B.2.2 · Organelles and compartmentalization

International Baccalaureate · IB Diploma · Biology · HL

Scope and prerequisites

A pancreatic cell makes enzymes for release, while a contracting muscle cell needs a continuing supply of ATP. Both are cells, but a micrograph shows different amounts of internal membrane and different organelle arrangements. How can dividing the inside of a cell into regions help it perform a particular job?

This unit covers B2.2 SL understandings plus additional higher-level content (marked AHL): organelle identity, cell fractionation, nuclear separation, local reaction conditions, mitochondrial and chloroplast adaptations, nuclear envelopes, ribosomes, rough ER, Golgi and vesicles. Prerequisites are cell structure, protein function and membrane transport. All numerical datasets are invented teaching examples. Sections marked **AHL** are for Higher Level only.

Identify organelles by structure and function

An **organelle** is a discrete cell subunit adapted to a particular function. The term does not mean only a membrane-bounded sac. In the convention used for this course, nuclei, vesicles, ribosomes and the plasma membrane count as organelles; the cell wall, cytoskeleton and cytoplasm do not. Ribosomes are not surrounded by a membrane. State the convention rather than classifying every useful cell component as an organelle.

Component	Structural evidence	Functional connection
Nucleus	Envelope enclosing chromatin; pores	Separates nuclear activities from the surrounding cytoplasm
Ribosome	Small particle with two subunits; no enclosing membrane	Joins amino-acid residues during translation
Mitochondrion	Two membranes; folded inner membrane	Supports aerobic respiration and ATP production
Chloroplast	Envelope and internal thylakoid membranes	Supports photosynthesis in suitable plant/algal cells
Lysosome	Membrane-bounded digestive compartment	Provides conditions for breakdown and recycling
Vesicle	Small membrane-enclosed compartment	Carries or stores selected contents

A cell's organelle content depends on its type and state. Some differentiated cells lose particular structures: mature mammalian red blood cells lack nuclei and mitochondria. A secretory cell can have extensive rough ER and Golgi; an active muscle cell can have many mitochondria. These are structure–function relationships, not a rule that one named organelle is the only important structure in that cell.

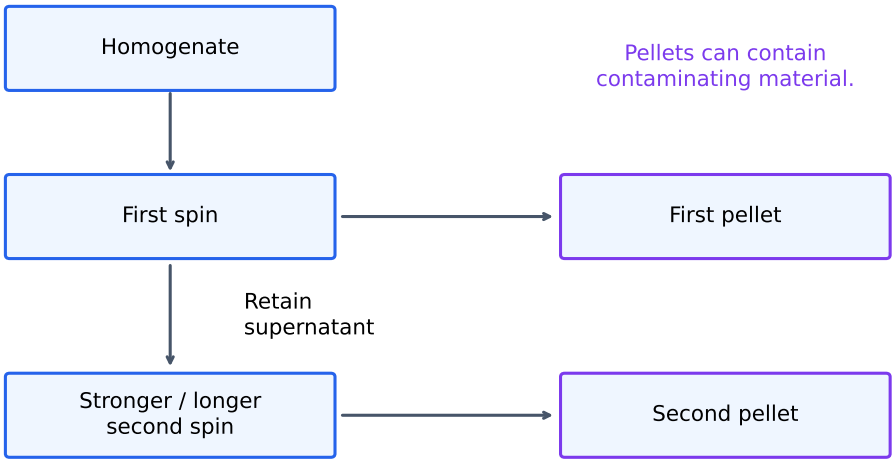
A schematic uses colour and simplified shapes to explain organization. An electron micrograph gives evidence from a specimen and preparation, usually without the diagram's identifying colours. Look for membranes, folds, stacked sacs and location. One circular profile could be a section through a tube; one image does not reveal a complete three-dimensional shape or prove an organelle's activity.

Cell fractionation makes organelle functions testable

A microscope can locate structures, but studying a separated fraction allows different questions. Does the fraction containing a particular organelle also contain a particular enzyme activity? The development of high-speed centrifuges helped researchers test such relationships. A new technique can make a previously inaccessible process measurable.

First homogenize tissue to break cell boundaries and release contents. Use a cold, suitable buffered medium: cooling slows unwanted reactions, buffering limits pH changes, and an appropriate osmotic composition helps preserve organelles. These measures reduce damage; they do not guarantee that every structure survives. Avoid detergent when intact lipid membranes are needed, because detergent can disrupt them.

Centrifuge the homogenate. Material that sediments forms a pellet; the liquid above it is the supernatant. Remove the supernatant and spin it under stronger or longer conditions to collect another fraction. Larger or more readily sedimenting structures can be collected before smaller ones. Sedimentation also depends on density, shape, medium and time, so size alone does not produce an exact universal order.



Further separation can use the second supernatant.

Carry the supernatant forward; do not spin the discarded pellet and call it the same sequence. Labels describe an experimental separation, not perfect purification.

Test fractions with microscopy and suitable marker assays. A nuclear marker should be enriched with nuclei; an enzyme associated with mitochondria should be enriched in a mitochondrial fraction. Broken organelles and trapped material can contaminate other fractions. Co-occurrence supports an association, but does not establish that a fraction is pure or that a single enzyme explains every function.

Worked example: distinguish recovery from enrichment

Invented recoveries, expressed as percentages of each marker's starting total, are:

Fraction	Nuclear DNA marker / %	Mitochondrial marker / %	Cytosolic enzyme / %
First pellet	80	2	5
Second pellet	10	82	5
Small-membrane pellet	5	10	10
Final supernatant	5	6	80

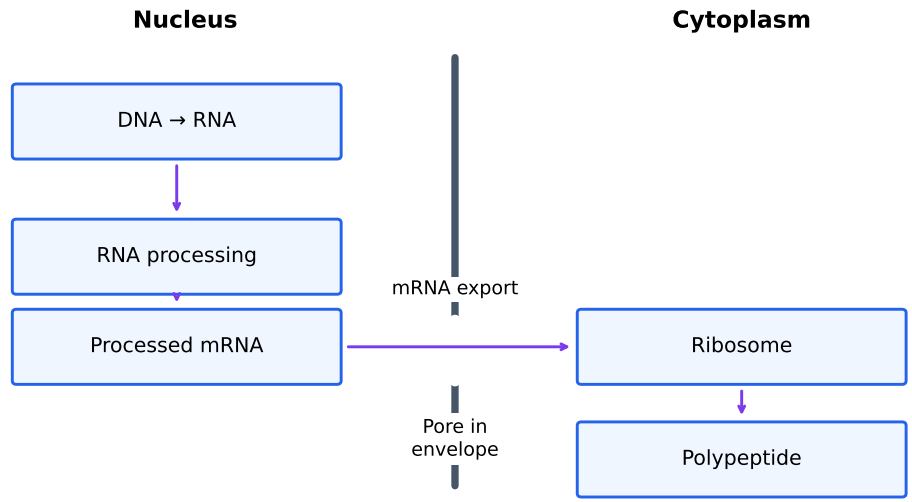
Each marker column totals 100%. The second pellet recovers most of the mitochondrial marker, but also contains some nuclear marker and cytosolic enzyme. It is enriched in mitochondrial material; it is not proved to contain only mitochondria. The final supernatant can also contain small particles not sedimented by the stated procedure.

In a separate invented assay, a starting sample has 30 enzyme units in 15 mg protein: specific activity $= 30/15 = 2$ units per mg. A collected fraction has 20 units in 2 mg: $20/2 = 10$ units per mg. Enrichment is $10/2 = 5$-fold, although recovery of enzyme activity is only $20/30 \times 100 = 66.7\%$. A concentrated, enriched fraction need not contain most of the starting material. Assay conditions must be comparable before activities can be divided meaningfully.

Nuclear separation allows RNA processing before translation

Compartmentalization creates regions with different contents or conditions. In a eukaryotic cell, the nuclear envelope separates the main DNA-containing compartment from the cytoplasm. Transcription produces RNA from a DNA template in the nucleus. Translation makes a polypeptide from an mRNA sequence at ribosomes outside the nucleus.

For a typical eukaryotic protein-coding gene, the first RNA transcript can be processed before export. Processing includes removing introns and joining the retained exons by splicing; a cap and poly(A) tail are commonly added. The processed mRNA passes through nuclear pores and can then meet cytoplasmic ribosomes. This separation provides an opportunity for processing and checking before translation. DNA instructions remain in the nucleus; the cell exports an RNA copy, not the gene itself.



The arrow across the envelope carries processed mRNA. The polypeptide is made outside the nucleus. The drawing is a pathway schematic, not a micrograph.

Prokaryotes have no nucleus enclosing their chromosome. Ribosomes may begin translating an mRNA while that RNA is still being transcribed. Do not say prokaryotes have no DNA or no ribosomes, or that all RNA processing is absent from them. The key comparison here is the absence of a nuclear boundary separating transcription

from translation.

A simple timing model illustrates the distinction. Suppose a eukaryotic transcript is made from minute 0 to 3, processing finishes at minute 5, and export finishes at minute 6. In this model cytoplasmic translation cannot start at minute 2: the processed RNA has not arrived. The numbers are invented and do not specify universal cellular rates. They describe the order of events for the stated transcript, not every RNA in every organism.

Compartments concentrate reactants and separate incompatible conditions

An enzyme and its substrate meet more readily when both are concentrated in a suitable region. Keeping them in a smaller compartment can increase their local concentrations without increasing their total amounts. The result still depends on transport, enzyme properties and other requirements; concentration alone does not guarantee the fastest possible reaction.

For an invented example, 12 pmol of a metabolite in 4 pL gives $12/4 = 3$ pmol per pL. The same amount in 12 pL gives 1 pmol per pL: a threefold concentration difference. This is a ratio model, not a realistic concentration claim for a named metabolite. It does not show that the larger compartment has fewer total molecules.

Membranes help preserve different conditions while allowing controlled exchange. Lysosomes maintain an acidic interior suitable for their digestive enzymes. The cytosol has different conditions, so the two environments can support different reactions in the same cell. A lysosomal membrane helps contain digestive activity; it is a selective boundary, not an absolute shield against every form of damage.

A phagocytic cell encloses an engulfed particle in a phagocytic vacuole, also called a phagosome. Fusion with lysosomal compartments supplies digestive enzymes and supports breakdown in the enclosed region. The material does not first have to disperse freely through the cytosol. Useful breakdown products can subsequently be transported out of that compartment. Explain both containment and controlled transfer, rather than describing an isolated bag with no exchange.

This organization has costs. Membranes must be made, maintained and supplied with transport proteins; sorting and transport can require energy. A useful explanation connects a boundary to the conditions it maintains and the process it enables. Simply naming a compartment does not establish its advantage.

Measure structures and report the limits of an image

For a prepared light-microscope slide, locate the specimen at low power first. Centre the region, increase the objective power as appropriate and use fine focus at high power. Draw clear outlines with straight label lines and record the scale. Do not add shading or structures that cannot be seen. Increased magnification does not automatically improve resolution.

Magnification is image length divided by actual length, with both lengths in the same unit. A scale bar supplies a known real distance within that image; a captioned magnification becomes wrong if an image is resized without updating it, while a scale bar resized with the image remains usable.

> ■ **Worked example: use a plausible organelle scale**
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> An invented mitochondrion image is 30 mm long; its actual length is 5.0 μm. Convert 30 mm = 30000 μm, then $M = 30000/5.0 = 6000$. Magnification is a dimensionless ratio, written $\times$6000. These numbers do not assert that all mitochondria have that size.
>
> On the same image, a vesicle measures 4.0 mm. Its real diameter is $4000/6000 = 0.667$ μm, about 0.67 μm. An image of a 2.0 μm structure at $\times$15000 is $2.0 \times 15000 = 30000$ μm, or 30 mm. A separate structure shown as 24 mm for an actual 120 μm has magnification $24000/120 = 200$; the arithmetic does not establish that this larger structure is a typical organelle.
>
> If a scale bar labelled 2.0 μm measures 8.0 mm on a print, a structure measuring 18 mm on that same print has actual length $(18/8.0)(2.0) = 4.5$ μm. Doubling both printed lengths leaves the ratio unchanged. Record the measured structure and method; a static micrograph does not directly show a vesicle moving or a mitochondrion making ATP.

AHL: Mitochondrial compartments support aerobic ATP production

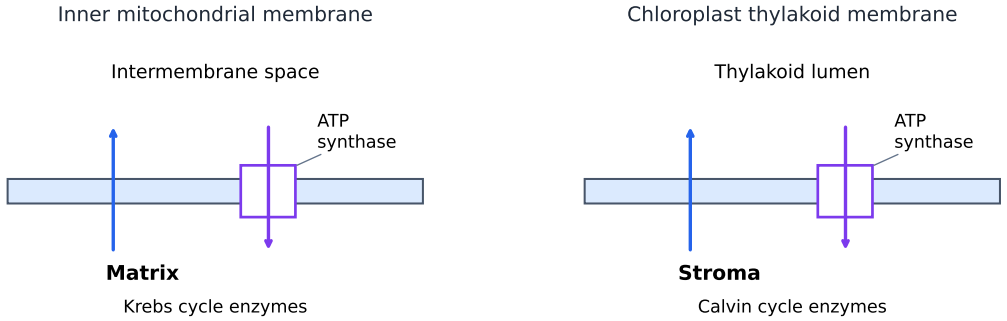
The outer and inner membranes enclose an intermembrane space and a matrix. Cristae are folds of the inner membrane. Their large area accommodates respiratory electron-transfer components and ATP synthase. The matrix contains enzymes and substrates used in the Krebs cycle, bringing related reactions into a suitable local environment.

Energy from electron transfer is used to move hydrogen ions from the matrix to the intermembrane space. The relatively small volume of that space allows a given transfer to produce a larger concentration change than it would in a much larger compartment. The inner membrane restricts uncontrolled ion movement, so a gradient can be maintained. Return of hydrogen ions through ATP synthase couples the stored electrochemical difference to ATP production.

Folds increase membrane area; they do not create energy or replace the need for substrates, oxygen in aerobic electron transfer and working proteins. Damaging the inner membrane can dissipate the gradient even if the cell still contains many mitochondria. ATP yield cannot be predicted from a count of cristae alone.

AHL: Chloroplast compartments support photosynthesis

A chloroplast has an envelope and internal thylakoids, often arranged in stacks called grana. Thylakoid membranes provide a large area for photosystems and other components of the light-dependent reactions. Light energy supports electron transfer and the accumulation of hydrogen ions in the thylakoid lumen. Its small volume helps a given ion transfer change local concentration.



These are local membrane sections, not whole organelles. Identify the two aqueous compartments before following the arrows.

Hydrogen-ion return through ATP synthase into the stroma supports ATP production. The stroma contains enzymes and substrates of the Calvin cycle, which incorporates carbon from carbon dioxide into organic compounds. It is not the thylakoid lumen, and photosystems are not free-floating in it. Carbon fixation and the light-dependent reactions are connected processes in different locations.

Compare functions precisely: mitochondrial ATP synthesis involves the inner membrane, whereas chloroplast light-dependent ATP synthesis involves the thylakoid membrane. A chloroplast's envelope is not a substitute for its thylakoid system. These arrangements illustrate how membranes provide both a surface for proteins and a boundary between regions.

AHL: Nuclear envelopes combine separation with controlled traffic

The nuclear envelope has two membranes with a space between them. Its outer membrane is continuous with the endoplasmic reticulum. Nuclear pore complexes provide controlled routes across the envelope, allowing appropriate RNAs and proteins to move between nucleus and cytoplasm. The boundary separates processes without sealing the DNA compartment away from every required molecule.

An mRNA can leave through a pore; a protein needed for DNA replication can enter by a regulated import process. It is not sufficient to say that all small molecules enter and every large molecule is excluded. Pore complexes, recognition and transport machinery contribute selectivity.

During open mitosis or meiosis, as in typical animal-cell examples, the envelope disassembles, permitting spindle access to chromosomes. Nuclear membrane material forms fragments/vesicles and reorganizes with the ER; envelopes later re-form around daughter chromosome sets. This is membrane rearrangement, not destruction of the genetic instructions. Some organisms retain a nuclear boundary during division, so envelope breakdown is not universal. Cell-biology descriptions of nuclear traffic explain why pores and envelope dynamics solve different access problems.

AHL: Ribosomes and rough ER direct proteins toward different destinations

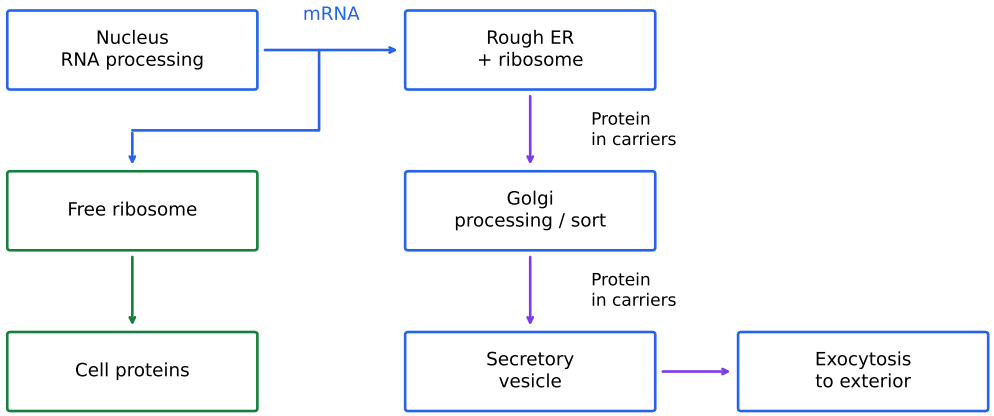
A ribosome contains ribosomal RNA and proteins arranged in two subunits. Free cytosolic and rough-ER-bound ribosomes use the same basic translation mechanism; they are not permanently different species of ribosome. Targeting information in a growing protein can lead a translating ribosome to the ER membrane.

Free ribosomes make many proteins used in the cytosol and proteins subsequently imported into the nucleus, mitochondria or chloroplasts. They do not make only proteins that remain forever in the cytosol. Many proteins entering the secretory pathway are made at ribosomes attached to the cytosolic face of rough ER. The ribosome is outside the ER lumen; the growing protein can be transferred into the lumen or inserted into the membrane.

The rough ER is a membrane system, not a pile of ribosomes. It supports entry, folding and early processing of appropriate proteins. Membrane-bound ribosomes make proteins for secretion and for destinations within the cell, such as parts of the endomembrane system. Not every protein made on rough ER leaves the cell, and not every protein used inside the cell is made on rough ER.

AHL: Golgi and vesicles process, sort and deliver proteins

A protein entering the secretory route passes from ER toward the Golgi in transport carriers. The Golgi contains flattened membrane sacs with receiving and shipping sides. It modifies and sorts appropriate proteins, then packages cargo into carriers for particular destinations. It does not translate an mRNA or synthesize a protein's amino-acid sequence from nothing.



Blue: RNA route. Purple: secretory protein route. Green: other protein products.

RNA export and protein transport are different arrows. The diagram does not send an already completed secreted protein out of the nucleus.

Vesicle membranes keep cargo separate from the cytosol during transport. Budding, movement, recognition and fusion deliver cargo to a suitable destination; random fusion with every membrane would undermine compartmentalization. Fusion during exocytosis joins the vesicle membrane to the plasma membrane and releases soluble contents outside.

Clathrin is a coat protein that helps form some vesicles, including appropriate endocytic and Golgi-associated carriers. It assembles on the cytosolic side, helping shape a bud and, through associated proteins, organize selected cargo. The coat is removed before later fusion steps. Clathrin is neither the lipid membrane nor a permanent coat on every vesicle; some transport carriers use other coat systems. The intracellular compartment account connects protein sorting with the maintenance of distinct destinations.

Practice

All numerical data are invented. Use a named structure, mechanism and limitation in each explanation.

1. Classify nucleus, ribosome, plasma membrane, cell wall, cytoskeleton and cytoplasm using this course's organelle convention. Explain why a membrane is not required around every organelle.
2. Predict two structural differences between an enzyme-secreting pancreatic cell and a mature mammalian red blood cell. Explain why one organelle count cannot establish a cell's complete activity.
3. Outline cell fractionation from homogenization to successive pellets. Explain the cold buffered medium and why supernatant, rather than the first pellet, proceeds to the next separation.
4. From the marker table, identify the fraction most associated with mitochondria and calculate how much mitochondrial marker remains elsewhere. Explain two reasons it is not a pure mitochondrial sample.
5. A starting assay has 30 enzyme units in 15 mg protein; a fraction has 20 units in 2 mg. Calculate both specific activities, fold enrichment and percentage recovery. Distinguish enrichment from total recovery.
6. Explain how nuclear separation permits RNA processing before translation. Compare with a prokaryote and identify the molecule exported for translation.
7. In the timing model, transcription ends at minute 3, processing at 5 and export at 6. Could cytoplasmic translation of this transcript start at minute 2? Explain without claiming universal timings.
8. Calculate concentrations for 12 pmol in 4 pL and in 12 pL. Explain an advantage and a cost of maintaining different reaction compartments.
9. Explain lysosome and phagocytic-vacuole functions. Distinguish containment from a complete absence of transport across their membranes.
10. Calculate magnification for a 30 mm image of a 5.0 μm mitochondrion, actual vesicle diameter from a 4.0 mm image at that magnification, and image length for a 2.0 μm structure at $\times$15000. Explain low-power focusing and clear scale-labelled drawing.
11. A 2.0 μm scale bar prints as 8.0 mm and a structure as 18 mm. Find actual size. Predict the result after both printed lengths double and explain two limits of a static micrograph.
12. **AHL** Compare mitochondrial and chloroplast ATP-producing membrane systems. Include membrane area, the small receiving compartment for hydrogen ions, enzyme locations, and one consequence of membrane damage.
13. **AHL** Explain nuclear-envelope pores and division-related disassembly. Contrast free and rough-ER-bound ribosomes, including two intracellular destinations for free-ribosome products and one rough-ER product retained inside the cell.
14. **AHL** Trace a secreted protein from its mRNA through translation, ER, Golgi, vesicles and exocytosis. Explain clathrin's role and why a static image of many vesicles does not alone prove a higher secretion rate.

■ Worked answers

1. Nucleus, ribosome and plasma membrane count as organelles in this course convention; cell wall, cytoskeleton and cytoplasm do not. Ribosomes are discrete functional subunits without an enclosing membrane. The classification does not imply excluded structures are unimportant.
2. The pancreatic cell can contain a nucleus, rough ER and Golgi for protein pro-

duction and processing; the mature mammalian red blood cell lacks these. Its organelle content reflects specialization. Counts alone do not measure protein activity, flux or ATP production; compare cell size, state and suitable functional measurements.

3. Homogenize in cold, appropriate buffered/osmotic medium, then centrifuge and retain the supernatant for stronger/longer spins. Cooling slows unwanted reactions; buffering and suitable osmotic conditions help preserve structures. The first pellet contains already sedimented material. Fractions differ in contents but can be contaminated or damaged.
4. The second pellet contains 82% of the mitochondrial marker. Elsewhere there is $2+10+6=18\%$, also $100-82=18\%$. Other markers occur in the second pellet, and broken/trapped material can contaminate it. A marker assay and microscopy support enrichment, not perfect purity.
5. Starting specific activity $= 30/15 = 2$ units per mg; fraction $= 20/2 = 10$ units per mg. Enrichment $= 10/2 = 5$-fold. Recovery $= 20/30 \times 100 = 66.7\%$. Activity per protein mass increased while about one-third of starting enzyme activity was not recovered in this fraction.
6. Transcription and RNA processing occur in the nuclear compartment before processed mRNA passes through pores to cytoplasmic ribosomes. Introns can be removed before translation. In prokaryotes, lack of a nuclear envelope permits translation to begin on RNA still being transcribed. mRNA, rather than the DNA gene or a completed secreted protein, is exported for translation.
7. No: at minute 2 this transcript has not finished processing or export. Under the model, cytoplasmic translation can start only after mRNA arrival at minute 6. Different real genes and conditions have different timings; the example establishes sequence, not a universal six-minute delay.
8. $12/4 = 3$ pmol per pL and $12/12 = 1$ pmol per pL, a threefold concentration ratio with the same total amount. Concentrating enzymes and substrates can support encounters, and separating incompatible conditions can support different reactions. Making and maintaining membranes and controlled transport require resources.
9. A lysosome supplies an acidic digestive environment. An engulfed particle lies in a phagocytic vacuole; fusion with lysosomal compartments supplies digestive machinery. Contents remain separated from cytosol during digestion, but useful products and other substances can cross through suitable transport pathways. The membranes are selective boundaries, not permanently sealed walls.
10. $30000/5.0 = 6000$, so $\times 6000$. Vesicle diameter $= 4000/6000 = 0.667$ μm, approximately 0.67 μm. The 2.0 μm structure gives $2.0(15000) = 30000$ μm $= 30$ mm. Locate and centre at low power; use fine focus at high power. Draw observed outlines, straight labels and a scale; magnification alone does not improve resolution.
11. $(18/8.0)(2.0) = 4.5$ μm. After resizing, $(36/16)(2.0) = 4.5$ μm. Section orientation and preparation affect apparent structure; a static image does not directly measure movement or activity. A scale bar must belong to that image and be resized with it.
12. **AHL** Cristae increase mitochondrial inner-membrane area; hydrogen ions accumulate in the small intermembrane space and return toward matrix through ATP synthase. Matrix enzymes support the Krebs cycle. Thylakoids provide photosystem area; hydrogen ions accumulate in the small lumen and return toward stroma through ATP synthase. Stroma enzymes support the Calvin cycle. Damage allowing uncontrolled ion leakage can dissipate gradients; membrane area alone cannot replace working components and an energy source.
13. **AHL** Nuclear pores permit controlled molecular exchange while the double membrane separates processes. During open division the envelope disassembles into rearranging membrane material/fragments/vesicles and later reforms; the DNA is not destroyed. Free ribosomes can supply cytosolic proteins and proteins imported into nucleus or mitochondria/chloroplasts. Rough-ER-bound ribosomes transfer appropriate growing chains into ER or membrane; a lysosomal enzyme is an intracellular example. Ribosomes attach at the cytosolic surface, and the two locations are not permanently different ribosome types.
14. **AHL** Export processed mRNA through a pore; a ribosome translates it and an appropriate targeting signal directs synthesis at rough ER. The protein enters ER, then transport carriers deliver it toward Golgi for modification/sorting. Secretory vesicles deliver selected cargo to plasma membrane; fusion releases contents by exocytosis. Clathrin helps shape some buds and organize cargo through associated proteins, then is removed; it is not every vesicle's permanent membrane. Many visible vesicles could reflect rapid production or blocked onward movement. Use timed labelling or secretion measurements to test flux.

Exam tips

Connect a named structure to a mechanism and function. Distinguish recovery from enrichment, local concentration from total amount, and an RNA route from a protein route. Convert both lengths to one unit and use a scale bar from the same image. Identify what a static specimen can and cannot establish.

- **AHL:** Name both sides of each ATP-producing membrane and the destination of each protein pathway.

Scope reference: IB Biology guide, B2.2. Numerical examples are invented.