Organelles and compartmentalization

B2.2 · SL

Why do a secretory cell and a muscle cell look different?

Cell contents support different demands

A pancreatic cell exports enzymes; a contracting muscle cell needs continuing ATP supply. Different internal arrangements support these different roles.

Common content: organelle classification, fractionation, nuclear separation, local reaction conditions and microscopy evidence.

Numerical datasets are invented teaching examples.

Use the course convention when classifying structures

Component	Classification and reason
Nucleus and vesicle	Organelles with enclosing membranes
Ribosome	Organelle without an enclosing membrane
Plasma membrane	Counts as an organelle in this course convention
Cell wall, cytoskeleton, cytoplasm	Do not count as organelles under this convention

Identify a structure before assigning activity to it

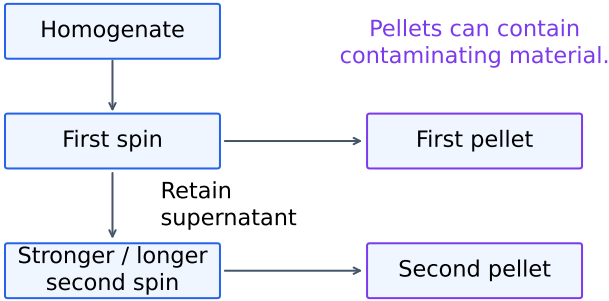
A micrograph supplies structural evidence

Look for membranes, folds, stacked sacs and location. A circular section may come from a tube; a schematic colour is not an identifying stain in every specimen.

Mature mammalian red blood cells lack nuclei and mitochondria. Secretory cells can have extensive rough ER and Golgi; active muscle cells can contain many mitochondria.

Organelle abundance supports a structure–function explanation but does not directly measure secretion rate or ATP production.

Follow the supernatant to the next spin



A pellet contains sedimented material; the next separation uses retained supernatant, and fractions can remain contaminated.

Preserve structures while separating them

Homogenize, centrifuge and test the resulting fractions

Break cell boundaries in a cold, suitable buffered/osmotic medium. Cooling slows unwanted reactions; the medium helps preserve pH and membrane-enclosed structures.

Collect an initial pellet and spin its supernatant under stronger or longer conditions. Sedimentation depends on density, shape, medium and time as well as size.

Use microscopy and marker assays to identify enrichment. High-speed centrifuges made functions of separated organelle fractions easier to investigate.

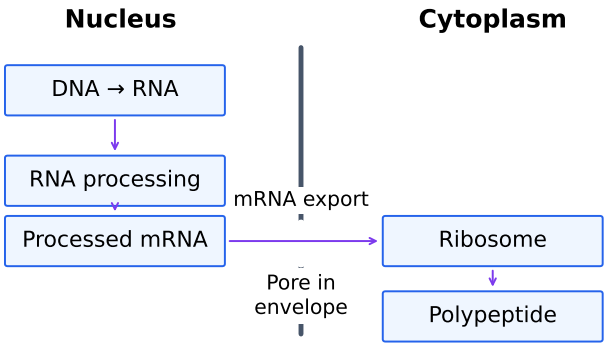
Read marker recoveries as an experimental distribution

Invented fraction	**Nuclear / mitochondrial / cytosolic marker recovery**
First pellet	80% / 2% / 5%
Second pellet	10% / 82% / 5%
Small-membrane pellet	5% / 10% / 10%
Final supernatant	5% / 6% / 80%

Separate enrichment from recovery

1. **Starting sample.** 30 enzyme units in 15 mg protein: specific activity $= 30/15 = 2$ units per mg.
2. **Collected fraction.** 20 units in 2 mg: $20/2 = 10$ units per mg. Enrichment $= 10/2 = 5$-fold.
3. **Recovery.** $20/30 \times 100 = 66.7\%$. Increased activity per protein mass does not mean all starting activity was recovered; compare the same assay conditions.

Export an RNA copy rather than the DNA gene



The nuclear envelope separates transcription and processing from cytoplasmic translation; processed mRNA crosses through pores.

A boundary allows processing before ribosomes meet mRNA

Compare eukaryotic separation with prokaryotic coupling

For a typical eukaryotic protein-coding transcript, splicing removes introns and joins retained exons. A cap and poly(A) tail are commonly added before export.

Ribosomes outside the nucleus translate processed mRNA. DNA remains in the nuclear compartment.

Prokaryotes lack this nuclear boundary, so translation may begin while transcription continues. They still have DNA and ribosomes; do not claim they lack all RNA processing.

Use the stated event order rather than a universal delay

1. **Invented model.** Transcription ends at minute 3; processing ends at 5; export ends at 6.
2. **Prediction.** Cytoplasmic translation of this transcript cannot start at minute 2. In this model its processed mRNA arrives only at minute 6.
3. **Limit.** These numbers establish the order for one stated transcript. They do not give universal times for all genes or organisms.

Connect a boundary to the conditions it maintains

Concentration and separation offer different advantages

Keeping enzymes and substrates together can support encounters without increasing total amounts. Transport and enzyme properties still matter.

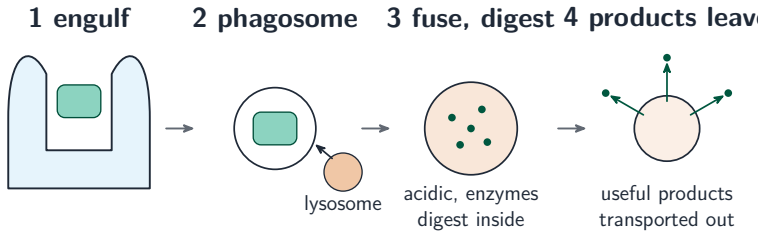
A lysosome supplies an acidic digestive environment while cytosol maintains different conditions. Its membrane helps contain digestive activity.

An engulfed particle remains within a phagocytic vacuole. Fusion with lysosomal compartments supports digestion; useful products can leave through suitable transport pathways.

Keep amount, volume and concentration separate

1. **Same amount.** Invented 12 pmol in 4 pL gives $12/4 = 3$ pmol per pL. In 12 pL it gives 1 pmol per pL.
2. **Compare.** The concentration is threefold greater in the smaller volume. Total amount remains 12 pmol in each case.
3. **Evaluate.** Compartments can separate incompatible conditions, but membranes, sorting and controlled transport require resources. This ratio model is not a measured concentration of a named metabolite.

A lysosome digests a bacterium



digestion stays inside a membrane compartment; the cytosol keeps its own conditions

> Acidic enzymes digest the bacterium safely

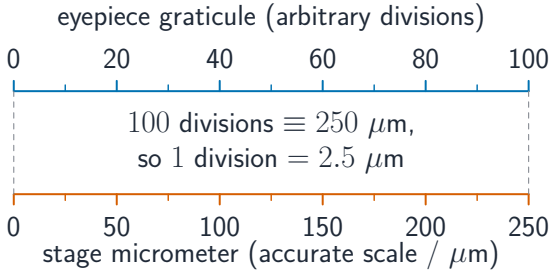
Convert lengths before finding magnification

1. **Mitochondrion.** A 30 mm image represents 5.0 μm. $M = 30000/5.0 = 6000$: magnification $\times 6000$, without a unit.
2. **Vesicle.** A 4.0 mm image at the same scale gives actual diameter $4000/6000 = 0.667$ μm, about 0.67 μm.
3. **Image length.** A 2.0 μm structure at $\times 15000$ gives 30000 μm $=$ 30 mm. All stated examples are invented.

A scale bar preserves the ratio when an image is resized

1. **Original print.** A 2.0 μm bar measures 8.0 mm. A structure measures 18 mm: actual size $= (18/8)(2) = 4.5$ μm.
2. **Doubled print.** Bar 16 mm and structure 36 mm: $(36/16)(2) = 4.5$ μm. Resize the scale bar with its image.
3. **Evidence.** Locate at low power first; centre before increasing power and use fine focus. Draw observed outlines with straight labels and scale. Magnification does not by itself increase resolution or reveal movement.

Calibrating an eyepiece graticule



Recalibrate for every objective lens

Common practice: fractions, nuclear separation and containment

1. **Questions 1–5.** Use the course organelle convention; explain specialization and fractionation. A mitochondrial marker is 82% in the second pellet, leaving 18% elsewhere.
2. **Enzyme calculations.** Specific activities 2 and 10 units per mg; fivefold enrichment; 66.7% recovery. A marker-enriched fraction is not automatically pure.
3. **Questions 6–9.** Export processed mRNA, not DNA. Apply the stated timing model and distinguish concentration from amount. Lysosomes and phagocytic vacuoles combine containment with controlled exchange.

Common practice: microscopy arithmetic and limitations

1. **Question 10.** Magnification $\times 6000$; vesicle about 0.67 μm; image of a 2.0 μm structure at $\times 15000$ is 30 mm.
2. **Question 11.** Scale-bar calculation gives 4.5 μm before and after proportional resizing.
3. **Method.** Begin at low power, use fine focus at high power, and draw observed structures with labels and scale. Section orientation, preparation and lack of time information limit conclusions.

Check yourself

Name the structure, mechanism, evidence and limitation

Distinguish recovery from enrichment and concentration from total amount. Use the same assay conditions and units.

A scale bar must belong to the same image and resize with it. A static micrograph does not directly measure a process rate.