Cell Biology

Unit 3 · Lesson 1 · Nucleus and ER

The notes for Lesson 1 of Unit 3: chapters 3.1 to 3.2 of the unit book, with their figures, worked examples, exam traps and quizzes, in one file. The full unit book, with every chapter, the exam question bank and the glossary, is linked at the end.

Lesson 1 of 4 | 2 chapters | Chapters 3.1 to 3.2

About these notes

These are the notes for **Lesson 1 · Nucleus and ER** of the University free n easy video lessons on Cell Biology Unit 3, Cell Organelles and Cytoskeleton. They are chapters 3.1 to 3.2 of the unit book, unchanged, so a page here is the same page in the full book.

3.1 Nucleus, nuclear envelope, nuclear pore and nucleolus

The nucleus is the room where a eukaryotic cell keeps its DNA. A double membrane, the nuclear envelope, separates it from the cytoplasm, so that copying DNA and making RNA happen apart from making protein. Traffic between the two sides passes through nuclear pores, and inside the nucleus a dense patch called the nucleolus builds ribosome subunits. This separation is what lets eukaryotes process RNA (cut, cap and tail it) before any ribosome can read it.

> You need: the prokaryote–eukaryote comparison from **Unit 1, Prokaryotic and eukaryotic cell organization**, and the lipid bilayer from **Unit 2, Plasma membrane structure**.

The nuclear envelope and lamina

The nuclear envelope is two lipid bilayers, an **outer nuclear membrane** and an **inner nuclear membrane**, with a thin gap between them called the **perinuclear space**. The outer membrane is continuous with the rough endoplasmic reticulum and carries ribosomes on its cytosolic face, so the perinuclear space is part of the ER lumen. The two membranes join only at the pores.

The inner membrane has its own proteins, which hold on to a mesh just beneath it: the **nuclear lamina**. The lamina is built from **lamins** (A, B and C), which are intermediate-filament proteins

(see Chapter 3.8). It gives the nucleus its shape and strength and anchors chromatin. When a cell enters mitosis, kinases add phosphate groups to the lamins, the mesh comes apart and the envelope breaks into vesicles; it reassembles around each daughter set of chromosomes (cell division itself is in **Unit 4, Mitosis**). Mutations in the lamin A gene cause laminopathies, including the premature-ageing disease progeria.

The nuclear pore complex

A **nuclear pore complex** (NPC) is a large protein channel where the inner and outer membranes fuse. It is made of about 30 different proteins called **nucleoporins**, each present in many copies, arranged with **eightfold symmetry** around a central channel. Some nucleoporins carry long, disordered chains rich in phenylalanine–glycine repeats (**FG repeats**) that fill the channel like a gel. Ions, sugars, nucleotides and small proteins diffuse through freely. Molecules larger than roughly 40–60 kDa are held back by the FG mesh unless a transport receptor carries them.

1. **Import.** A protein meant for the nucleus carries a **nuclear localization signal** (NLS), a short stretch of basic amino acids (lysine, arginine). The classic example is PKKKRKV from the SV40 virus large T antigen. The adaptor **importin α** binds the NLS, and **importin β** binds importin α and slides through the FG mesh.
2. **Release.** Inside the nucleus, the small GTPase **Ran** in its GTP-bound form binds importin β, and the cargo is let go.
3. **Recycling.** The importin–Ran-GTP complex goes back out. In the cytosol a GTPase-activating protein (RanGAP) makes Ran hydrolyse its GTP, and importin is freed for another round.
4. **Export.** Proteins with a **nuclear export signal** (leucine-rich) bind an exportin together with Ran-GTP inside the nucleus and are released in the cytosol when GTP is hydrolysed. mRNA leaves bound to its own export proteins after processing.

The direction comes from the **Ran gradient**: the enzyme that loads Ran with GTP (RanGEF, also called RCC1) sits on chromatin inside the nucleus, while RanGAP sits in the cytosol. So Ran-GTP is high inside and low outside. Importins bind cargo where Ran-GTP is low; exportins bind cargo where it is high.

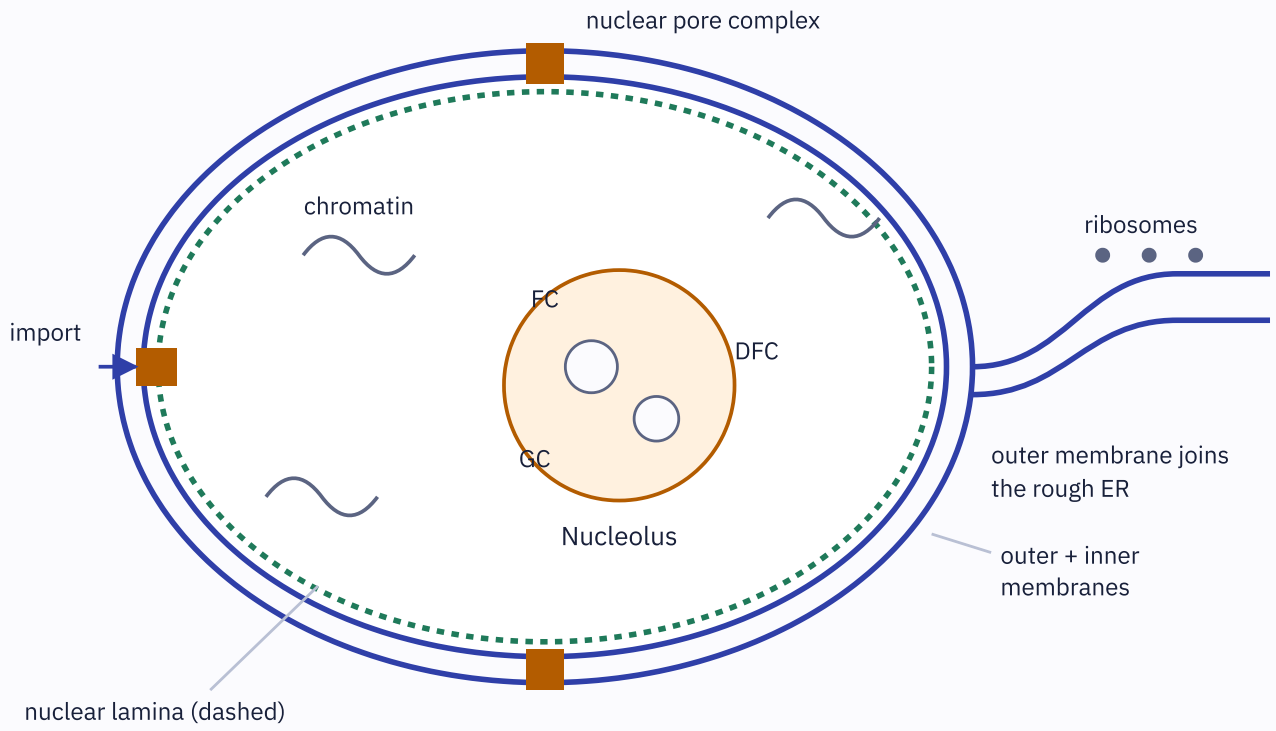


Figure 3.1.1 A nucleus in section. The envelope is two membranes; the outer one runs straight into the rough ER, so the space between them is ER lumen. Pores (orange) cross both membranes. Under the inner membrane lies the lamina. In the nucleolus, FC is the fibrillar centre (pale circles), DFC the dense fibrillar component around it, and GC the granular component outside.

The nucleolus: a ribosome factory

The **nucleolus** is not bounded by a membrane. It forms around the **nucleolar organizer regions** (NORs), the chromosome loops that carry many tandem copies of the ribosomal RNA (rRNA) genes. In humans the NORs sit on the short arms of the five acrocentric chromosomes, 13, 14, 15, 21 and 22. Under the electron microscope it shows three zones:

- **Fibrillar centre (FC):** holds the rRNA genes not being read, and RNA polymerase I waiting to start.
- **Dense fibrillar component (DFC):** where the rRNA genes are transcribed and the first processing begins.
- **Granular component (GC):** where rRNA folds and joins ribosomal proteins into maturing subunits.

RNA polymerase I makes one long precursor (45S in mammals). It is cut into the 18S, 5.8S and 28S rRNAs. The fourth rRNA, 5S, is made by RNA polymerase III from genes outside the nucleolus and brought in. Ribosomal proteins are made in the cytoplasm, imported through the pores, assembled with rRNA in the nucleolus, and the finished small and large subunits are exported separately. They join only in the cytoplasm, on an mRNA (see Chapter 3.5). A cell

making a lot of protein, such as a plasma cell or a growing cancer cell, has a large and prominent nucleolus. The nucleolus disappears during mitosis and re-forms afterwards.

The rest of the nucleus holds **chromatin**, DNA wrapped on histones. Loosely packed, active chromatin is **euchromatin**; tightly packed, mostly silent chromatin is **heterochromatin**, often found against the lamina. That belongs to molecular biology and is not repeated here.

WORKED EXAMPLE

Question (8 marks). A researcher joins green fluorescent protein (GFP, about 27 kDa) to three different partners and looks at where the fusion sits in living cells. (a) GFP alone; (b) GFP joined to a 100 kDa cytosolic enzyme; (c) the same 100 kDa fusion with PKKKRKV added at one end. Predict where each fluoresces and explain. (d) What happens to (c) if the cells make a Ran mutant that cannot hydrolyse GTP?

Solution.

1. **(a)** 27 kDa is below the passive limit of about 40–60 kDa, so GFP diffuses both ways through the pores and is seen in both cytoplasm and nucleus.
2. **(b)** The fusion is about 127 kDa. It is too large to diffuse and has no NLS, so no importin carries it. It stays in the cytoplasm; the nucleus looks dark.
3. **(c)** PKKKRKV is the SV40 NLS. Importin α binds it, importin β carries the complex through the pore, Ran-GTP releases it inside. The nucleus glows and the cytoplasm is dim.
4. **(d)** Importin β stays locked to Ran-GTP after it returns to the cytosol, because GTP is never hydrolysed. Free importin runs out, import stops, and newly made fusion protein builds up in the cytoplasm.

Answer: (a) both compartments; (b) cytoplasm only; (c) nucleus; (d) import fails, protein stays cytoplasmic. The marks are for using size, the NLS and the Ran cycle, not just for the location.

EXAM TRAPS

- The nucleolus has no membrane. Students who draw a membrane around it lose the mark.
- 5S rRNA is not made in the nucleolus; it is made by RNA polymerase III elsewhere in the nucleus and brought in.
- The NLS is not cut off after import (unlike an ER signal sequence), so a protein can be imported again after every mitosis.
- Ran-GTP is high inside the nucleus, not outside. Reversing this gradient reverses every arrow in your answer.
- The perinuclear space is continuous with the ER lumen, not with the cytosol.

CHECK YOURSELF

– REMEMBER **Name the three zones of the nucleolus.** 2 marks

Fibrillar centre, dense fibrillar component and granular component.

– REMEMBER **What proteins make up the nuclear lamina?** 1 mark

Lamins (A, B and C), which are intermediate-filament proteins.

– UNDERSTAND **Why is the nuclear envelope described as part of the ER?** 2 marks

The outer nuclear membrane is continuous with the rough ER membrane and carries ribosomes, and the perinuclear space is continuous with the ER lumen.

– UNDERSTAND **How does the Ran gradient make nuclear transport one-way?** 4 marks

RanGEF on chromatin keeps Ran-GTP high in the nucleus; RanGAP in the cytosol keeps it low there. Importins release cargo when Ran-GTP binds them, which happens only inside; exportins bind cargo only together with Ran-GTP, inside, and release it when GTP is hydrolysed outside.

– APPLY **A 20 kDa protein with no NLS and a 90 kDa protein with an NLS are made in the cytosol. Where does each end up?** 3 marks

The 20 kDa protein diffuses through the pores and spreads over both compartments. The 90 kDa protein is too large to diffuse but is carried in by importins, so it collects in the nucleus.

– APPLY **Electron micrographs show a very large nucleolus in one cell type. What does that tell you about the cell?** 2 marks

It is making ribosomes fast, so it is making a lot of protein, as in a secretory cell, a plasma cell or a rapidly growing cell.

– ANALYZE **Explain how separating transcription from translation lets a eukaryote do things a bacterium cannot.** 4 marks

In bacteria ribosomes start reading an mRNA while it is still being made, so it cannot be edited first. Behind the envelope, a eukaryotic pre-mRNA can be capped, spliced and tailed, and only finished mRNA is exported. This allows alternative splicing and quality control, and the pore becomes another point where gene expression is controlled.

BEYOND THE SYLLABUS

Cryo-electron tomography and AlphaFold models have now given near-complete structures of the human nuclear pore, and the FG-repeat channel is studied as an example of liquid-like protein phases. The nucleolus is a classic example of a membraneless organelle formed by phase separation, a current research theme also linked to neurodegenerative disease. Nuclear import signals are used in gene therapy and CRISPR delivery to bring Cas9 into the nucleus. Search "nuclear pore complex cryo-ET" and "nucleolus phase separation" to read more. None of this is for the exam.

3.2 Endoplasmic reticulum

The endoplasmic reticulum (ER) is a network of membrane tubes and flat sacs that spreads from the nuclear envelope through the cytoplasm. Its inside, the **lumen**, is one connected space. The ER is the entry gate of the secretory pathway: every protein that will be secreted, or that will live in the plasma membrane, Golgi, lysosome or ER itself, enters here. The ER also makes most of the cell's membrane lipids, and in muscle it stores the calcium that triggers contraction.

You need: membrane lipids and proteins from **Unit 2, Membrane proteins and lipids**, and the nuclear envelope from Chapter 3.1.

Rough and smooth ER

Rough ER (RER) is studded with ribosomes on its cytosolic face and is usually built of flattened sacs (cisternae). It makes secretory and membrane proteins. Cells that secrete a lot of protein, such as pancreatic acinar cells (digestive enzymes) or plasma cells (antibodies), are packed with RER.

Smooth ER (SER) has no ribosomes and is usually tubular. Its jobs depend on the cell:

- **Lipid synthesis:** phospholipids and cholesterol in all cells; steroid hormones in the adrenal cortex, testis and ovary.
- **Detoxification:** in liver cells, cytochrome P450 enzymes add hydroxyl groups to drugs and toxins so they dissolve in water and leave in urine. Long use of drugs such as phenobarbital makes the liver SER expand.
- **Calcium store:** in muscle the SER is called the **sarcoplasmic reticulum**; it pumps Ca^{2+} in and releases it on a nerve signal.
- **Glucose release:** in liver, glucose-6-phosphatase in the ER membrane frees glucose from glycogen breakdown.

The point where vesicles bud off towards the Golgi is a ribosome-free patch of RER called the **ER exit site** (transitional ER).

How a protein enters the ER: the signal hypothesis

All protein synthesis starts on free ribosomes in the cytosol. What sends a ribosome to the ER is the protein it is making. Günter Blobel's **signal hypothesis** (Nobel Prize 1999) explains how. A secretory protein begins with an **ER signal sequence**: about 15–30 amino acids at the N-terminus, with a core of 6–12 hydrophobic residues.

1. The signal sequence comes out of the ribosome's exit tunnel.
2. The **signal recognition particle** (SRP), a complex of six proteins and a 7S RNA, binds the signal sequence and the ribosome, and translation pauses.
3. SRP brings the ribosome to the **SRP receptor** on the ER membrane. Both bind GTP.
4. The ribosome is handed to the **translocon**, a channel called **Sec61**. GTP is hydrolysed, SRP and its receptor separate and SRP is recycled.
5. Translation restarts and the growing chain is threaded through Sec61 into the lumen. This is **cotranslational translocation**: the protein crosses while it is still being made.
6. **Signal peptidase** on the lumen side cuts off the signal sequence. The finished protein is released into the lumen when synthesis ends.

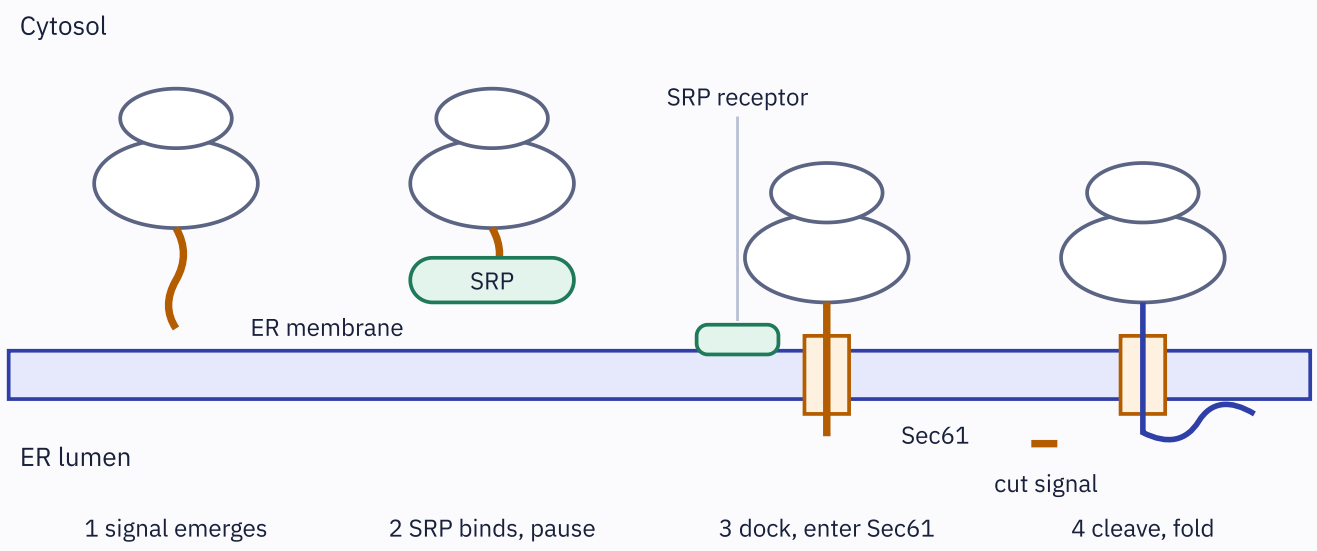


Figure 3.2.1 Cotranslational import into the ER. The signal sequence (orange) appears, SRP grabs it and pauses translation, the ribosome docks on the SRP receptor and Sec61 channel, and the chain enters the lumen; signal peptidase then cuts the signal off. Notice the ribosome stays on the cytosolic side the whole time.

Membrane proteins, folding and quality control

A protein that must stay in the membrane carries a second hydrophobic stretch, a **stop-transfer anchor sequence**. When it enters Sec61, the channel opens sideways and releases it into the bilayer as a membrane-spanning α-helix, while the rest of the chain finishes on the cytosolic side. An internal **signal-anchor sequence** (not cleaved) can start transfer in either orientation, and alternating start and stop sequences give proteins that cross the membrane many times.

Once in the lumen, proteins are folded and modified:

- **Chaperones** such as BiP hold unfolded chains so they do not clump.
- **Disulfide bonds** are made by protein disulfide isomerase (PDI); the lumen is oxidising, unlike the cytosol.
- **N-linked glycosylation:** the enzyme oligosaccharyltransferase moves a ready-made 14-sugar block ($Glc_3Man_9GlcNAc_2$, built on the lipid dolichol) on to asparagine in the sequence Asn–X–Ser/Thr, where X is any amino acid except proline.
- **Calnexin and calreticulin** check folding: a protein that is still misfolded gets a glucose added back and is held for another try.

Proteins that never fold are pulled back to the cytosol and destroyed by the proteasome; this is **ER-associated degradation** (ERAD). When misfolded proteins pile up, the ER sends the **unfolded protein response** (UPR) through three sensors, IRE1, PERK and ATF6: the cell makes more chaperones and slows translation, and if the stress lasts it triggers apoptosis (Unit 4).

Resident lumen proteins such as BiP carry the C-terminal signal **KDEL** (Lys-Asp-Glu-Leu); if they escape to the Golgi, a KDEL receptor returns them.

WORKED EXAMPLE

Question (6 marks). Predict the final location and orientation of each protein. (a) An N-terminal cleavable signal sequence, nothing else hydrophobic. (b) An N-terminal cleavable signal sequence and one stop-transfer sequence in the middle. (c) Same as (a) plus KDEL at the C-terminus. (d) No signal sequence at all.

Solution.

1. **(a)** SRP sends it to the ER, the whole chain is translocated and the signal is cut off. It is a soluble lumen protein; with no other signal it moves on to the Golgi and is secreted (the default route, see Chapter 3.3).
2. **(b)** Translocation starts at the N-terminus and stops at the stop-transfer sequence, which becomes the single membrane-spanning helix. The N-terminus is in the lumen and the C-terminus in the cytosol. This is a **single-pass type I** membrane protein. After the Golgi it reaches the plasma membrane with its N-terminus outside the cell, because the lumen side becomes the outside after vesicle fusion.
3. **(c)** It enters the lumen like (a), but the KDEL receptor keeps returning it from the Golgi. It is an ER-resident lumen protein.
4. **(d)** It is finished on a free ribosome and stays in the cytosol, unless it carries another signal (nuclear, mitochondrial or peroxisomal).

Answer: (a) secreted; (b) plasma membrane, type I, N-terminus outside; (c) ER lumen; (d) cytosol.

EXAM TRAPS

- Ribosomes are not permanently fixed to the RER. The same ribosome can be free or bound; the protein it makes decides.
- The ER signal sequence is cleaved; the nuclear localization signal is not. Do not mix them.
- N-linked sugars go on asparagine (Asn) in the ER. O-linked sugars go on serine or threonine, mostly in the Golgi.
- The lumen of the ER is topologically equivalent to the outside of the cell. This is why the lumen side of a membrane protein faces outward at the plasma membrane.
- Sarcoplasmic reticulum is smooth ER, not rough ER.

CHECK YOURSELF

– REMEMBER **Give two functions of the smooth ER.** 2 marks

Any two of: lipid and steroid synthesis, detoxification of drugs by cytochrome P450, Ca^{2+} storage in muscle, glucose release in liver.

– REMEMBER **What is the consensus sequence for N-linked glycosylation?** 1 mark

Asn–X–Ser/Thr, where X is any amino acid except proline.

– UNDERSTAND **Why does translation pause when SRP binds?** 2 marks

So that the chain is not finished in the cytosol before the ribosome reaches the ER; the protein must cross the membrane while it is still unfolded.

– UNDERSTAND **What is the unfolded protein response and why does a cell need it?** 3 marks

A signal sent by ER stress sensors (IRE1, PERK, ATF6) when misfolded proteins build up. It raises chaperone levels and slows new protein synthesis so the ER can catch up; if stress persists it triggers apoptosis to remove the damaged cell.

– APPLY **A liver cell is given phenobarbital for several days. Predict the change in its ER.** 2 marks

The smooth ER expands and makes more cytochrome P450 enzymes to detoxify the drug.

– APPLY **A cell-free translation system has ribosomes and mRNA for a secreted protein but no ER membranes. What length of protein is made, and why?** 3 marks

Without SRP, the full-length protein is made with its signal sequence still attached, because there is no signal peptidase. With SRP but no membranes, translation stalls. Adding microsomes (ER vesicles) gives the shorter, cleaved protein inside the vesicles. This was the original evidence for the signal hypothesis.

– ANALYZE **Compare how proteins enter the ER and the nucleus.** 4 marks

ER: N-terminal hydrophobic signal, usually cotranslational, unfolded chain through Sec61, signal cleaved, GTP used by SRP and receptor. Nucleus: internal basic NLS, post-translational, folded protein through a large pore, signal kept, GTP used by Ran.

BEYOND THE SYLLABUS

Many therapeutic proteins, including insulin and monoclonal antibodies made in CHO cells, pass through the ER of the producing cell, and bioprocess engineers tune chaperone and UPR genes to raise yield. Signal-peptide choice is a standard design step when expressing a protein for secretion in yeast or mammalian cells. ER stress is studied in diabetes, where insulin-making β cells work near their ER limit. Search "signal peptide optimisation recombinant secretion" and "unfolded protein response diabetes". This is not for the exam.

Key terms of this lesson

TERM	MEANING OR FORMULA	CHAPTER
Nuclear localization signal	short basic sequence (e.g. PKKKRKV) read by importin α; not cleaved	3.1
Ran gradient	Ran-GTP high in nucleus (RanGEF on chromatin), low in cytosol (RanGAP)	3.1
Nucleolus	membraneless site of rRNA synthesis and subunit assembly: FC, DFC, GC	3.1
Signal sequence	N-terminal hydrophobic stretch read by SRP; cut by signal peptidase	3.2
N-linked glycosylation	14-sugar block added to Asn in Asn–X–Ser/Thr (X ≠ Pro), in the ER	3.2
KDEL	C-terminal retention signal returning ER lumen proteins from the Golgi	3.2

Glossary (terms used in this lesson)

Cisterna A flattened membrane sac of the ER or Golgi.

Importin Transport receptor that carries NLS proteins into the nucleus.

Lamina Mesh of lamin filaments under the inner nuclear membrane.

Nucleolus	Membraneless nuclear region where rRNA is made and ribosome subunits assembled.
Nucleoporin	One of the proteins that make up the nuclear pore complex.
Sec61	The channel (translocon) through which proteins cross the ER membrane.
Signal recognition particle	RNA–protein complex that brings ribosomes making signal-sequence proteins to the ER.
Unfolded protein response	ER stress signal that raises chaperones and slows translation.

The full unit book

Every chapter of Unit 3, the unit summary, the exam question bank with model answers and the full glossary: Cell Biology · Unit 3 · Cell Organelles and Cytoskeleton (one file, works offline).

Sources and further reading

Standard textbooks

- Bruce Alberts and others, *Molecular Biology of the Cell* (W. W. Norton / Garland Science): chapters on intracellular compartments and protein sorting, intracellular membrane traffic, energy conversion in mitochondria and chloroplasts, and the cytoskeleton. (from memory; check)
- Harvey Lodish and others, *Molecular Cell Biology* (W. H. Freeman / Macmillan). (from memory; check)
- Geoffrey M. Cooper, *The Cell: A Molecular Approach* (Sinauer / Oxford University Press). (from memory; check)
- Gerald Karp, *Cell and Molecular Biology: Concepts and Experiments* (Wiley). (from memory; check)
- P. S. Verma and V. K. Agarwal, *Cell Biology, Genetics, Molecular Biology, Evolution and Ecology* (S. Chand), widely prescribed in Indian universities. (from memory; check)

Open resources

- OpenStax, *Biology 2e*, Chapter 4 "Cell Structure", section 4.3 "Eukaryotic Cells" and 4.4 "The Endomembrane System and Proteins" (CC BY 4.0): an open textbook covering the organelles of

this unit at an introductory level. Reached through the course pages below; section titles from memory; check.

- Penn State DuBois, Biology 110 course pages built on OpenStax *Biology 2e*: psu.pb.unizin.org/biol110/part/cell-structure/
- City Tech OpenLab, Biology 1101 Course Hub, Lecture 11 "The Eukaryotic Cell", with readings and slides: openlab.citytech.cuny.edu/bio1101coursehub/?p=3597

Numbers in this book (pore size limit, ribosome protein counts, mtDNA size, filament diameters, motor step sizes) are standard textbook values written from memory; check them against your prescribed book. Worked-example inputs such as step rates and translation speeds are given in the question and are for practice.

Syllabus: the University free n easy universal syllabus, built from top universities' syllabi.

Built for the University free n easy YouTube channel from the universal syllabus of Cell Biology (Oct 2026). Written with top universities' syllabi and standard textbooks; check a formula against your prescribed book before an exam, and report mistakes on the channel.