KAP5 Biology: Cell Biology and Enzymes

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<u>1.1 Sub-cellular Structures: Eukaryotic & Prokaryotic Cells</u></u>

Revision Notes

Cells are either eukaryotic (animals, plants, fungi) or prokaryotic (bacteria).

Eukaryotic Cells: Have a membrane-bound nucleus and organelles.

- a) Animal Cells:
 - **Nucleus:** Contains DNA, controls cell activities.
 - Cell Membrane: Controls entry/exit of substances.
 - Mitochondria: Site of aerobic respiration, releases energy (ATP).
 - **Ribosomes:** Site of protein synthesis.
- b) Plant Cells: Have animal cell structures plus:
 - **Cell Wall:** Made of cellulose, provides structural support and protection.
 - **Chloroplasts:** Site of photosynthesis, contain chlorophyll.
 - Vacuole (Permanent Large Central): Contains cell sap, maintains turgor pressure, storage.
- **c) Bacteria (Prokaryotic Cells):** Simpler, smaller, lack a nucleus and complex organelles.
 - **Chromosomal DNA:** Single, circular loop in the cytoplasm (nucleoid region), main genetic material.
 - **Plasmid DNA:** Small DNA rings in cytoplasm, carry extra genes (e.g., antibiotic resistance).
 - Cell Membrane: Controls entry/exit of substances.
 - **Ribosomes:** Site of protein synthesis (smaller than eukaryotic).
 - Flagella: Whip-like tail for movement (not always present).
 - (Also have a cell wall, but made of peptidoglycan).

Comparison of Cell Types

Feature	Animal Cell (Eukaryotic)	Plant Cell (Eukaryotic)	Bacterial Cell (Prokaryotic)
Nucleus	Present	Present	Absent (Nucleoid region)
Genetic Material	Linear DNA	Linear DNA	Circular DNA
Plasmid DNA	Absent	Absent	Often Present
Cell Membrane	Present	Present	Present
Cell Wall	Absent	Present (Cellulose)	Present (Peptidoglycan)
Mitochondria	Present	Present	Absent
Chloroplasts	Absent	Present	Absent
Ribosomes	Present	Present	Present
Vacuole	Small/Absent	Large, central	Absent
Flagella	Absent (except sperm)	Absent (most)	Sometimes Present

Practice Questions (1.1)

- 1. Identify two structures present in plant cells but absent in animal cells, and state the function of each.
- 2. Describe the location and function of chromosomal DNA and plasmid DNA in a bacterial cell.
- 3. Compare the function of ribosomes in eukaryotic (animal/plant) and prokaryotic (bacterial) cells.
- 4. Explain the role of the cell membrane and mitochondria in an animal cell.

Answers (1.1)

- 1. **Cell Wall:** Provides structural support and protection. **Chloroplasts:** Site of photosynthesis. (Also acceptable: Large central vacuole maintains turgor pressure and storage).
- 2. **Chromosomal DNA:** Located in the cytoplasm (nucleoid region), contains the main genetic information. **Plasmid DNA:** Small DNA rings in the cytoplasm, carry extra genes.
- 3. In both cell types, ribosomes are the site of protein synthesis.
- 4. **Cell Membrane:** Controls entry/exit of substances. **Mitochondria:** Site of aerobic respiration, releasing energy (ATP).

1.2 Specialised Cells

Revision Notes

Cells differentiate to become specialised, developing adaptations for specific functions.

- a) Sperm Cells: Function is to fertilise the egg.
 - Adaptations:
 - Acrosome: Cap with enzymes to penetrate the egg.
 - Haploid Nucleus: Half the chromosome number for fertilisation.
 - Mitochondria: Packed in mid-piece to provide energy for swimming.
 - **Tail (Flagellum):** Propels the sperm towards the egg.
- **b) Egg Cells (Ova):** Function is to be fertilised and nourish the early embryo.
 - Adaptations:
 - Nutrients in Cytoplasm: Food store for the developing embryo.
 - Haploid Nucleus: Half the chromosome number for fertilisation.
 - Changes in Cell Membrane after Fertilisation: Forms a barrier to prevent multiple sperm entering.
- **c) Ciliated Epithelial Cells:** Function is to move substances (like mucus) along surfaces (e.g., airways).

- Adaptations:
 - **Cilia:** Hair-like projections that beat rhythmically to move mucus.
 - **Mitochondria:** Provide energy for cilia movement.

Summary of Specialised Cell Adaptations

Cell Type	Function	Key Adaptations
Sperm Cell	Carry male DNA to egg	Acrosome: Enzymes to penetrate egg. Haploid Nucleus: Half chromosome set. Mitochondria: Energy for swimming. Tail: Propulsion.
Egg Cell (Ovum)	Be fertilised; provide early nutrients	Nutrient-rich Cytoplasm: Food store. Haploid Nucleus: Half chromosome set. Membrane changes post-fertilisation: Prevents polyspermy.
Ciliated Epithelial Cell	Move mucus along airways	Cilia: Hair-like structures beat to move substances. Mitochondria: Provide energy for cilia movement.

Practice Questions (1.2)

- 1. Explain how the acrosome and mitochondria contribute to the function of a sperm cell.
- 2. Describe two adaptations of an egg cell and explain how each relates to its role in reproduction.
- 3. What are cilia, and how do they enable ciliated epithelial cells in the trachea to perform their function?
- 4. Why is it essential for both sperm and egg cells to contain a haploid nucleus?

Answers (1.2)

- 1. Acrosome: Contains enzymes to digest egg layers for fertilisation. Mitochondria: Provide energy for tail movement.
- 2. Nutrient-rich cytoplasm: Provides food for the early embryo. Membrane changes post-fertilisation: Prevents multiple sperm entry. (Also: Haploid nucleus half chromosome set).
- 3. Cilia are hair-like projections. They beat to sweep mucus and debris away from the lungs.
- 4. So that when they fuse during fertilisation, the resulting zygote has the correct (diploid) number of chromosomes.

1.3 Microscopy Technology

Revision Notes

Microscopes allow visualisation of cells. Electron microscopes (EM) offer higher magnification and resolution than light microscopes (LM).

- Light Microscopes (LM): Use light and glass lenses. Max magnification ~1500x, resolution ~200 nm. Can view living cells. Relatively simple, affordable, colour images.
- Electron Microscopes (EM): Use electron beams and electromagnets. Max magnification >1,000,000x, resolution <1 nm. Reveal ultrastructure (fine details like ribosomes, internal membranes). Specimens must be dead (complex preparation, vacuum). Very expensive, complex, black and white images.
- **Impact:** EM revealed ultrastructure (e.g., cristae in mitochondria, ribosomes), linking structure to function and advancing understanding of cell biology.

Comparison of Light and Electron Microscopes

Feature	Light Microscope (LM)	Electron Microscope (EM)
Radiation Source	Visible Light	Beam of Electrons
Lenses	Glass	Electromagnets
Max Magnification	~1,500x	>1,000,000x
Max Resolution	~200 nm	<1 nm
Specimen State	Living or Dead	Dead only
Preparation	Simple	Complex
Environment	Air	Vacuum
Cost/Complexity	Lower	Very High
Image Туре	Colour	Black and White

Practice Questions (1.3)

- 1. State two advantages of using a light microscope compared to an electron microscope.
- 2. State two advantages of using an electron microscope compared to a light microscope.
- 3. Why can living cells not be observed using an electron microscope? Give two reasons.
- 4. Explain how the invention of the electron microscope increased biologists' understanding of sub-cellular structures like mitochondria.

Answers (1.3)

- Can view living specimens; simpler preparation; cheaper; colour images. (Any two)
- 2. Much higher magnification; much higher resolution (reveals ultrastructure).
- 3. Requires a vacuum; preparation process kills cells.
- 4. Higher resolution allowed visualisation of internal structures like cristae, helping understand how structure relates to function (large surface area for respiration).

1.4 Number, Size, Scale & Estimation

Revision Notes

Biological structures range vastly in size (cells ~ μ m, organelles ~ μ m/nm). Understanding scale and units is key.

Estimation: Making a rough calculation.

- When to Use: Checking precise calculations; approximating size/area of structures difficult to measure directly (e.g., small organelles).
- How to Use: Compare the unknown dimension to a known reference (e.g., cell diameter, scale bar). Estimate how many times the unknown fits into the known. Calculate the estimate (Known dimension / Estimated fits).

Practice Questions (1.4)

- A student calculates the magnification of a mitochondrion in an electron micrograph to be x500. They estimate the mitochondrion in the image is about 1/10th the width of the cell image, which is 5 cm wide, and know the actual cell width is 20 μm. Use estimation to check if the student's calculation is reasonable.
- 2. Why might a biologist use estimation to determine the size of ribosomes seen in

an electron micrograph?

- 3. An image of a plant cell is 80 mm long. The actual cell is 100 μ m long. Estimate the actual length of a chloroplast that appears to be about one-quarter of the length of the cell image.
- 4. Explain one situation during a microscopy practical where estimation would be a useful skill.

Answers (1.4)

- 1. Estimated image size of mitochondrion ≈ 0.5 cm = 5000 μ m. Estimated actual size $\approx (1/10) * 20 \ \mu$ m = 2 μ m. Estimated magnification $\approx 5000 \ \mu$ m / 2 μ m = x2500. The student's x500 calculation seems potentially low.
- 2. Ribosomes are very small and may lack clear boundaries, making direct measurement difficult. Estimation provides an approximate size by comparison.
- 3. Estimated image length of chloroplast = 20 mm. Magnification = 80 mm / 100 μ m = x800. Estimated actual length = 20 mm / 800 = 25 μ m.
- 4. Estimating organelle size within a cell; checking a magnification calculation; estimating cell size based on field of view diameter. (Any one).

<u>1.5 Quantitative Units & Standard Form</u>

Revision Notes

Metric prefixes denote fractions of base units:

- milli (m): 10–3 (thousandth)
- **micro (µ):** 10–6 (millionth)
- nano (n): 10–9 (billionth)
- pico (p): 10–12 (trillionth)

Unit Conversions: Multiply/divide by 1000 for each step (e.g., 1 mm = 1000 μ m; 1 μ m = 1000 nm).

Standard Form: Expresses numbers as $A \times 10n$, where $1 \le A < 10$ and n is an integer. Useful for very large/small numbers.

- Multiplication: Multiply A values, add powers (n).
- **Division:** Divide A values, subtract powers (n).

Metric Prefixes Summary

Prefix	Symbol	Power of 10	Meaning
milli	m	10–3	thousandth
micro	μ	10–6	millionth
nano	n	10-9	billionth
pico	р	10–12	trillionth

Practice Questions (1.5)

- 1. Convert 50 millimeters (mm) into micrometers (μ m) and express the answer in standard form.
- 2. A virus particle has a diameter of 80 nanometers (nm). Convert this diameter into meters (m) and express the answer in standard form.
- 3. Calculate the following and give the answer in standard form: $(4 \times 10 6) \times (2 \times 10 3)$.
- 4. Calculate the following and give the answer in standard form: $(8 \times 103) \div (4 \times 10-2)$.

Answers (1.5)

- 1. 50 mm = 50,000 μm=5×104 μm.
- 2. 80 nm = 80×10−9 m = 8×10−8 m.
- 3. 8×10-9.
- 4. 2×105.

1.6 Core Practical: Microscopy, Magnification & Drawings

Revision Notes

Using a Light Microscope:

- 1. Start with lowest power objective lens.
- 2. Place slide on stage, secure with clips.
- 3. Use coarse focus to bring image into view.
- 4. Use fine focus to sharpen image.
- 5. Switch to higher power objective, refocus using fine focus only.

Preparing a Wet Mount (e.g., Onion Epidermis):

- 1. Drop of water on slide.
- 2. Place thin specimen layer in water.
- 3. Add drop of stain (e.g., iodine).
- 4. Lower coverslip slowly at an angle to avoid air bubbles.
- 5. Blot excess liquid.

Magnification Calculations:

- **Total Magnification** = Eyepiece Lens Mag. × Objective Lens Mag.
- Image Size, Actual Size, Magnification: M=I/A (Magnification = Image size / Actual size). Rearrange as needed: A=I/M, I=A×M.
- Units: Ensure Image (I) and Actual (A) sizes are in the *same units* (e.g., convert mm to μm) before calculating. Magnification (M) has no units.

Labelled Scientific Drawings:

 Rules: Use sharp pencil only; large and clear drawing; solid, continuous lines (no shading); draw accurately what you see; label lines straight (use ruler), horizontal, touching structure, no arrowheads, lines don't cross; include title and total magnification.

Practice Questions (1.6)

- 1. A student observes an onion cell using an eyepiece lens (x10) and an objective lens (x40). What is the total magnification?
- The student draws the onion cell, and the drawing measures 60 mm in length. Using the magnification calculated in Q1, calculate the actual length of the onion cell in micrometers (μm).
- 3. List three key rules that must be followed when producing a biological drawing.
- 4. Why is it important to lower the coverslip slowly and at an angle?

Answers (1.6)

- 1. Total Magnification = $10 \times 40 = x400$.
- Image size (I) = 60 mm = 60,000 μm. Magnification (M) = 400. Actual size (A) = I / M = 60,000 μm / 400 = 150 μm.
- 3. Use sharp pencil; Draw large and clear; Use solid continuous lines; Label lines drawn with ruler, pointing accurately; Include title and magnification. (Any three)
- 4. To prevent trapping air bubbles.

1.7 Enzyme Mechanism: Active Site & Specificity

Revision Notes

Enzymes: Proteins acting as biological catalysts, speeding up reactions by lowering activation energy without being used up.

- Active Site: Specific 3D region on the enzyme where the substrate binds. Its shape is determined by the enzyme's tertiary structure.
- **Substrate:** Molecule(s) the enzyme acts upon.
- **Specificity:** Enzymes are highly specific; the active site shape is complementary to only one or a few specific substrates.
- Mechanism:

- 1. Substrate collides and binds to the active site.
- 2. Forms an enzyme-substrate complex.
- 3. Enzyme catalyses conversion of substrate to product(s).
- 4. Product(s) detach from the active site.
- 5. Enzyme is regenerated.
- Models:
 - Lock and Key (Old): Rigid enzyme and substrate fit perfectly.
 - **Induced Fit (Accepted):** Active site is flexible, changes shape slightly upon substrate binding for a better fit, optimising catalysis.

Practice Questions (1.7)

- 1. What is the role of the active site in enzyme function?
- 2. Explain what is meant by enzyme specificity, referring to the active site and substrate.
- 3. Briefly describe the 'lock and key' model of enzyme action.
- 4. How does the 'induced fit' model differ from the 'lock and key' model?

Answers (1.7)

- 1. It's the specific region where the substrate binds and the reaction is catalysed.
- 2. Each enzyme acts on a specific substrate because the active site shape is complementary only to that substrate's shape.
- 3. The active site (lock) and substrate (key) have rigid, perfectly complementary shapes.
- 4. Induced fit suggests the active site is flexible and changes shape slightly on binding, unlike the rigid shapes in the lock and key model.

1.8 Enzyme Denaturation

Revision Notes

Denaturation: A change in the enzyme's specific 3D shape, especially the active site, leading to loss of function. Often irreversible.

- Causes: Extreme conditions, mainly high temperatures or non-optimal pH values.
- **Mechanism:** These conditions break the weak bonds (e.g., hydrogen, ionic) maintaining the enzyme's tertiary structure.
- **Effect:** The active site shape changes, so the substrate can no longer bind effectively. The enzyme loses its catalytic activity.

Practice Questions (1.8)

- 1. Define denaturation in the context of enzymes.
- 2. Explain how high temperatures cause enzyme denaturation.
- 3. How does a change in pH away from the optimum lead to denaturation?
- 4. What is the direct consequence of the active site changing shape during denaturation on enzyme function?

Answers (1.8)

- 1. Loss of the enzyme's specific 3D shape (especially active site) due to extreme conditions (heat/pH), causing loss of function.
- 2. Increased vibrations break weak bonds holding the enzyme's structure.
- 3. Alters charges on amino acids, disrupting ionic/hydrogen bonds maintaining enzyme shape.
- 4. The substrate cannot bind to the altered active site, so the enzyme cannot catalyse the reaction.

<u>1.9 Effect of Temperature, Substrate Concentration & pH on</u> Enzyme Activity

Revision Notes

- Temperature:
 - Low temp: Slow rate (low kinetic energy, fewer collisions).
 - **Increasing temp:** Rate increases (more kinetic energy, more collisions) up to the optimum.
 - **Optimum temp:** Maximum activity (e.g., ~37°C for human enzymes).
 - **High temp:** Rate decreases sharply due to denaturation (active site changes shape).
 - **Graph:** Rises to peak (optimum), then falls steeply.
- pH:
 - **Optimum pH:** Maximum activity (varies by enzyme, e.g., pH 2 for pepsin, pH 8 for trypsin).
 - **Extreme pH (too high or low):** Rate decreases due to denaturation (active site changes shape).
 - **Graph:** Bell-shaped curve, peaking at optimum pH.
- Substrate Concentration: (Constant enzyme conc.)
 - **Low conc:** Rate increases as substrate conc. increases (more collisions). Substrate is limiting factor.
 - **High conc:** Rate plateaus. Active sites become saturated. Enzyme concentration is limiting factor.
 - **Graph:** Increases steeply, then levels off (plateau).

Practice Questions (1.9)

- Describe what happens to the rate of an enzyme-catalysed reaction as temperature increases from 0°C to 60°C, assuming the optimum temperature is 37°C. Explain why.
- 2. Explain why the graph of enzyme activity versus pH is typically bell-shaped.
- 3. What happens to the rate of reaction when substrate concentration is increased from low to very high levels, assuming enzyme concentration is constant? Explain

the shape of the graph.

4. Identify the limiting factor for the reaction rate at point A (low substrate conc.) and point B (high substrate conc./plateau) on a typical graph of reaction rate vs. substrate concentration.

Answers (1.9)

- 1. Rate increases up to 37°C (more kinetic energy/collisions), then decreases sharply as enzyme denatures above optimum.
- 2. Activity peaks at optimum pH; activity decreases on either side as non-optimal pH causes denaturation (active site change).
- 3. Rate increases initially as more substrate leads to more collisions. Rate plateaus when all active sites are saturated (enzyme concentration becomes limiting).
- 4. Point A: Substrate concentration. Point B: Enzyme concentration (active site availability).

1.10 Core Practical: Effect of pH on Enzyme Activity

Revision Notes

Objective: Investigate how pH affects amylase activity.

Principle: Amylase breaks down starch. Iodine tests for starch (blue-black = present; orange-brown = absent). Time how long it takes for starch to disappear at different pHs. Method Outline:

- 1. Set up spotting tile with iodine drops.
- 2. Prepare tubes with amylase + buffer solution at different pH values. Keep a separate tube of starch solution.
- 3. Control temperature (water bath).
- 4. Mix starch with one amylase/buffer tube, start timer.
- 5. Sample mixture into iodine wells every 10-30 secs.
- 6. Record time when iodine stays orange-brown (starch gone).
- 7. Repeat for all pH values. Variables:
- Independent: pH (using buffers).
- **Dependent:** Time taken for starch digestion (or Rate = 1/time).

• Control: Temperature, enzyme concentration, starch concentration, volumes. Results: Fastest rate (shortest time) at optimum pH. Slower rates at non-optimal pH due to denaturation. Graph of Rate vs. pH is bell-shaped.

Aspect	Details	
Enzyme Used	Amylase	
Substrate	Starch solution	
Independent Variable	pH (using buffer solutions)	
Dependent Variable	Time taken for starch to disappear (or Rate = 1/time)	
Method of Measurement	Timing how long it takes for samples to stop turning iodine blue-black	
Key Control Variables	Temperature, enzyme concentration, starch concentration, volumes	
Expected Graph Shape	Rate vs. pH: Bell-shaped curve	

Summary of Core Practical (pH and Amylase Activity)

Practice Questions (1.10)

- 1. In the core practical investigating the effect of pH on amylase activity, what is the independent variable and how is it controlled?
- 2. What is the dependent variable, and how is it typically measured in this experiment using iodine?
- 3. List two important control variables that must be kept constant during this investigation.
- 4. Describe the expected results if amylase has an optimum pH of 7 and is tested at pH 4, pH 7, and pH 10.

Answers (1.10)

- 1. Independent variable: pH. Controlled using buffer solutions.
- 2. Dependent variable: Time taken for starch breakdown (or rate). Measured by timing until samples no longer turn iodine blue-black.
- 3. Temperature; enzyme concentration; substrate concentration; volumes. (Any two)
- 4. Fastest reaction at pH 7 (optimum). Much slower reactions at pH 4 and pH 10 due to denaturation.

1.11 Rate Calculations for Enzyme Activity

Revision Notes

Rate quantifies how fast an enzyme works.

- Formula 1: Rate=Time takenChange in quantity (substrate used or product formed)
 - Units: e.g., g/min, cm³/s, mol/min.
- Formula 2: Rate=Time taken1 (Used when measuring time to a fixed endpoint).
 - \circ Units: e.g., s–1, min–1.
- From Graphs: (Quantity vs. Time)
 - \circ Rate = Gradient (slope) of the line (Change in Y / Change in X).
 - Initial rate = Gradient of tangent at time = 0 (steepest part).

Practice Questions (1.11)

- 1. An enzyme reaction produces 12 g of product in 3 minutes. Calculate the rate of reaction in g/min.
- In an experiment investigating the effect of temperature on catalase activity, it took 25 seconds for a certain amount of oxygen to be produced at 30°C. Calculate the rate of reaction in s–1.
- 3. A graph shows the volume of gas produced (cm³) against time (s). How would you calculate the rate of reaction at 10 seconds?
- 4. Why is the initial rate of reaction often measured in enzyme experiments?

Answers (1.11)

- 1. Rate = 12 g / 3 min = 4 g/min.
- 2. Rate = 1 / 25 s = 0.04 s-1.
- 3. Draw a tangent to the curve at 10 seconds and calculate its gradient (Change in volume / Change in time).
- 4. Initial rate reflects maximum speed under those conditions before substrate concentration drops or product inhibits the reaction. Allows valid comparison when changing other factors.

<u>1.12 Importance of Enzymes in Synthesis & Breakdown</u></u>

Revision Notes

Enzymes control metabolism (both breakdown and synthesis).

- Breakdown (Catabolism/Digestion): Breaking large, insoluble molecules into small, soluble ones.
 - **Carbohydrates:** Broken down by **carbohydrases** (e.g., amylase: starch \rightarrow maltose; maltase: maltose \rightarrow glucose).
 - **Proteins:** Broken down by **proteases** (e.g., pepsin, trypsin) into amino acids.
 - Lipids: Broken down by lipases into fatty acids and glycerol.
- Synthesis (Anabolism): Building complex molecules from simpler units.
 - **Carbohydrates:** Simple sugars joined to form polysaccharides (e.g., glycogen synthase: glucose \rightarrow glycogen).
 - Proteins: Amino acids joined to form proteins (protein synthesis).
 - Lipids: Fatty acids and glycerol joined to form lipids.
- **Importance:** Enzymes allow these essential reactions to happen quickly enough at body temperature and pH, enabling energy release, nutrient absorption, growth, and repair.

Key Digestive Enzymes Summary

Enzyme Type	Specific Example	Substrate	Product(s)
Carbohydrase	Amylase	Starch	Maltose
Carbohydrase	Maltase	Maltose	Glucose
Protease	Pepsin	Proteins	Polypeptides
Protease	Trypsin	Polypeptides	Amino Acids
Lipase	Lipase	Lipids (Fats)	Fatty Acids & Glycerol

Practice Questions (1.12)

- 1. State the role of enzymes as biological catalysts in both synthesis and breakdown reactions.
- 2. Name the type of enzyme that breaks down proteins and state the smaller molecules produced.
- 3. Give an example of a specific enzyme involved in carbohydrate digestion and state its substrate and product.
- 4. Explain why enzymes are essential for the synthesis of glycogen from glucose.

Answers (1.12)

- 1. Enzymes speed up both breakdown (catabolic) and synthesis (anabolic) reactions without being used up.
- 2. Proteases break down proteins into amino acids.
- 3. Amylase breaks down starch into maltose. OR Maltase breaks down maltose into glucose.
- 4. Enzymes (like glycogen synthase) catalyse the joining reactions, lowering activation energy so synthesis occurs fast enough at body conditions for energy storage.