidence for this using the data given. [2]

1. The data supports the hypothesis that the overexpression of CDKN3 results in decreased survival rate of patient/ increased patient death across all time points;
2. Quote values (high vs low gene expression at any one year)
3. The data is insufficient to support the hypothesis that the overexpression of CDKN3 results in decreased survival rate of patient/ increased patient death across all time points;
4. e.g. correlation does not mean causation; no statistical analysis carried out

(1&2 or 3&4)

(ii) *CDKN3* gene has been identified as an oncogene. Explain what an oncogene is. [2]

1. Oncogene is a ***proto-oncogene**** that has undergone a ***gain-in-function mutation****;

2. leading to the overexpression of gene products / production of hyperactive or degradation-resistant protein causing ***uncontrolled cell division****and cancer;

(c) **(i)** Explain the need for the tight control of the mitotic cycle. [3]

1. Cell cycle is regulated for normal growth and development;
2. Cell cycle is regulated at ***checkpoints**** at G1, G2 and M, which determines if the cell cycle can proceed;
3. Dysregulation of checkpoints cause cell to escape cell cycle control mechanism;
4. Leading to ***uncontrolled division**** of cells and cancer;

AVP

- G1 checkpoint can proceed if DNA is not damaged;
- G2 checkpoint can proceed if DNA is not damaged/chromosome has replicated/sufficient cyclin can bind with Cdks to form MPF;
- M Check point can proceed if all chromosome are attached to spindle fibres from both poles;

(ii) Studies have shown that CDKN3 proteins inhibits CDK which is associated with DNA replication in the cell cycle.

With reference to Fig. 4.3, state which protein is inhibited by CDKN3. [1]

CDK2;

(d) Based on the evidence provided, suggest, with a reason, which of the labelled cells would be present in the higher numbers in c-REL -/- cervical cancer cells as compared to c-REL cervical cancer cells. [2]

1. A;
2. Since c-REL -/- cervical cancer cells spend a longer time at prophase, there will be more cells at prophase, A;

[Total:11]

5(a) Match each mutant to the possible types of mutations:

operator deletion ; Mutant 1

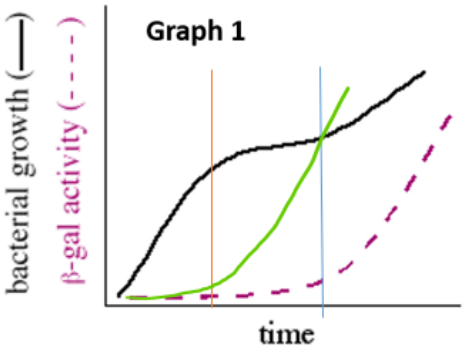
promoter deletion : Mutant 2 [2]

(b) Explain your answer for the mutant with the operator deletion in the presence of lactose only. [2]

1. No operator, hence, no binding of repressor, hence RNA pol not blocked from to promoter / RNA pol accessible to promoter.
2. Absence of glucose, hence high levels of cCAMP, which can activate CAP, increase affinity of RNA pol to promoter
3. Hence resulting in high level / 100 units of β-galactosidase, as in the WT.

(i) Sketch on graph 1, how the levels of cAMP is expected to change with time. [1]

Similar shape to β-galactosidase activity graph but shifted to the left.



(ii) State and explain if the results shown in graph 2 is expected in the presence of IPTG. [3]

1. The results are not expected;
2. In the presence of the inducer IPTG (inducer), the lac repressor is inactivated
   Or
   RNA polymerase will bind to the promoter and the lac operon is immediately turned on;
3. However, in graph 2, in the presence of glucose, cAMP levels should be low and thus the ß-galactosidase activity should not immediately increase/ the graph should be similar to graph 1;

(iii) Suggest why operons are necessary in bacteria. [2]

1. Genes coding for proteins with related function under control of single promoter, hence so as to respond rapidly and appropriately to the environment
2. Bacterium only produces enzymes when required / in respond to certain changes/conditions so allowing the bacteria to make economical use of / prevent wastage of energy and resources
3. Especially since bacteria are able to use a variety of metabolites e.g. glucose is metabolised preferentially over lactose, thus not economical to produce *lac* genes in the presence of glucose; so allowing the bacteria to make economical use of / prevent wastage of energy and resources;
4. The abovementioned provide a selective advantage to such bacteria;

[Total: 10]

6(a) (i) In the space below, draw a genetic diagram to explain the results of crossing the F_1 offspring to produce the phenotypic ratio of squash fruit colours shown in the F_2 generation. Use the symbols **A, a** and **B, b** to represent the alleles. [4]

Let A represent the dominant allele for production of inhibitor that prevents colourless precursor to be converted to yellow intermediate.
Let a represent the absence of inhibitor.

Let B represent the dominant allele for production of inhibitor that prevents yellow intermediate to be converted to green pigment.
Let b represent the absence of inhibitor.

F_1 phenotype	White fruit	X	White fruit
F_1 genotype	AaBb		AaBb
Gametes	**AB** **Ab** **aB** **ab**		**AB** **Ab** **aB** **ab**

Fertilisation

Gametes	**AB**	**Ab**	**aB**	**ab**
AB	AABB white fruit	AABb white fruit	AaBB white fruit	AaBb white fruit
Ab	AABb white fruit	AAbb white fruit	AaBb white fruit	Aabb white fruit
aB	AaBB white fruit	AaBb white fruit	aaBB yellow fruit	aaBb yellow fruit
ab	AaBb white fruit	Aabb white fruit	aaBb yellow fruit	aabb green fruit

F_2 genotypes	9 A_B_ :	3 A_bb :	3 aaB_ :	1 aabb
F_2 phenotypic ratio	12 white fruit	:	3 yellow fruit :	1 green fruit

1. F_1 genotypes and corresponding F_1 phenotypes
2. F_1 gametes
3. F_2 genotypes and corresponding F_2 phenotypes
4. F_2 phenotypic ratio
5. Showing how the ratio combines to give the epistatic ratio 12:3:1

(max 4)

(ii) Test crosses were carried out on two white-fruited plants, **P** and **Q**, from the F_2 generation. Each of these plants had its female flowers pollinated with pollen from a green-fruit plant.

For plant **P**, half of the offspring were white and half were yellow.
For plant **Q**, half of the offspring were white and half were green.

Deduce the genotypes of plants **P** and **Q.**

plant **P** ……..

plant **Q** …….. [2]

1. plant **P AaBB;** (A: AABb depends on genetic diagram if A_bb is yellow fruit)
2. plant **Q Aabb;** (A: aaBb depends on genetic diagram if A_bb is yellow fruit)

(b) **(i)** Calculate the percentage of seeds which germinated in the parental generation.

Show your working. [2]
56/70 x 100 = 80%

1. correct calculation and working shown
2. correct answer

(ii) Name the type of variation shown in Fig. 6.1(c).

Explain your answer. [3]

1. ***Continuous variation****; (Must have)
2. The offspring show a range of phenotypes for height (25-46 cm); (quote values)
3. The range of phenotypes indicate ***polygenic**** inheritance where multiple genes are involved;
4. There is an additive effect of each gene where each gene has a small overall effect;
5. In addition to genotypic factors, ***environmental**** factors affect the phenotype;

[Total: 11]

7 ATP is the universal energy currency which provides the immediate source of energy for cellular
processes.

Fig. 7.1 shows some ways in which ATP may be synthesised and used in cells.

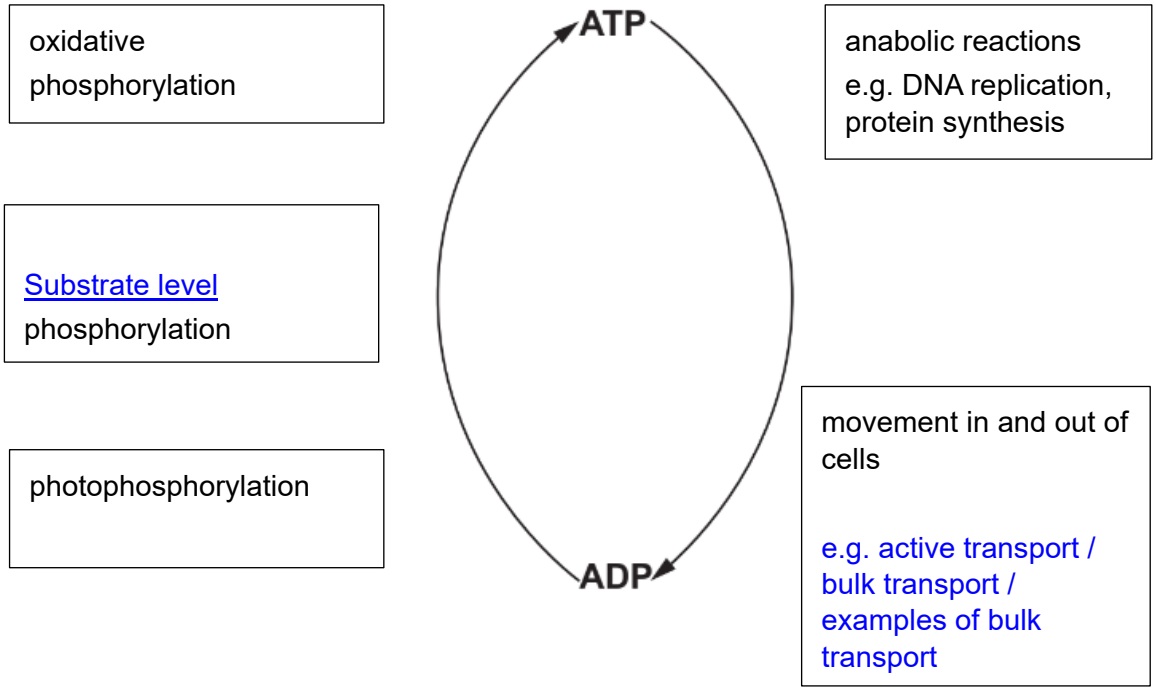


Fig. 7.1

(a) Complete Fig. 7.1 by writing correct terms or examples on the dotted lines provided. [2]

(b) **(i)** With reference to Fig. 7.2, state one structural similarity between coenzyme A and ATP. [1]

Both contain

1. nitrogenous base/ adenine;
2. pentose/ ribose; R: 5 carbon ring I: deoxyribose
3. contains three phosphate groups;

(ii) Suggest why the actual net number of ATP molecules synthesised is less than the theoretical number. [1]

1. some energy lost as heat;
2. glucose may not be completely broken down / some intermediates are used in different metabolic processes;
3. reduced NAD may be used for other (metabolic) reactions;

(c) As respiration takes place, oxygen is used by the seeds and the coloured liquid moves down the tube.

Describe the role of oxygen in aerobic respiration. [3]

1. Oxygen is ***final electron acceptor**** at the end of ***electron transport chain**** (ETC);
2. where it will combine with electrons and protons to form water (accept: $2e^- + 2H^+ + \frac{1}{2}O_2 \rightarrow H_2O$)
3. By removing electrons, oxygen re-oxidises ETC so that ***NADH**** and ***$FADH_2$**** can continue to donate electrons, thereby allowing ***oxidative phosphorylation**** to continue to produce ***ATP****;
4. This allows ***regeneration**** of ***NAD**** and ***FAD**** allowing them to pick up more electrons (and protons) from ***glycolysis****, ***link reaction**** and ***Krebs cycle****;

(d) **(i)** Calculate the rate of oxygen uptake in cm^3 per minute for respirometer **C** between 5 and
20 minutes.

Give your answer to two significant figures. Show your working. [1]

$\frac{1.7 - 0.4}{20 - 5} = \frac{1.3}{15} = 0.08666 = 0.087$ (2 s.f) ;

(ii) Explain why there is an increased rate of respiration of germinating pea seeds between
10°C and 25°C. [2]

1. Thus with increasing temperature , ***kinetic energy**** of the substrates and the enzymes involved in respiration/ germination increases resulting in the increase in frequency of ***effective collisions**** between the enzyme ***active sites**** and substrate;
2. which results in the formation of more ***enzyme-substrate (ES) complexes**** per unit time;

which will increase the rate of respiration.

[Total: 10]

8(a) **(i)** Using the information in the table above and your own knowledge, suggest why the cheetah is the cat at highest risk if the environment changes. [3]

1. Cheetahs have the lowest percentage of genes with different alleles 4% compared to the other cats, 23%, 12% and 21% in the domestic cat, lion and ocelot respectively;
2. Cheetahs have the lowest genetic diversity / least genetic variation of the listed cats ;
3. Hence when the environment changes, the cheetahs are least likely to have the advantageous allele that will confer a selective advantage / be selected for and are thus least likely to survive / most at risk of extinction;

(ii) Cheetahs are unusual amongst the big cats. A female cheetah often mates with several different males and gives birth to two or three cubs at a time, each having a different father.

Suggest why this may be advantageous to cheetahs. [2]

By mating with different males,

1. there will be greater genetic variation/diversity amongst the litter in the fastest possible time;
2. an increased chance of fertilization/pregnancy;
3. an increased chance that the cubs will survive and reproduce when the environment is unfavourable;
   and hence the population size will increase

(b) **(i)** The scientific name for the Neanderthal is considered to be either *Homo sapiens* or *Homo neanderthalensis*.

A scientist considers that *Homo sapiens* is the correct name to use.

State the assumption that the scientist is making and explain why it is difficult to confirm this assumption. [3]

assumption:[1](max 1)

1. Neanderthals and modern humans are the same species;
2. Neanderthals and modern humans could interbreed and produce fertile, viable offspring;

explanation /Why it is difficult to confirm this assumption: [2] (max 2**)**

3. Since only Neanderthal fossils are available/Neanderthals are extinct;
4. it is not possible to confirm if they were able to interbreed with modern humans and produce fertile, viable offspring; (linked to point 3)
5. it is not possible to obtain sufficient, morphological / physiological / behavioural /ecological /molecular data ; (linked to point 3)
6. small sample of Neanderthals fossils;
7. AVP;

(ii) Suggest what the data in Fig. 8.1 indicate about the evolutionary relationships between modern humans, the chimpanzees and the macaques. [3]

1. All 3 species share a common ancestor ;
2. the smaller the number of differences the, more closely related the different species are;
3. chimpanzees are most closely related to modern humans as there is only a 2 nucleotide difference between them and they share a most recent common ancestor;
4. macaques, are as distantly related to humans and as they are to chimpanzees as there is 10 nucleotide difference between macaques and humans and macaques and chimpanzees;

(iii) Explain why any such conclusions made in b(ii) need to be treated with caution. [2]

The data must be treated with caution because

1. the nucleotide sequences in Fig. 8.1 are only a small fraction of the whole genome (A: only studying part of one gene) and hence not representative of the entire genome;
2. there will be variation within each species due to mutations;
3. these sequences come from one individual from each species ;
   AVP: insufficient morphological/ physiological/anatomical data made available to make a conclusion;
   R: convergent evolution

[Total: 13]

9(a) Describe and explain the relationship between mean water temperature and the trend observed for symbiont cell densities during the warming period. [3]

1. Increase in mean temperature from 23.5°C to 28.0°C during the warming period saw a corresponding increase in symbiont cell density from 0.06a.u.to 0.15a.u for colonies with clade D/ 0.04a.u. to 0.07a.u. for colonies with clade C;
2. Quote values correctly;
3. Increase in temperature leads to higher metabolism for the algal cells due to greater enzyme activity/rate of photosynthesis/respiration, hence greater growth rates and higher cell density;

(b) Compare the symbiont cell density between colonies with group C and colonies with group D *Symbiodinium* during the bleaching event. [2]

Similarities

1. Both show a decreasing trend;
2. Decrease was more gradual from 13 Aug to 18 Aug/ decrease was more steep from 14 Jun to 13 Aug;

Differences

3. Cell density for group D was much higher at all sampling dates;
4. Cell density decreased to 0.00a.u. for colonies with group C/fully bleached but colonies with group D did not fully bleach (Reject: just stating cell densities without emphasizing that colonies with group D did not fully bleach)/ decrease in symbiont cell densities in colonies with group C was greater than in colonies with group D.
5. Standard deviation was greater for colonies with group D

(c) Suggest an explanation for the difference in symbiont cell density between colonies with group C and colonies with group D *Symbiodinium* during the bleaching event. [1]

1. (Coral colonies with) group D *Symbiodinium* are more tolerant to higher temperatures;
2. Presence of group D *Symbiodinium* conferred some protection to the corals against higher temperatures (i.e. the corals didn't bleach even with >80% of other corals bleached)
3. AVP

(d) Suggest why although there was no change in temperature between 13 and 18 August, there was a decrease in symbiont cell density. [1]

1. Prolonged exposure to elevated temperatures caused corals to undergo prolonged period of stress resulting in bleaching;
2. 31°C is already beyond the thermal tolerance;
3. There were additional stress factors due to climate change like increased bacterial infection, lower pH due to ocean acidification

(e) Suggest the impact of the loss of coral biodiversity. [1]

1. Loss of potential biomedicines sourced from coral reefs;
2. Loss of genetic diversity of foods such as fish that use coral reefs as nursery for their younbg;
3. Loss of habitat for many organisms that depend on coral reefs for shelter and food (A: symbiotic relationships)
4. AVP with sufficient support e.g. economic impacts

[Total: 8]

10 (a) (i) On Fig. 10.1, complete the diagram to illustrate the process of antigen presentation on the dendritic cell.

You are required to label the structures that you have drawn. [1]

Drawing showing peptide-MHC complex on dendritic cells

- There must be a part embedded in at least 1 layer of the bilayer
- 2 separate parts representing 1) the MHC and 2) the antigen / pepitide
- Antigen must be complementary in shape to the T cell receptor

(ii) Describe the processes that occurs after the event illustrated in a(i). [2]

1. Secretion of ***cytokines**** by dendritic cell;
2. Naïve T cell becomes activated to undergo ***clonal expansion and differentiation**;*
   into effector cells, such as ***cytotoxic T/helper T**** and ***memory T cells**** (one named example of cell);

(b) Compare the structure of the TCR shown in Fig. 10.1 to the structure of an antibody molecule secreted by plasma cells. [3]

Similarities

1. Both have disulfide bonds;
2. Both have antigen binding site(s);
3. Both have quaternary structure / more than 1 polypeptide chain;

Differences

	Feature	Fig. 1.1	Antibody
5	Number of polypeptides	2 polypeptides	4 polypeptides
6	Hydrophobic R-groups of amino acids on the surface	Present (embedded in the membrane)	Absent
7	Number of antigen binding sites	1	2

Must have at least one similarity and one difference.
Note: There is no point 4

[Total: 6]

PAPER 3

1(a) **(i)** Identify the cell structures that fluoresced in Fig. 1.2 and name the specific antigen (protein X) that the primary antibodies bind to. [2]

cell structure	spindle fibres / microtubules [1]
specific antigen (protein X)	tubulin [1]

(ii) Identify the stage of mitotic division that the cell in Fig. 1.2 is in and describe the events that occur at this stage. [3]

1. ***Anaphase**** **;**
2. ***centromere**(s)* **divides; (**R;replicate/split);
3. and ***sister chromatids**,* now called daughter ***chromosomes**** are pulled/separate to **opposite *poles**** **(**Reject: ends) of the cell;
4. By shortening ***kinetochore microtubules**** **(**Reject: contract) with centromeres leading resulting in characteristic v shape of the chromosomes;

OR

1. ***Metaphase****:
2. Kinetochore microtubules/spindle fibres attached to the centromeres;
3. will align the chromosomes at the ***metaphase plate/equator*;**

(iii) The secondary antibodies are goat antibodies that bind to the primary antibodies which are mouse antibodies.
With reference to Fig. 1.1, state the name of the specific site that the secondary goat antibodies bind to and explain why the secondary goat antibodies do not bind to other sites. [3]

1. The secondary goat antibodies bind to the constant domain of heavy chain / fragment crystallisable (Fc) of primary mouse antibodies;
2. This is because the antigen binding site / fragment antigen binding (Fab) region of secondary goat antibodies;
3. are not ***complementary in shape**** to other sites, and thus cannot bind to them;
   OR
   are only complementary to Fc region of primary mouse antibodies;

(b) **(i)** Explain why plasma cells alone cannot be used directly for the long term production of antibodies. [3]

1. Plasma cells are ***differentiated**** cells;
2. Unable to undergo mitosis/self-renewal/divide indefinitely to produce a constant pool of plasma cells;
3. short life-span/dies after a few days;

(ii) The Hayflick limit is the number of times a normal somatic cell population will divide before cell division stops. In order for the 'immortal' hybrid cells in Fig. 1.3 to divide indefinitely, the cells need to overcome the Hayflick limit.
Explain how the fusion of the plasma cells with the tumour cells enable the 'immortal' hybrid cells to overcome the Hayflick limit. [4]

1. Named gene: ***telomerase****
2. Expression of telomerase is switched off in normal/plasma cells;
   (A: telomerase active in tumour cells)
3. but switched on in immortal hybrid cells;
4. Telomerase anneals to the ***3' overhang**** at the end of the telomeres and lengthens the telomeres;
5. Hence critical length is not reached thus allowing the cells to divide continuously and overcome the Hayflick limit;

(c) **(i)** Using the information above, help Dr N to design a 10 nucleotide long 'forward' and 10 nucleotide long 'reverse' primer that can be used to amplify the coding region of the protein X gene.
State the sequences of the respective primers in the table below. [2]

type of primer	sequences of primer
'forward'	5' CATGTCAATT3' (must include polarity)
'reverse'	5'AACTTTACAG3' 3'GACATTTCAA5' (must include polarity)

(ii) Comment if Dr N's proposed PCR conditions will successfully produce the desired PCR product. [4]

1. No PCR product will be formed in the first cycle;
2. hydrogen bonds between complementary base pairs/DNA strand are not broken, hence template/target DNA remain double stranded/are not single stranded;
3. Primers are unable to anneal to the target DNA sequences at 65°C/annealing stage;
4. No primers to provide the 3'OH group, Taq DNA polymerase unable to add nucleotides/extend DNA at the 72°C elongation stage;

And/or

5. PCR product will be formed after cycle 1;
6. Ref to idea that after first stage 3 of cycle 1, stage 1, 2 and 3 were repeated sequentially for 29 cycles;
7. Ref to idea that denaturation, annealing and elongation stages can proceed;
8. Less PCR product formed than if conditions was correctly set as there are only 29 full cycles;

(d) **(i)** Name the non-coding sequence that must be present in the bacteria plasmid to enable the expression of protein X. [1]

Bacterial ***promoter****;

(ii) Further analysis shows that the polypeptide produced is much longer than expected.

Suggest an explanation for this observation. [3]

1. Protein X/tubulin is an ***eukaryotic**** gene that contains non-coding ***introns****;
2. ***RNA splicing**** does not occur in bacteria as the spliceosomes are absent;
3. as the introns are not excised and the exons are not joined;
4. Hence mRNA is longer and is translated into a longer polypeptide; (point accepted only if context is correct)

(iii) Explain why this polypeptide can still be injected into the mouse to produce the correct primary antibodies as shown in Fig. 1.3. [3]

1. Some sections of the polypeptide will have the same sequence of amino acids as protein X;
2. 3D conformation of some epitopes/ antigens/ regions of the protein remains unchanged;
3. which can still stimulate the production of the correct primary antibodies that may bind to these sections;
4. As the antibodies will have a ***Fab/ antigen binding site**** that is complementary to conformation to the antigen;

[Total: 28]

2(a) Explain why both the SARS-CoV-2 and the influenza viruses recognize the surface epithelial cells of the respiratory tract. [2]

1. Both the ***ACE 2**** receptor and ***sialic acid receptor**** are cell surface receptors located on the cell surface of epithelial cells of the respiratory tract allowing SARS-CoV-2 and influenza viruses to bind respectively;
2. The glycoproteins on the cell surface receptors of SARS-CoV-2 and influenza viruses are complementary in shape to the respective receptors;

(b) Describe two differences between the replication cycle of SARS-CoV-2 and influenza viruses. [2]

	SARS-CoV-2	influenza viruses
+ or – strand RNA used to synthesise proteins	1. + ss RNA used directly to synthesize viral proteins;	-ssRNA genome is used to synthesise a +strand RNA before it can be used to synthesise viral proteins ;
polyprotein synthesis and proteolysis	2. proteolysis of polyproteins (pp1ab and pp1a);	proteins not synthesized as polyproteins;
entry of virus	3. either by direct ***fusion**** of the host cell and viral membranes **and/or** ***endocytosis**** using the S2 subunit of the spike protein;	**Only by** into the host cells by ***endocytosis****;
assembly	4. occurs within vesicle;	at plasma membrane;
release	5. by ***exocytosis****;	by ***budding****;

(c) Describe a strategy that can target a named protein which can be used to block key steps of the life cycle of SARS-CoV-2. [1]

1. Prevent attachment/binding of the virus to the host cell by blocking the binding of the ***spike protein**** to ACE2;
2. Prevent replication of RNA genome by inhibiting ***RNA dependent RNA polymerase****;
3. Prevent proteolysis of polyprotein, pp1ab, pp1a, by inhibiting viral protease;

(i) Describe the differences in the proportion of cases against age before and after vaccination was introduced. [2]

1. Before vaccine era : nearly all cases occurred in early life, after which protection was continually reinforced;
2. Vaccine Era: Lower proportion of cases in early age but peaked past middle age ;

(ii) Explain how vaccination causes a rise in antibody titre to a level similar to that produced during an infection.[4]

1. Vaccine stimulates an immune response because specific surface antigens of pathogen are retained;
2. ***Antigen presenting cells**** (APCs) (A: ***macrophages** / **dendritic cells***) take up the vaccine by phagocytosis, process the antigen and present it as a ***peptide:MHC complex***;
3. Each naïve T cell has a specific T cell receptor (TCR) that specifically binds to complementary peptide:MHC complex on APC;
4. APC secretes ***cytokines**** that activates naïve T cell which will undergo ***clonal expansion and differentiation**** to form ***helper T cells***, cytotoxic T cells and memory T cells;
5. ***Helper T cells**** secrete ***cytokines**** that activate specific naïve ***B cells**** to undergo ***clonal expansion and differentiation*;***
6. and form antibody-secreting ***plasma cells**** and that produce antibodies at levels similar to that produced during an infection;

(iii) Suggest how the vaccination programme can be modified to prevent the re-emergence. of VPD. [1]

Administer another booster jab in after middle age/ when people are older.

[Total: 12]

3(a) Describe how the structure of adenylyl cyclase relates to its function.[2]

1. It contains a ***G-protein binding site**** so that an activated G protein can bind to it and activate it;
2. It also contains an ***active site**** that is ***complementary in shape**** and charge to ***ATP**** to catalyse the conversion of ***ATP**** to ***cAMP****;
3. It contains amino acids with ***hydrophobic R groups**** on its exterior that allows it to be embedded/ stabilised in the ***hydrophobic core**** of the phospholipid bilayer;

(b) With reference to Fig. 3.1, identify a stage where signal amplification is observed and explain how the signal is amplified in this stage. [2]

1. Conversion of ATP to ***cAMP**** by ***adenylyl cyclase****;
2. Each adenylyl cyclase / enzyme produces many molecules of cAMP;

OR

3. PKA will phosphorylate the next protein in the pathway;
4. One PKA molecule can activate many proteins in the next step;

OR

5. Activation of G protein by ligand/adenosine binding to receptor;
6. One ligand/adenosine binding to the GPCR can activate many G proteins;

(c) With reference to Fig. 3.1 and Fig. 3.2, explain the effect of consuming several caffeinated drinks on an individual.[4]

1. The structure of caffeine is similar to (part of) the structure of adenosine;
2. Hence both molecules are ***complementary in shape**** and charge to the ***ligand binding site**** of the G-protein coupled/ adenosine receptor;
3. As there is a high level of caffeine in the body from consuming several caffeinated drinks;
   caffeine is more likely to bind to the receptor and prevent adenosine from binding to it /caffeine competes with adenosine for binding to receptor;
4. Hence the adenosine signalling pathway is not initiated, no activation of G protein, no activation of adenylyl cyclase, no production of cAMP, PKA is not activated (A: any 2)
5. drowsiness does not occur;

(d) Drugs known as cAMP phosphodiesterase inhibitors are used in the management and treatment of chronic obstructive pulmonary disease, psoriasis, psoriatic arthritis and erectile dysfunction.

With reference to Fig. 3.1, discuss the impact of treatment with cAMP phosphodiesterase inhibitors on an individual who

- suffers from psoriasis;
- is highly stressed; and

does not consume anything with caffeine.[2]

1. cAMP phosphodiesterase inhibitors will prevent the conversion of cAMP to AMP as;
2. Hence cAMP levels will increase further which in turn will activate PKA;
3. and cause the individual will feel drowsy while also treating the symptoms of psoriasis;

[Total:10]

4(a) Discuss whether you agree or disagree that the impacts of human activity on climate change will lead to the greater spread of mosquitoes.

Comment if this matter requires both urgent and global action. [15]

(A) Impacts of human activity on climate change:

1. Human activities contribute to increased production of greenhouse gases + any 2 activities:
   a. human induced burning of fossil fuels releasing CO_2;
   b. increase in decomposing materials, e.g in landfills/sewage increase CO_2 release;
   c. industrial processes, such as in cement works, release CO_2;
   d. burning land and vegetation to clear land for agriculture;
   e. food choices that have greater carbon footprint for production;

   (Students must give 2 examples of human activities to score one mark for A1)

2. these activities have led to increase in greenhouse gas emissions since 1750, the start of the industrial revolution;

3. Greenhouse gases (GHG) such as CO_2 and methane/nitrous oxide allow short-wave radiation from the sun to pass through to heat the Earth's surface and lower atmosphere;
4. The heated surfaces radiate out-going long-wave radiation/infra red/heat which is absorbed and re-emitted by the GHGs back to the Earth's surface;
5. Increased concentration of GHGs leads to increased trapping of radiation/heat causing more warming;

(B) Agree (the above will lead to the spread of aedes/anopheles mosquito)

1. increase in global temperatures at higher latitudes and/or altitudes;
2. become more optimal for insects including aedes mosquitoes ;
3. higher temperatures, duration from first instar to emergence is shorter as rate of enzyme-catalysed reactions increases;
4. this leads to faster development rate and decreased length of reproductive cycles;
5. climate change could result in more rainfall in certain areas creating more pools of stagnant water for mosquito breeding (ORA less rainfall/drought in some areas so less standing water for mosquitoes to breed);

(C) Disagree (climate change is not the only factor/reduced populations in the tropics)

1. Changes in land use and human population growth and migration may also lead to greater spread of the aedes mosquito;
2. Poor mosquito control measures can also lead to the spread (e.g. less use of insecticides, lack of education/awareness, enforcement of measures);
3. Population increase / more dense and urbanisation (together poor control measures) can lead to more breeding grounds;
4. Increase in global temperatures may cause tropics to be uninhabitable due to small thermal safety margins of most insects (range shift rather than spread/expansion);

(D) Require urgent and global action (MAX?)

1. *Aedes aegypti* /*Anopheles* female mosquito is a vector for human disease like dengue fever and malaria
2. Greater disease transmission;
3. Increased temperature also results in shorter extrinsic incubation period as virus/plasmodium parasite can reproduce faster within mosquito vector;
4. Dengue fever caused by spread of dengue virus through the bite of infected female *Aedes aegypti* mosquito;
5. Highest risk in highly populated regions (human host) with rainy seasons (breeding ground for mosquito vector);
6. Dengue virus infects cells of the immune system and can be potentially fatal with secondary dengue infection;
7. Malaria is caused by spread of *Plasmodium* parasite through the bite of infected female *Anopheles* mosquito;
8. [global action] Both diseases, while mainly prevalent in tropical and sub-tropical, can affect human populations globally due to migration and human travel;
9. [global action] Climate change cannot be mitigated through the action of certain countries/regions only, (A: Climate change requires a global effort involving all countries);

QWC: 1 point from B/C + D

(b) Explain why, in a mammalian cell, glucose is a better respiratory substrate compared to triglycerides and why triglycerides are suitable as storage molecules. [10]

A. Why glucose is a better respiratory substrate:

Comparison needed for points to be awarded

Feature	Glucose	Triglycerides
Solubility	1. Glucose is a ***small, polar and hydrophilic**** molecule;	2. while triglycerides are ***large, nonpolar and hydrophobic**** molecules due to the hydrocarbon chains;
	3. It is soluble in an aqueous medium and can be easily transported via the bloodstream and within the cytoplasm of the cell;	4. Hence, they are ***insoluble**** in an aqueous medium, cannot be easily transported in aqueous medium / needs to form lipoprotein complexes with proteins to be transported in blood;
Size	5. It is small enough to undergo facilitated diffusion through the cell surface membrane via carrier proteins to cells where it can be oxidised;	6. The large size make it difficult to cross the cell surface membrane / needs to enter the cell via receptor-mediated endocytosis;
Production of ATP	7. Glucose can be phosphorylated to make it reactive and committed to the glycolytic pathway where a net 2 ATP molecules can be produced;	8. Triglycerides have to undergo many oxidation reactions before it can be broken down fully to produce energy/ ATP;

9. Glucose is the only substrate that can be used by the brain cells for respiration; (bonus)

(max 4 marks from part A)

B. Why triglycerides are good storage molecules

10. For ***same mass of food**** , triglycerides produce about **twice** the amount of metabolic energy compared to carbohydrates (e.g. glycogen or starch) upon oxidation during respiration;
11. Triglycerides contain more carbon and hydrogen but less oxygen atoms per gram compared to glucose/ Triglycerides contain a higher proportion of carbon-hydrogen bonds and lower proportion of oxygen atoms per gram compared to glucose; [R: ratio of C:H]
12. Thus, triglycerides are a more efficient, compact store of energy;

13. Having a compact energy store is useful in for animals who are mobile and need to carry their energy stores with them;
14. Triglycerides are ***insoluble in water**** because the long, ***non-polar hydrophobic**** hydrocarbon tails make up most of the molecule. They are ***non-polar**** and ***cannot form hydrogen bonds with water****;
15. Thus, triglycerides do not affect the water potential within living cells and can serve its role as a good energy store in organisms.
16. ***Oxidation*** of triglycerides also releases ***metabolic water**** in the process which is used as a source of water for animals especially desert animals;
17. Other functions
    a. Protect internal organs by cushioning organs such as kidneys from mechanical damage;
    b. Thermal insulation by thick layer of subcutaneous fat in seals and other marine mammals, protecting them from cold weather;
    c. Improve ***buoyancy*** in marine mammals like the whale as lipids are less dense than water;
    d. Lipids can function as a reservoir for storage of ***fat soluble vitamins*** like vitamins A, D and K;
18. AVP;

QWC (At least 2 comparisions between glucose and triglycerides in part A and must have part B)

[Total: 25]

5(a) The fluid mosaic hypothesis for the plasma membrane was formulated by Singer and Nicolson in the early 1970s and is universally accepted.

With named examples, discuss the significance of the properties of fluid mosaic model in living organisms. [15]

The fluid mosaic hypothesis for the plasma membrane was formulated by Singer and Nicolson in the early 1970s and is universally accepted.

With named examples, discuss the significance of the properties of fluid mosaic model in living organisms. [15]

Properties of cell membrane.

1. The cell membrane is ***fluid****, phospholipids and proteins which are free to move laterally within a layer;
2. ***Selectively permeable**** [R: partially]
    a) due to ***hydrophobic core**** of the phospholipid bilayer ;
    10b. due to the presence of proteins with hydrophilic channels / pore;
3. The cell membrane is ***mosaic**** because embedded proteins are arranged randomly among the phospholipid bilayer;

Significance 1: Importance/significance of being fluid (Max 8, Max 2 for each sub-point)
– need to link the following point to fluidity (property), not just describe each point / process.

4. Permeability of the membrane

a) Transient pores allows small / hydrophobic molecules to cross the membrane e.g steroid hormones / ligand – cell signaling

5. Self-sealing allows membranes to pinch off to form vesicles

6. Endomembrane system

a) Fluidity of membrane allows the endomembrane system to be inter-connected / budding / pinching of vesicles to from 1 set of internal membrane to another such as rough endoplasmic reticulum
b) to transport materials between organelles for protein modification and packaging
c) vesicles to fuse with plasma membrane to place surface proteins on plasma membrane.

7. Bulk transport

a) Fluidity allows transport of large or hydrophilic material through bulk transport/endocytosis and exocytosis;
b) **Exocytosis e.g. secretion of Ab / enzymes / insulin / hormones**
c) **phagocytosis**, APC / macrophages taking in pathogen.
d) **Receptor-mediated endocytosis** – B lymphocytes taking in pathogen upon BCR binding to specific complementary pathogen;

8. Cell division / elongation / changing shape of cells

a) elongation of cell – e.g. during anaphase / growth of root cell ;
b) During cytokinesis in animal cell, cell surface membrane invaginates / forming the furrow
c) **binary fission*** in bacteria during division
d) mobility - unicellular organisms moving

9. Bacteria

a) the fluidity of the membrane allows bacteria to undergo binary fission;
b) Allows the uptake of foreign DNA during ***transformation****
c) The fluidity of the membrane allows the formation of sex pilus to facilitate transfer of F plasmid during ***bacterial conjugation****;

Other AVP
Allows RTK subunits to move along the membrane to dimerise.

Significance 2 : selectively permeable (Max 5, Max 2 for each sub-point)

10. a. hydrophobic core of the bilayer, the membrane only allows small hydrophobic molecules to cross the membrane directly without assistance / repels charged and most polar molecules such as ions, amino acids, organic acids and simple sugars.

10b. transport proteins provides **hydrophilic* channel / pore** for **charged** and **most polar** molecules to pass through.

11. Compartmentalization / Formation of unique environment for highly specialised activities - e.g lysosomes, acidic environment that allows its enzymes to work at its optimal pH.

12. Accumulation of charged ions and formation of chemical gradients across membranes.

a. e.g. establishes a proton gradient / accumulation H^+ ions in the intermembranal space in the mitochondria / within the thylakoid space in chloroplasts
b. e.g. accumulation of high concentration of Na^+ ions outside the axons and high concentration of K^+ inside the axons generates the resting potential in preparation for an action potential

13. Storage of food source

Significance 3 : Importance of being mosaic / role of protein (Max 8, Max 2 for each sub-point)
Proteins embedded on membrane

14. **photo / respiration** – ETC / pigments /
    a) Require proteins (electron carriers) to be arranged in order;
    b) To facilitate electron transfer along electron transport chain – OP and PP
    c) Idea of carrying out chemiosmosis (ATP synthase / proton pumps);
    d) Photosynthetic pigment

15. **cell signalling**
    a) specific protein receptors to be present on cell membrane e.g. insulin/glucagon
    To allow specific signalling ligand to recognize and bind signal to be transmitted into cells
    b) signal to be transduced e.g. adenylyl cyclase

16. **cell-cell recognition / Immunology**
    a) Ref immune cells eg. B cell/macrophage/ T helper cell
    b) specific receptor bind to specific antigen eg. BCR / TCR with specific antigen binding sites
    c) antigen presenting, e.g. MHC on APC / B cell
    d) opsonisation, e.g Fc receptors on macrophages
    e) cell adhesion in formation of tissue.

17. **transport proteins**
    Allow water-soluble ions, e.g. glucose, amino acids, Na^+ etc to be transported in and out of cell

18. enzymes
e.g. cellulose synthase complexes on the plasma membrane - actively synthesize cellulose

(b) Outline how phenotypic variation is brought about in named animal viruses. [10]

Introduction

1. Viruses contain ***genetic material (DNA or RNA)*;***
2. The genetic material can undergo accumulation of ***mutations**** when viruses are in a living ***host cell**** ;
3. Ref. to an e.g. of a mutation;
4. Hence, viruses can express differences in their surface protein structure, leading to ***different conformation**** thus phenotypic variation;

HIV

5. HIV lack of ***proof reading ability/mechanism**** of ***reverse transcriptase**** in HIV results in errors in genome replication;
6. Viral RNA is ***single stranded***/does not have backup copy to carry out repair mechanism;
7. There is fast/high rate of replication of the virus resulting in faster accumulation of errors over time,
8. Hence modified surface antigens eg ***GP120 and GP41****;
9. HIV undergoes a rapid rate of ***antigenic drift****; (only if a proper explanation for antigenic drift is given for HIV)

Influenza virus

10. When the influenza virus replicates in its host cell, mutations frequently occur due to the poor ***proof reading mechanism/ability**** of the viral ***RNA-dependent RNA polymerase****
11. fast replication rate of the virus resulting in accumutation of mutations in the viral genome over time.
12. These mutations produce viruses with modified surface antigens eg. ***haemagglutinin* or neuraminidase****;
13. Hence influenza undergoes ***antigenic drift****; (only if a proper explanation for antigenic drift is given for influenza)

14. ***Antigenic shift**** **involves** ***random genetic reassortment**** ;
15. Thus when new viruses are assembled in the host cell, new combinations of RNA segments can come together;
16. A species barrier may be crossed;
17. Random genetic reassortment can produce viruses with ***NEW**** **surface antigens** (i.e. new glycoproteins such as haemagglutinin or neuraminidase);

18. AVP (max 1m)

QWC: mention of 2 different named viruses and both antigenic drift and shift – 1m

[Total: 25]

PAPER 4

(a) You will need to carry out a serial dilution of the 10.00% protein solution, **P**, to reduce the concentration by half between each successive dilution.
You will need to prepare four concentrations of protein solution in addition to the 10.00% protein solution, **P**.
After the serial dilution is completed, you will need to have 5 cm^3 of each concentration available for use.

(i) In the space below, draw a table to show how you will prepare the serial dilution. [3]

Final concentration of protein solution/%	Concentration of protein solution used/%	Volume of protein solution used/cm^3	Volume of **W**/water used/cm^3
10.00	10.00	10.0	0.0
5.00	10.00	5.0	5.0
2.50	5.00	5.0	5.0
1.25	2.50	5.0	5.0
0.63	1.25	5.0	5.0

1. **C** – Headings and units with correct final concentration of protein solution and concentration of protein solution used;
2. **V** - correct volumes of protein solution and water (**W**) used;
3. **P** - concentration recorded to 2 dp and volume recorded to 1 dp ;

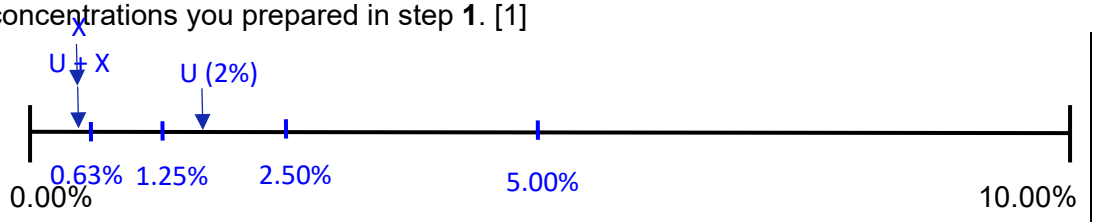
(ii) Record your results in an appropriate table. You may use the same symbols for more than one test-tube. [2]

protein concentration/%	symbol
10.00	+++++
5.00	++++
2.50	+++
1.25	++
0.63	+
0.00	-

1. **H** - correct heading with units for independent variable (protein concentration/%) correct heading for dependent variable (symbol);
   A: colour intensity, colour after 1 min, etc (instead of symbol) in heading
   R: concentration of P in heading
2. **P** - records readings for all concentrations with symbols with decreasing trend;

(iii) Fig. 1.1 shows a scale of protein concentrations used in this investigation. The position for 0.00% and 10.00% are shown on the scale.

Complete the scale in Fig. 1.1 by showing the positions of the protein concentrations you prepared in step **1**. [1]



correct positions of protein concentrations**;**

(iv) Use your results in **(a)(ii)** and **(a)(iii)** to estimate the protein concentration of **U**, **X** and **U+X**. Show your estimates on Fig. 1.1 by drawing labeled arrows (↓) at the correct positions on the scale. [2]

1. U correctly labeled at 1.25%<U<2.5%
   A: U=1.25 or U=2.5
   R: if U is greater than 2.5
2. Both X and U+X labeled at 0%<X<0.63% and 0%<U+X<0.63%
   R: if X or U+X is pointing to 0

(v) Using your result in **(a)(iv)**, estimate the concentration of protein in **U** over a period of 24 hours and state the possible medical condition of the patient indicated by **U**. Show your working in the space provided. [2]

1. Correct calculation
   10.00% ➔ 1000mg
   2.00% ➔ 1000/10 X 2 = 200 mg
   (A: range of values between 125 to 250mg)
   (ECF: if U in a(iv) is incorrect)
2. Correct medical condition - Urinary tract infection or kidney tubular disease

(vi) Solution **X** was extracted from the human gastrointestinal tract.

Suggest the identity of solution **X** and explain the basis of your suggestion. [3]

1. X is an enzyme/protease/pepsin/peptidase. (A gastric juice containing protease)
2. which digests the proteins into amino acids;
3. Thus after X was added the concentration of U decreased but it did not reach 0 due to the presence of X which is an enzyme that is protein in nature and hence a slight purple colouration was noted when the biuret test was conducted on both U+ X and X.

(b) **(i)** Outline how a colorimeter is set up so as to obtain a correct measurement of the colour intensity of a coloured solution placed in the cuvette. [2]

*any **two** from:*

1. set wavelength to use**;**
2. set colorimeter (absorbance) to zero/calibrate**;**
3. using water (to calibrate colorimeter)**;**

(ii) State the limitation of the experiment described in part **(a)**, that can overcome by the use of the colorimeter. [1]

1. human perception/interpretation of colour intensity is subjective and this affects the determination of the colour intensity;
2. colorimeter enable objective, numerical determination of colour intensity;

R: determine colour

(iii) Plot a calibration curve using the data given, on the grid in Fig. 1.3.

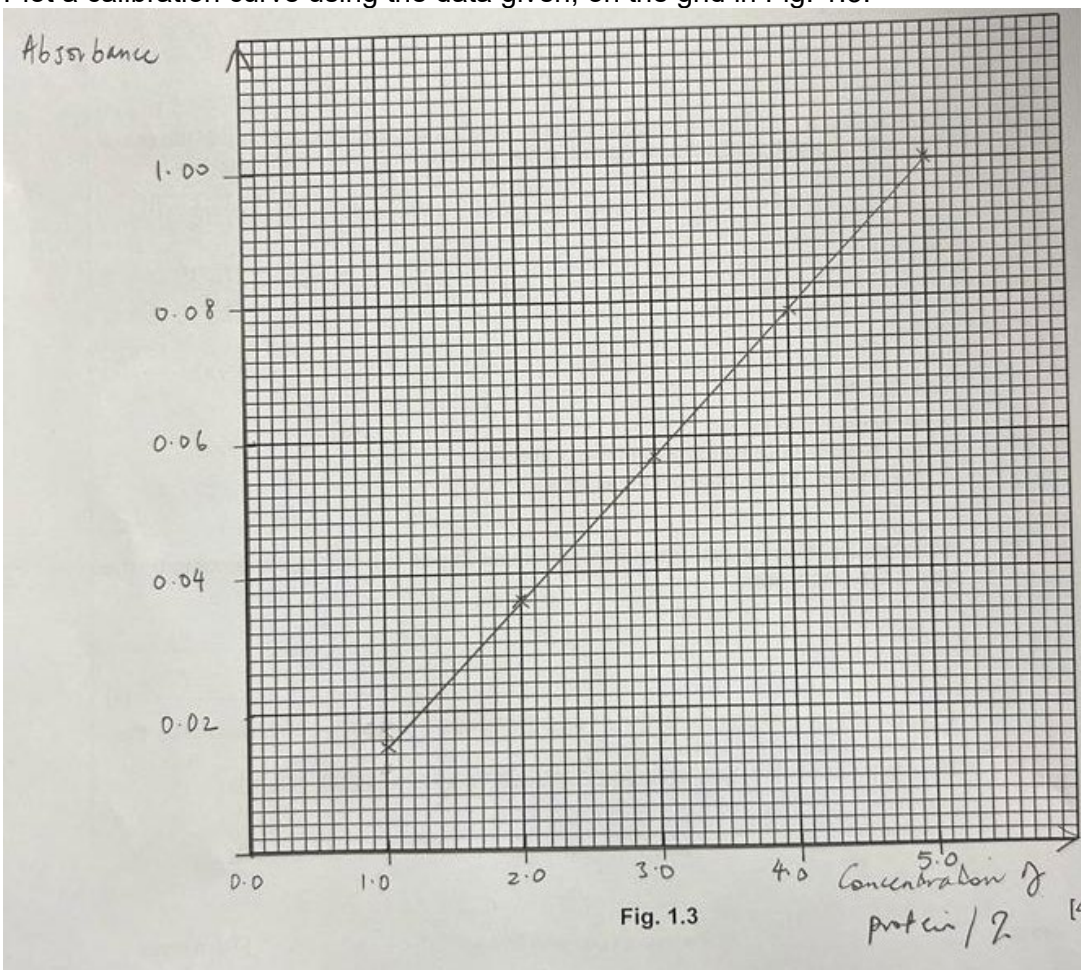


Fig. 1.3 [4]

1. **S**cale : 2/3 of graph provided
2. **A**xes : correct with units (x-axis: protein concentration/% and y-axis: absorbance/ arbitrary units)
   Penalise for: No intervals markings, No 0.0 (origin)
3. **P**ts : all 5 points correctly plotted using X
4. **L**ine : smooth curve/line passes all pts
   Penalise for extrapolation

(iv) Describe how the student could modify the previous experiment to determine the concentration of protein in sample **R**.
*Do **not** repeat any detail of how the standard protein solutions are prepared, the biuret test and how the colorimeter would be used.*
Your method should be set out in a logical way and be detailed enough to let another person follow it. [5]

1. Using a 10cm^3 syringe, add 5 cm^3 of distilled water to 5 cm^3 of sample R; This is a 2x dilution. (compulsory point) A other dilutions e.g. 1 in 2, 1 in 4, etc (must match with pt 5)
2. Conduct biuret test on the diluted sample R from step 1;
3. Transfer the diluted sample R after the biuret's test into a cuvette, measure the absorbance of the solution using the colorimeter;
4. Use the absorbance of the diluted sample R, read off the calibration/standard curve to find the concentration of protein;
5. Multiply the concentration of protein by the dilution factor of 2 (must match with point 1) to obtain the protein concentration in undiluted sample R;

[Total: 25]

2(a) Compare the morphology of **W1** and **W2** in the space below. [3]

Differences

	Point of comparison	**W1**	**W2**
1	number of seed	There are less seeds.	There are more seeds.
2	hair on skin	There are less hair on skin/shorter hair on skin. R no hair A smoother	There are more hair on skin/ longer hair on skin.
3	colour of flesh	The flesh is yellow.	The flesh is green.
4	size of middle core	The middle core is smaller.	The middle core is larger.
5	shape of core/middle	The shape is circular.	The shape is oval.
AVP	colour of skin	The skin yellowish brown in colour.	The skin darker brown in colour.
AVP	shape of cross section of fruit	The shape is circular.	The shape is oval.

Max 2

Similarities:	
6	Both have black seeds/flesh/skin;
7	Both have a pale middle core.
8	Both have hair.
9	Both have brown skin.
10	Both have numerous seeds arrange around the centre core/middle of the fruit

Max 2

(b) **(i)** Cut a 5mm thick transverse slice of specimen **W1** on the cutting board. The slice should contain a full complement of seeds and should not be taken from the extreme ends of the fruit.

Blot dry the cut surface with a paper towel. Cover one cut surface of the slice with bromocresol green, **G**. Leave it to stain for one minute. **G** stains proteins blue.

In the space provided, draw a large, detailed diagram of the cut surface of the slice as shown by the shaded area in Fig. 2.1.

You are required to use a sharp pencil for drawings.

Your drawing should show details of the arrangement of different regions and their correct shapes and proportions. Indicate where the highest concentration of protein is present. [4]

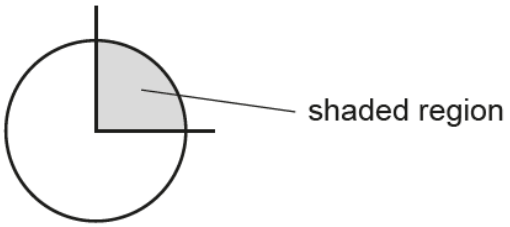
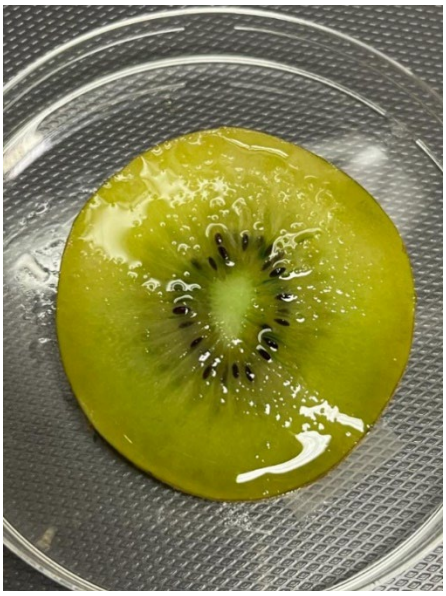
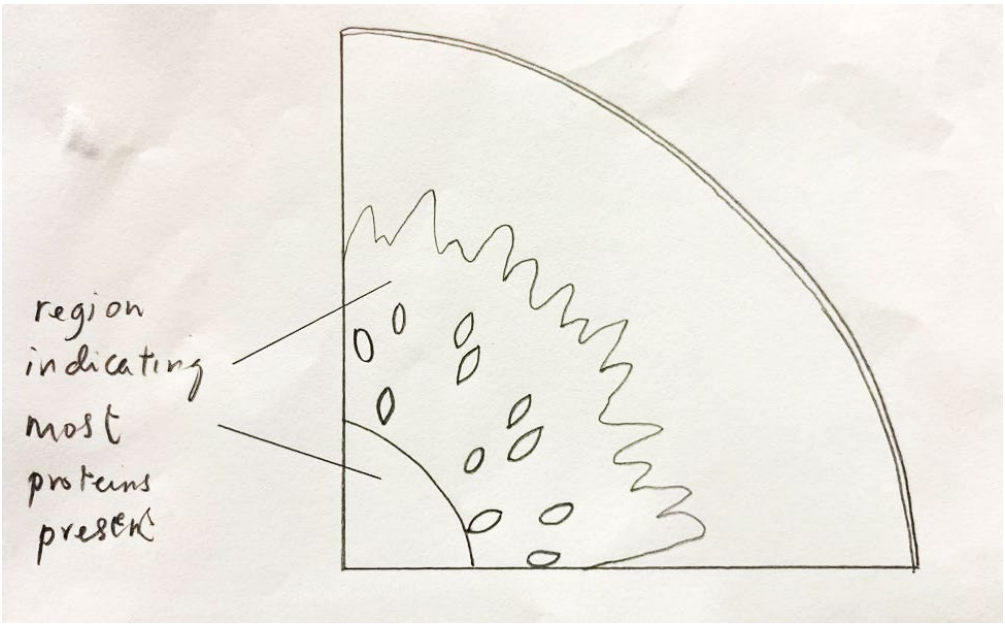


Fig. 2.1

1. Scale : detailed diagram of appropriate size (>3/4 space given) + no shading;
2. Proportion: Correct proportion of layers including correct proportions of core compared to the diameter of the fruit, and skin, size of the seeds;
3. Accuracy : skin, seeds, locules + correct section drawn;
   R if draw LS
4. Annotations : blue green core/locules indicating highest concentrations of proteins present;





(ii) You are required to estimate the total number of seeds that were present in the fruit from which **W1** was taken.

- Cut the 5mm slice of **W1** into equal quarters.
- Place one of these quarters on the cutting board and, using the forceps, squeeze out all of its seeds.

Count the number of seeds present: ……6 - 26……....

Repeat this for another quarter.

Count the number of seeds present: ……6 - 26…… [1]

(iii) Plan how you would estimate the number of seeds in the whole fruit, taking into consideration and explaining any allowance that you may think is necessary. [4]

1. Find the mean of the number of seeds by adding the 2 samples and divide by 2;
2. Estimate the number of seeds in a slice by multiplying the mean by 4;

3a. Show how they derive 10 slices

3. Fig 2.2 shows that 10 slices can be obtained from the whole fruit, multiplying the number of seeds per slice by 10 would give the total number of seeds in a fruit;
4. As the region containing seeds taper at the ends, there should be less seeds at the ends and hence should be taken into consideration. Hence a good estimate would be the total number in a slice multiplied by 7/8/9 instead;

(iv) Using the method you described in **(b)(iii)** and data collected from **(b)(ii)**, estimate the number of seeds found in **W1**. Show your working clearly. [2]

1. Total number of seeds in 2 quarters 12 + 15 = 27
   Mean number of seeds in 1 quarter 27/2 = 13.5;

2. Total number of seeds in 1 slice = 13.5 x 4 = 54;
   Total number in 8 slices = 54 x 8 = 432 seeds;

(c) **(i)** Complete the calculation to find the value of *t* for the concentration of sucrose in the fruits. [2]

$$t = \frac{|23-15|}{\sqrt{\frac{4^2}{30}+\frac{3^2}{30}}}$$

= 8/0.9 [1]

t = 8.9 [1]

(ii) State the null hypothesis for **(c)(i)**. [1]

There is no significant difference (R: insignificant) in the mean sucrose concentration (R: sweetness) in the samples of W1 and W2 variety of fruit; Any difference is due to random chance.

(iii) State the conclusion that could be gathered from the *t*-test. [3]

1. At **58** degrees of freedom (v) and t = 8.9, p < 0.001; A t > 2.02 or 2.00
2. At 5% level of significance, ***p < 0.05**** so we reject the null hypothesis, H_0;
3. and conclude that mean of concentration of sucrose in W1 is ***significantly different**** than the mean concentration of sucrose in W2 and the difference is not due to chance alone;

[Total: 20]

3(a) You are given slides **S1** and **S2** in a petri dish

(i) Using the slide **S2**, determine the diameter of the field of view at high power magnification. [1]

5 stage micrometer division x 100 µM
= 500 µM

1. Correct working and correct final ans expressed nearest µM
   R mm

(ii) Calculate the volume of blood under the field of view at high power magnification. [2]

$V = \pi r^2 h$
$= \pi \times 250^2 \times 100$
$= 19\ 634\ 954\ \mu M^3$

1. Correct working using ans from part **(i)**
2. Correct final ans expressed to nearest μM^3

(b) **(i)** Place slide **S2** under higher power magnification.
Use slide **S2** to determine the dimensions of each eye piece graticule unit.
Show your working. [1]

40 eye piece graticule unit = 100μM
1 eye piece graticule unit = 100/40 μM = 2.5 μM

1. Correct working and final ans

(ii) Place slide **S1** under high power magnification.
Locate a lymphocyte and use the markings on the eye piece to measure the diameter of the:

a) nucleus (N); and [1]

4 eyepiece graticule unit x 2.5 μM = 10 μM

1. Correct working and final ans. (ecf allowed for 2.5 μM)

b) cell (C). [1]

5 eyepiece graticule unit x 2.5 μM = 12.5 μM

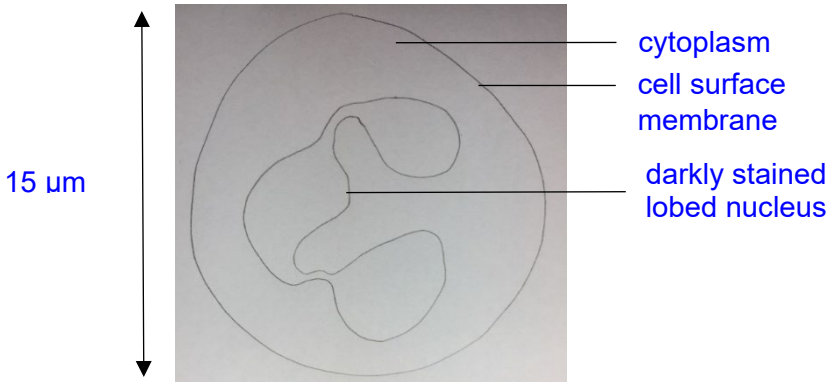
1. Correct working and final ans. (ecf allowed for 2.5 μM)

(iii) Calculate the N:C ratio of the lymphocyte, and express your answer in whole numbers. [1]

10 : 12.5 = 20: 25 = 4 : 5

1. Correct working and final ans (in whole numbers)

(c) Place slide **S1** under high power magnification.
In the space below, make a labeled drawing of a neutrophil. [3]



High power drawing (X400) of a human white blood cell (neutrophil)

1. **S**ize (use 2/3 of the space provided)
2. **A**ccuracy → Drawing shows nucleus with at least 2-5 lobes
3. **P**roportion →Size of nucleus proportional to cell
4. **L**abels → lobed nucleus, cytoplasm, cell surface membrane

[Total: 10]