Section A
Answer all the questions in this section.

1 (a) Evaluate the effectiveness of vaccines against the rise of antibiotic resistant strains of pnemococcal bacteria. [4]

[Max 4]

- Highly effective/effective to a large extent;
- Antibiotic is administered when a bacterial infection has already occurred;
- Ref. to the idea that bacteria have reached a population large enough to cause disease / symptoms;
- Ref. to the idea that bacteria have multiplied many times;
- Ref. to the idea that each time the bacteria divide, their DNA is copied and mistakes in this process can create variations within the population;
- Antibiotic resistance can be one of the variations;

- Presence of antibiotics can act as a selective pressure;
- Select for more resistant bacteria;
- Resistant bacteria survive and reproduce;
- Pass down their antibiotic resistance alleles;
- Increase the frequency of the antibiotic resistance alleles within the bacterial population;

- Vaccines are administered prior to an infection;
- Ref. to the idea that infection can be brought under control before bacteria has the chance to multiply;

- Antibiotics tend to target one specific bacterial protein / mechanism;

- Vaccines can expose the immune system to a huge number of bacterial proteins / antigens;
- Promotes the development of a vast repertoire of antibodies that can fight against the bacterial infection in various ways;
- Ref. to action of antibodies: e.g. opsonisation, agglutination, neutralisation
- Ref. to idea that chances of bacteria simultaneously evolving resistance to counter every type of antibody produced is slim;
- Vaccines can curb overuse of antibiotics;

(b) Describe how a typical T4 bacteriophage can work against an antibiotic-resistant bacterium. [3]

- T4 bacteriophage binds to specific receptors on antibiotic-resistant bacterium;
- The T4 phage penetrates the bacterium cell wall by contracting its contractile sheath of the tail which drives the hollow tube of the tail into the host bacterium; facilitating the entry of viral DNA;
- The phage then replicates its genome and uses the bacterium's protein synthesis machinery to synthesise phage proteins and structural components;
- The T4 phage components assemble around the viral genome to form new mature phage particles;

- A phage-coded lysozyme breaks down the peptidoglycan cell wall of the host cell, causing cell lysis and release of the new T4 bacteriophages;
- Resulting in the death of the bacterium;

(c) (i) Using the information given, describe how a bacterium appears to have acquired antibiotic resistance due to the release of such plasmid DNA. [2]

- Competent bacteria cells within the population will take up the plasmid DNA via transformation;;
- Plasmid DNA may contain genes for antibiotic resistance;;

(ii) Suggest how this could lead to an increase in the proportion of bacteria with antibiotic resistance in the population. [3]

- The transformed bacteria cells can then undergo conjugation with the bacteria near them; or binary fission
- Within the population, there are now antibiotic resistant bacteria and those that are not;
- Use of antibiotics act as a selection pressure;
- Antibiotic resistant bacteria have a selective advantage and they are selected for;
- These selected bacteria will survive, reproduce and pass on their alleles to the next generation;
- increasing frequency of antibiotic-resistant bacteria within the population;

(d) Briefly comment on the validity of the hypothesis based on the results shown in Fig. 1.2. [3]

- The hypothesis seems valid;;
- T4 bacteriophage results in less intact plasmid and chromosomal DNA within the bacteria whereas bacteria that have interacted with SUSP1 and SUSP2 has more intact plasmid and chromosomal DNA;;
- Endonucleases present in the T4 bacteriophages allows for the digestion of the bacteria's plasmid and chromosomal DNA whereas these endonucleases are absent in SUSP1 and SUSP2;;

(e) As a good Biology student, name and describe a procedure that the researchers can undertake to determine the presence or absence of the hydrolytic endonucleases in the "superspreaders". [4]

- Southern Blotting;
- Place the gel in a mixture of alkali and salt to denature the double stranded DNA fragments into single stranded DNA;
- The gel is then covered with a nitrocellulose filter;
- Additional absorbent papers are added on top of the nitrocellulose filter to draw up the single stranded DNA from the gel by capillary action and transferred them onto the nitrocellulose filter;
- Nitrocellulose filter is baked at 80^{o}C so that the DNA is permanently bound to the filter;
- The filter is then exposed to a solution containing radioactively labeled single-stranded DNA probes;

- ref to nucleic acid
- which are complementary to the DNA sequence of the endonucleases;
- Excess probes are then washed off and the bands are visualised under autoradioraphy;

(f) Besides mutation, suggest how the environmental scientists' prediction miht come true. [1]

- As temperature rises, permafrost melting occurs;;
- releasing new bacteria and viruses, which were once trapped in the frozen soil, into the environment;;

OR

- As sea level rises, flooding of low lying lands occurs, less land available;;
- different species of organisms may find themselves in closer proximity, allow for greater antigenic shift in viruses to occur, resulting in new strains of viruses formed;;

- AVP

[Total: 20]

2 (a) (i) Label on Fig. 2.1, two critical positions where a cell can be regulated in the mitotic cell cycle. [2]

- label - meataphase checkpoint; DNA damage checkpoint; G2 checkpoint

(ii) Explain the significance of the critical positions where a cell can be regulated in the mitotic cell cycle. [2]

- metaphase checkpoint – ensure all chromosomes are attached to mitotic spindle
- G2 checkpoint – check no DNA damage and chromosomes all replicated
- G1 checkpoint – cell size is large enough, nutrients available, growth factors present.

3 Identify the two areas labelled A in the dividing cells in Fig. 2.2 and outline their function. [3]

- spindle apparatus / spindle fibres; Accept spindle / microtubules / tubulin / centrioles / microtubule organising centres / MTOCs [1]

function to max 2

- attach to chromosomes / kinetochores ;
- detail of, elongation / structure / shortening, of microtubules ;
- for movement of chromosomes ;
- during mitosis ;

Accept if centrioles given as identity

- forms poles of the cell ;
- organises the spindle ;

4 (i) Explain why it is necessary to increase the permeability of the cell surface membranes before staining cells using the technique of immunofluorescence. [2]

- Membrane has as hydrophobic hydrocarbon core;
- Antibodies are hydrophilic;
- Antibody molecules too large to fit between phospholipid molecules in the bilayer;;

(ii) Suggest one advantage of using immunofluorescence in studying the changes that occur in cells during cell division. [1]

- locate position of specific, proteins / structures ;
- antibody molecules have complementary shape to target, proteins / structures ;

- can see distribution of, proteins / structures, in light microscope ;
- do not need to prepare sections for the electron microscope ;
- easier to look at a large number of cells than in EM ;

- higher degree of specificity than using other staining techniques ;
- idea of variable regions of antibodies giving greater specificity ;

5 (i) Explain why embryonic stem cells do not give rise to tumour. [3]

1. They are able to divide continuously but not excessively/uncontrollably;
   Cell division of stem cells can be regulated;
   Stem cells will only divide when necessary e.g. presence of growth factors;
2. As they obey cell cycle control/ stop appropriately at cell cycle checkpoints when conditions at previous stage are not met;;
   Accept if students refer to arrest of cell cycle to repair DNA damage/ cells send to apoptosis when DNA damage is irreparable;
3. Tumor suppressor genes (TSG) and proto-oncogenes are both functional / not mutated;;
   Note: A maximum of 2 marks is warded if there is no mention of TSG and proto-oncogenes anywhere in the answer.

Others: stem cells show contact inhibition;

(ii) Discuss one ethical concern that this type of research attempts to address. [2]

<u>Ethical concerns</u>: (any 1 with elaboration)

- Using adult cells does not involve use and destruction of embryos;;
- Reason – destruction of embryos (even excess embryos from IVF programme) is morally unacceptable/ repugnant to a sector of society that considers the embryo as life or potential of life;;
- AVP

[Total: 17]

6 (a) Explain why ectotherms are "susceptible to the negative effects of rising temperatures". [2]

- Ectotherms cannot regulate body temperature;
- Increased temperatures may result in the lizard's body temperature exceeding an upper critical temperature;
- Exceed optimum temperature of enzymes;
- can inhibit biochemical processes;

(b) With reference to Figure 3.1, account for two effects of global warming on the lizard's foraging period. [3]

- Global warming decreases the total length of foraging period / to about 2/3 of the period before global warming;;
- Only one foraging period instead of two / no more foraging period towards end of the day;;
- Global warming increases atmospheric temperature which increases minimum and maximum operative temperatures of lizard throughout the day (as they are ectotherms);;
- Pushes operative temperatures above acceptable range / narrows timeframe where operative temperatures fall within acceptable range;;
- E.g. minimum operative temperature towards the end of the day remains higher than acceptable range;;

(c) Explain why the change in the foraging activity due to increased temperatures can lead to the extinction of the local lizard populations.[1]

- Limited food acquisition due to shorter foraging periods will mean that the lizards feed less;;
- This leads to lower growth rates of lizards;;
- Idea of limiting reproduction: Eg. Limit the energy they have available for reproduction/ or limit the number of offspring they have;;
- Decrease in number of offspring in subsequent generations/ Without new individuals to replace those that die, the population will eventually go extinct;;

(d) Justify the predictions made by the scientists as shown in Fig 3.2. [3]

- When lizards move higher up in terms of elevation or towards higher latitude where the temperature is lower;
- Longer foraging period, more food and energy and
- hence probability of extinction is lower;

- Probability of extinction is greater at the lower elevation of the lizards' distributions and lower latitude/ latitude that is closer to the equator;

- Temperature at lower elevation and latitude is already high / near critical limit of lizards;
- Global warming leads to an increase in temperature at the lower elevation and lower latitude, resulting in decrease in foraging;
- Less energy for growth and reproduction, resulting in extinction;

- Viviparous (live-bearing) lizards have a higher probability of extinction than oviparous(egg laying) lizards as live bearing lizards would need more energy to sustain the development of the embryo;
- A decrease in foraging activity due to increased temperatures would decrease their intake of food, decrease energy available to sustain embryo growth and development;;
- Compared to those who lay eggs/ oviparous as the egg yolk provides the source of energy for the developing embryo;;

(e) Suggest how the live-bearing and egg-laying lizards evolved to become different species from a common egg-laying ancestor.[4]

- Different populations of the ancestral egg laying species are separated from each other due to physical barriers (allopatric) or behavioural reasons (sympatric) / idea of isolation;
- * No gene flow between the groups;;
- * Mutations also arise independently in each population ;;
- Fertile individuals from the different populations are no longer able to mate with each other (i.e. absence of gene flow) and so result in two distinct gene pools.
- *Over a long period of time, the genetic variations in the different populations increase (eg ability to bear young live);

[Natural selection – cap at 2 mk]

- Different environments have different selection pressures on the organisms;
- One group faces high predation (egg eating predators);
- hence live-bearing individuals in that area are selected for;
- survive and reproduce and pass down advantageous alleles to offspring;

<u>* MUST HAVE POINTS</u>

[Total: 13 marks]

Section B

4 (a) Compare the differences between B and T lymphocytes and describe how cell signalling helps B lymphocytes to play a role in the production of high-affinity antibodies with different effector function. [13]

Difference between B & T cells [max 8]

- Naïve B cells develop from immature B cells in the bone marrow;;
- Naïve T cells develop from immature T cells in the thymus;;

- B lymphocytes are important in humoral immunity in which antibodies neutralise and eradicate extracellular microbes and toxins;;
- T lymphocytes are important in cell-mediated immunity in which cytotoxic T cells eradicate intracellular microbes/microbes that have infected body cells;;

- Only B cells lymphocytes can secrete antibodies;;
- Helper T cells secrete cytokines to activate other cells of the immune system e.eg. phagocytes to destroy microbes and activate B cells to class switch and become antibody-secreting plasma cells;
- Cytotoxic T cells secrete perforins and granzymes to kill infected cells by apoptosis;

- B lymphocytes express membrane antibodies (BCR) that recognise intact antigens which could be found on the surface of the pathogen
- T lymphocytes express T-cell receptors (TCR) that recognise peptide fragments of protein antigens displayed on other cells

- B cells ingest protein antigens, degrade them and display peptides bound on MHC molecules
- T helper cells with complementary receptors bind to MHC bound peptides and
- release cytokines to cause the B cell to differentiate to form plasma cells

Cell signalling

- Cytokines released by T helper cells then bind to cytokine receptors on the B cell,
- there is a conformation change in the receptor that
- results in a signal transduction into the B cell through the triggering of a phosphorylation cascade/ signal amplification
- Which leads to a cellular response that causes the gene for cytidine oxidase to be switched on
- The enzyme cytidine oxidase causes hypermutation of the VDJ regions of the antibody genes
- To produce antibodies of higher affinity/ specificity to the antigen
- Hence T helper cells also stimulate the production of antibodies with higher affinity for the antigen through the process of affinity maturation

- Another cellular response is to cause genetic recombination of the antibody genes to result in heavy chain class-switching to produce antibodies of different classes (IgG, IgA or IgE) of the same specificity.

(b) Describe how bacteria reproduce asexually and suggest some advantages of such a process. [12]

- Binary fission;; [MUST HAVE]

<u>DNA attachment</u>

- First the DNA attaches itself to the cell membrane or to a mesosome which is a highly folded region of the cell membrane.

<u>DNA replication</u>

- Replication of the DNA starts at the origin (Ori C) that is attached to the cell wall, near the midpoint of the cell.
- As the DNA uncoils, a new complementary strand is being constructed on each strand in a semi-conservative manner.
- Replication occurs bidirectionally.
- This is helped by the enzyme called DNA gyrase which removes positive supercoiling.
- DNA replication ends at the termination sequence situated opposite the origin of replication.
- Plasmids are replicated the same way as the bacterial chromosome.

<u>Cell growth and division</u>

- After DNA replication, cell growth occurs. As the cell elongates, each circular DNA strand which is still attached to the cell membrane separate.
- When the cell divides, the cell membrane folds inwards between the DNA molecules to form a double layer across the long axis of the cell.
- New cell wall layers are secreted within the membrane layers.
- This divides the cell into two smaller, identical cells which may remain together or may separate.

<u>Advantages (max 3)</u>

- It allows for rapid populating.
- Ref. idea that it conserves energy and resources.
- Ref. to idea that daughter cells are genetically identical to the parents → all favourable alleles from the parents are guaranteed to be passed down

QSE: Candidates must be able to name and describe the process of binary fission and suggest at least 2 advantages.

5 (a) Environmental factors affect the phenotype of organisms. For example, fur colour of Himalayan rabbit is affected by temperature.

Explain the significance of the environment on variation and the formation of new species. [13]

Max 12m for (1) + (2) + (3)
1m for QWC

(1) Effect of environment on phenotype [Max 6]
- Effects of environment increases variation between individuals;;
- Ref. to space / nutrients for growth / Environment affecting gene expression;;
- Plays a part in continuous variation which is controlled by the combined (additive) effects of many genes (polygenes) and environmental factors;;

(2) Role of environment on natural selection
- Genetic variation within population (giving rise to variation in phenotype further enhanced by effects of the environment) due to mutations / meiosis / sexual reproduction;;
- Environment factors serving as the selective pressure;;
- E.g. Source of food / Predator / etc.;;
- Variations in phenotype give rise to differential reproductive success;;
- Favourable alleles are passed down to later generations;;
- Change in allele frequency between generations;;

(3) Formation of new species
- Geographical / behavioural / physiological isolation between populations;;
- No gene flow between populations;;
- Accumulation of different genetic changes in each populations;;
- Correct definition of new species based on a named species concept;;
- Correct ref. to allopatric / sympatric speciation;;

QWC
- Answer includes at least 2 points from each section;;

(b) The Polymerase Chain Reaction (PCR) was a revolutionary method developed by Kary Mullis in the 1980s. Outline the main principles of PCR and discuss how DNA replication by PCR differs from the process of how lambda phage replicates its genome via a lysogenic cycle within its host cell. [12]

Principles of PCR [max 6]
- PCR is based on using the ability of DNA polymerase to synthesize a new strand of DNA complementary to the offered template strand.
- ref to semi-conservative replication
- ref to complementary base pairing
- A thermostable Taq polymerase is used which works at an optimum temperature of 72^0C

- so that the PCR reaction can be carried out in a thermocycler.
- Because DNA polymerase can add a nucleotide only onto a preexisting 3'-OH group,
- a primer is needed to which it can add the first nucleotide.
- PCR involves the use of a forward and a reverse primer to flank the region to be amplified
- This makes it possible to delineate /mark out a specific region of template sequence that the researcher wants to amplify.
- As the PCR involves cycles of denaturation, annealing and elongation, at the end of 20-30 cycles, the specific sequence will be accumulated in billions of copies

Difference between PCR and DNA replication in prokaryotes [max 6]

PCR	DNA replication of lambda phage
DNA primers, a forward and a reverse primer designed in the laboratory, are used	RNA primers, which are synthesized by primase, are used
Specific DNA sequences is being synthesized/copied	Entire genome on the circular chromosome is being copied
Replication starts where the forward and reverse primers bind	Replication starts at the single origin of replication
High temp (95oC) is being used to denature (break hydrogen bonds) and separate the DNA into single strands	Helicase is used to unwind DNA into single strands
Variation in temperature required in 1 cycle	Temperature is fairly constant throughout the entire process
No Okazaki fragments formed	Okazaki fragments formed in lagging strand
Does not require DNA ligase, primase	Requires DNA ligase, primase