3

CELLULAR ORGANELLES

ENDOPLASMIC RETICULUM

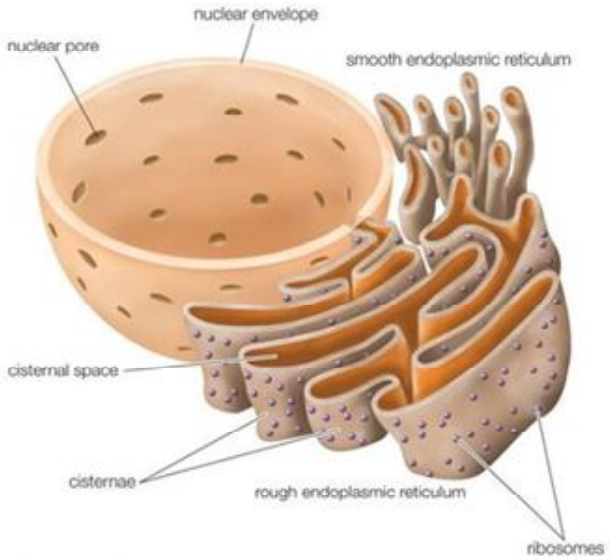


Figure 3.1: Rough and Smooth endoplasmic reticulum.

→ It is single membrane-bound and the largest intracellular compartment.

→ It is a network of closed and flattened structure.

→ The ER membrane enclose a space, separating it from the environment known as, the **ER lumen.**

→ The composition of the luminal (or, cisternal) space is quite different from the surrounding cytosol.

→ ER membranes interact with cytoskeleton and also contain specialized domains for distinct functions.

→ It is universally present in all eukaryotic cells, **except**, red blood cells (mature erythrocytes) and sperm cells.

→ Depending upon the presence of ribosomes on the surface of ER membrane, ER is differentiated into two types, Smooth endoplasmic reticulum and Rough endoplasmic reticulum.

Smooth endoplasmic reticulum (SER):

→ ER lacking membrane-bound ribosomes are called *smooth endoplasmic reticulum* (SER).

→ They are also called as ***Transitional ER***.

→ SER is found in a number of cell types including, *skeletal muscles*, *kidney tubules*, and *steroid-producing endocrine glands.*

Functions of SER:

- Involved in the synthesis of steroid hormones in the endocrine cells of the gonad and adrenal cortex.
- Detoxification of a wide variety of organic compounds, including alcohol, in the liver.
- Sequester calcium ions within the cytoplasm of the cells.
- Regulated release of Ca^{2+} from the SER of skeletal and cardiac muscle cells triggers contraction.

Rough endoplasmic reticulum (RER):

→ RER is characterized by the presence of ribosomes bound to its cytosolic surface.

→ Usually, composed of network of flattened sacs (or, cisternae).

→ It is found continuous with the outer membrane of the nuclear membrane.

Functions of RER:

- RER is the site of biosynthesis of proteins, carbohydrate chains and phospholipids that journey through the membranous compartments of the cell.
- Certain polypeptides such as, secreted proteins, integral membrane proteins, and soluble proteins that reside within compartments of the endomembrane system (ER, golgi complex, lysosomes, endosomes, vesicles and plant vacuoles), are synthesized on ribosomes attached to the cytosolic surface of RER membranes.

Signal hypothesis:

The site of protein synthesis in a cell is determined by the sequence of amino acids at the N-terminal of the polypeptide. The **Signal sequence** (a stretch of 6-15 hydrophobic amino acid residues) present at the N-terminus of the secretory proteins directs them to the ER membrane where, the polypeptide moves into the cisternal space of the ER through a protein-lined, aqueous channel in the ER membrane.

❖ The polypeptide moves through the membrane cotranslationally.

Synthesis of Secretory, Lysosomal, or Plant Vacuolar proteins on membrane-bound ribosomes (Figure 3.2):

- The synthesis begins with the binding of m-RNA to the free ribosome.
- As the polypeptide emerges out of the ribosome, the hydrophobic signal sequence (present at the N-terminus) is recognized by a signal recognition particle (SRP).
- SRP binds to both, the signal sequence on the nascent polypeptide as well as the ribosome, and serves as a tag, enabling the binding of the entire complex (SRP-ribosome-nascent polypeptide) to the cytosolic surface of the ER membrane.
- This binding occurs through 2 interactions:
  - ♦ between SRP and SRP receptor, and
  - ♦ between ribosome and translocon.
- Once the binding takes place, the SRP releases from the receptor, the ribosome attaches to the cytosolic end of the translocon and the signal sequence on the nascent polypeptide is inserted into the narrow aqueous channel of the translocon.
- The growing polypeptide is translocated through the hydrophobic pore ring into the ER lumen.
- Upon termination of translation, the bound ribosome is released from the ER membrane.

❖ Most of the steps involved in the synthesis and trafficking of secretory proteins are regulated by the binding or hydrolysis of GTP.

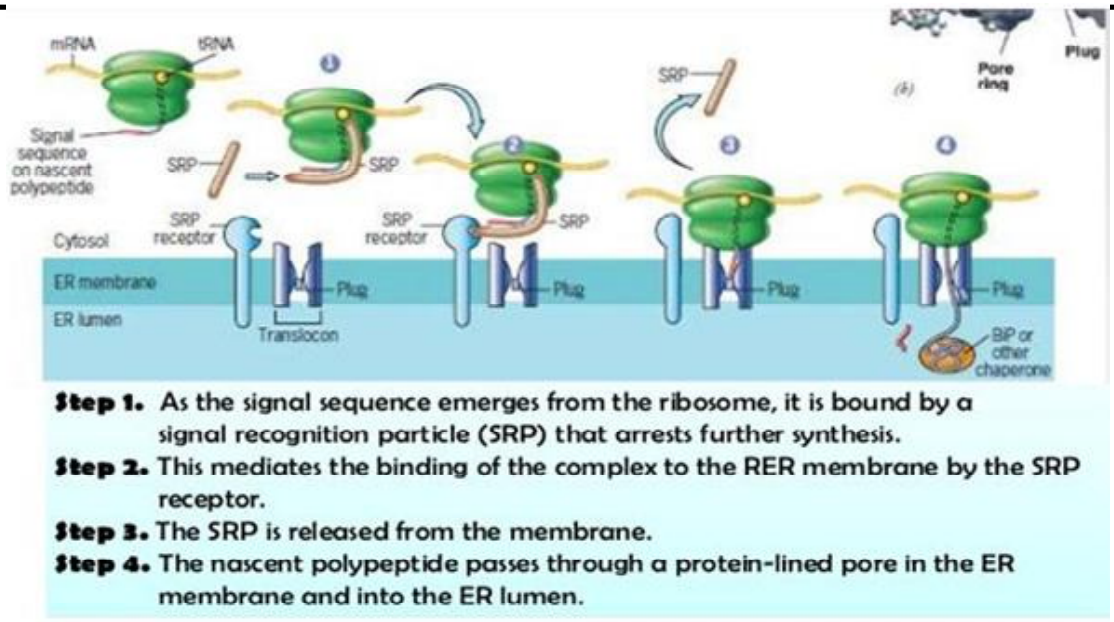


Figure 3.2: Synthesis of Secretory, lysosomal, or Plant Vacuolar Proteins on membrane-bound ribosomes.

Protein processing in ER:

→ Signal sequence present at the N-terminal of the polypeptide is removed by the proteolytic enzyme, ***Signal peptidase***.

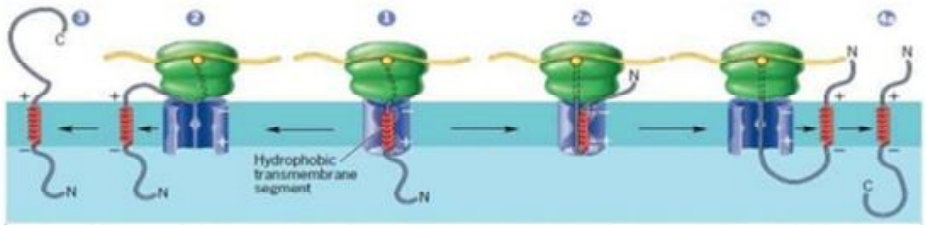
→ The enzyme, ***Oligosaccharyl transferase*** adds carbohydrates to the nascent protein.

→ ***Molecular chaperons*** present in the RER lumen recognize and bind to unfolded/ misfolded proteins thereby, facilitating their proper folding.

→ ***Protein disulfide isomerase*** **(PDI)** catalyzes the formation of disulfide bonds. [The reduced form of cysteine residue (-SH) in the polypeptides are oxidized to disulfides (-SS-) in the ER].

Synthesis of integral membrane proteins on membrane-bound ribosomes (Figure 3.3):

- The translocation of membrane proteins into the ER (cotranslational) uses the same machinery as that of the secretory, lysosomal, or plant vacuolar proteins (as mentioned above).
- Although, unlike secretory, lysosomal, or plant vacuolar proteins which pass entirely through ER membrane during translocation, integral proteins contain one or more hydrophobic transmembrane segments that are shunted directly from the channel of the translocon into the lipid bilayer.



Step 1. The nascent polypeptide enters the translocon just as if it were a secretory protein.
Steps 2–3 show the synthesis of a transmembrane protein whose N-terminus is in the lumen of the ER and C-terminus is in the cytosol.
In **step 2**, the translocon has opened laterally and expelled the transmembrane segment into the bilayer.
Step 3 shows the final disposition of the protein.
Steps 2a–4a show the synthesis of a transmembrane protein whose C-terminus is in the lumen and N-terminus is in the cytosol.
In **step 2a**, the translocon has reoriented the transmembrane segment, in keeping with its reversed positively and negatively charged flanks.
In **step 3a**, the translocon has opened laterally and expelled the transmembrane segment into the bilayer.
Step **4a** shows the final disposition of the protein.

Figure 3.3: Synthesis of Integral membrane proteins on membrane-bound ribosomes

Membrane biosynthesis in ER:

→ Membranes do not arise *de novo* but, from pre-existing membranes. These are formed when newly synthesized proteins and lipids are inserted into existing membranes in the ER.

→ Membrane components move from the ER to various other membrane compartments. In this time course, the proteins and lipids present in the membrane gets modified by the enzymes residing in the particular cell organelle. Such modifications gives each membrane compartment a unique composition and distinct identity.

→ The asymmetric nature of the phospholipid bilayer is also established in the ER.

→ Almost all membrane lipids are synthesized **entirely** within the ER, except,

- *Sphingomyelin and Glycolipid:* Synthesis begins in the ER and completes in the Golgi complex.
- Some unique *lipids of mitochondria and chloroplast* membranes: Synthesized by enzymes present in their own membranes.

Glycosylation in the RER (Figure 3.4):

→ Almost all proteins produced on membrane-bound ribosomes become glycoproteins.

→ Carbohydrate groups play a crucial role in the function of many glycoproteins such as, binding sites during interactions with other macromolecules or, proper folding of proteins to which they are attached, etc.

→ The sugar sequence that comprise the oligosaccharide of glycoprotein are highly specific.

→ The addition of oligosaccharide chain is catalyzed by the enzymes **glycosyltransferases**. Each of these enzymes transfers a specific monoaccharide from a nucleotide sugar, such as **GDP-mannose of UDP-N-acetylglucosamine** to the growing end of the carbohydrate chain.

→ The basal or core segment of each carbohydrate chain is not assembled directly on the protein but put together on a lipid carrier and then transferred to the specific **arginine** residue of the polypeptide.

→ The lipid carrier, ***dolichol phosphate***, is found embedded in the ER membrane. Sugars are added to this molecule one at a time by glycosyl transferases. In this process, glycosylation begins with the transfer of another *N-acetyl glucosamine 1-phosphate*, followed by the transfer of another *N-acetyl glucosamine, nine mannose* and *three glucose* units in the precise pattern.

→ The pre-assembled block of the 14 sugars is then transferred by the ER enzyme oligosaccharyl transferase from dolichol phosphate to asparagines in the nascent polypeptide, as the polypeptide is being translocated into the ER lumen.

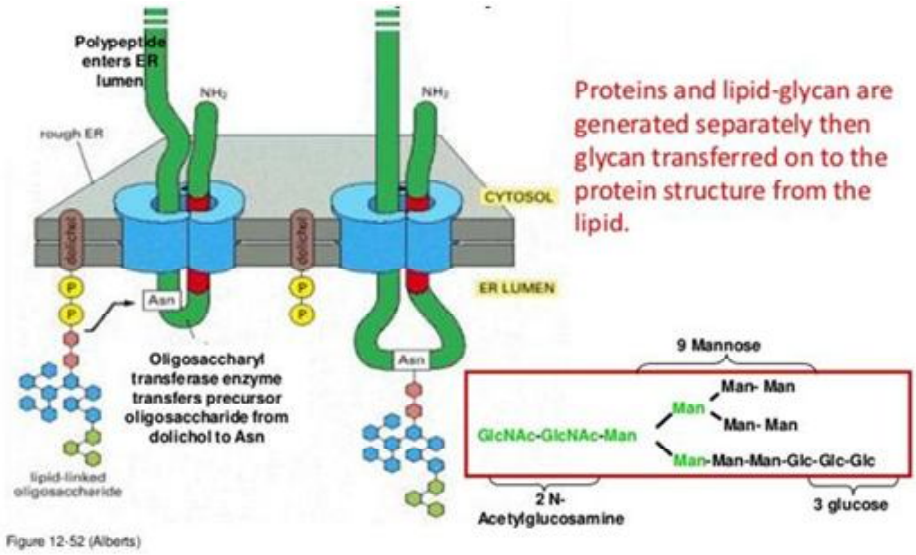


Figure 3.4: Protein glycosylation in RER.

Vesicular transport from ER to Golgi complex:

- The exit sites of RER cisternae lack ribosomes and is the place where the first transport vesicles in the biosynthetic pathway are formed.
- As soon as the vesicle buds out from the ER membrane, the transport vesicles fuse with one another to form larger vesicles and interconnected tubules in the region between the ER and Golgi complex. This region is known as **ERGIC** (**endoplasmic *reticulum Golgi intermediate compartment***), and the vesicular-tubular carrier that form there are called **VTCs**.
- Once formed, the VTCs move away from the ER towards the Golgi complex. This movement of VTCs occurs along tracks composed of microtubules.

GOLGI COMPLEX

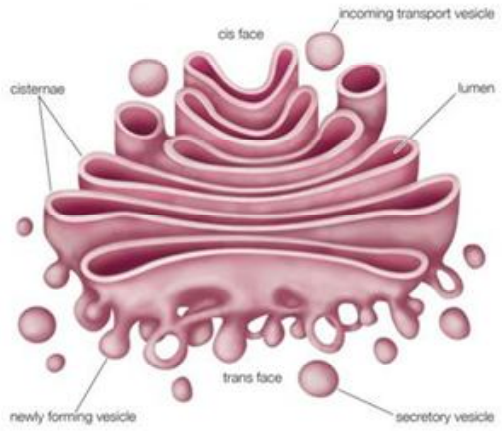


Figure 3.5: Golgi Complex.