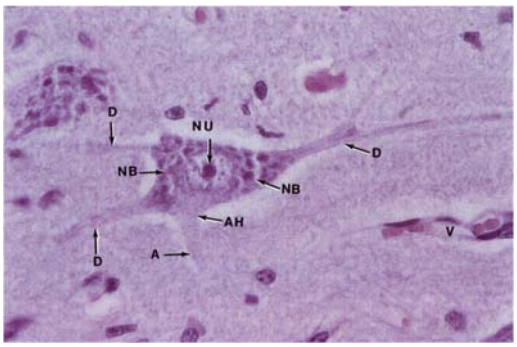
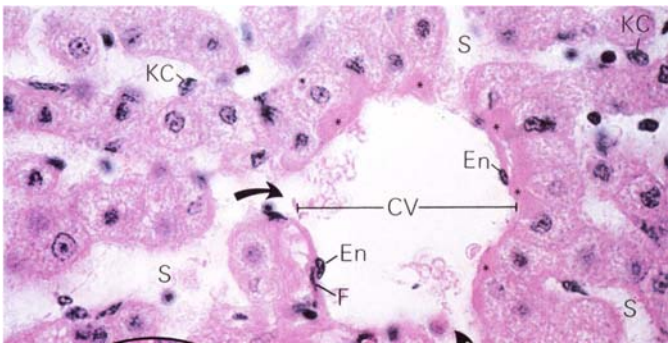
Endoplasmic Reticulum

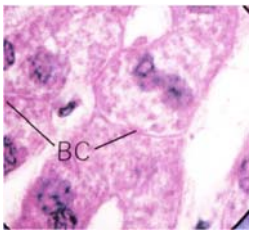
- What's ER? – definition
- How is ER? – description
- Why is ER? – functions

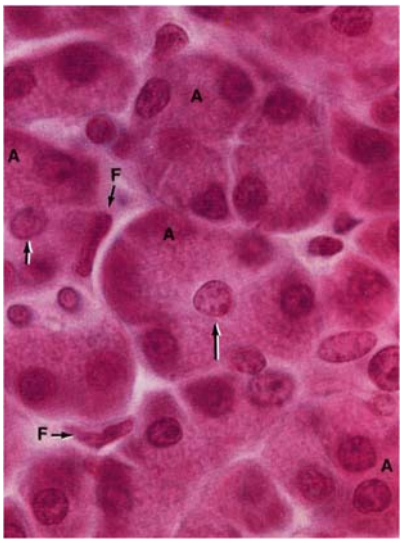


Nissl's bodies – neurons



Berg's bodies – hepatocytes



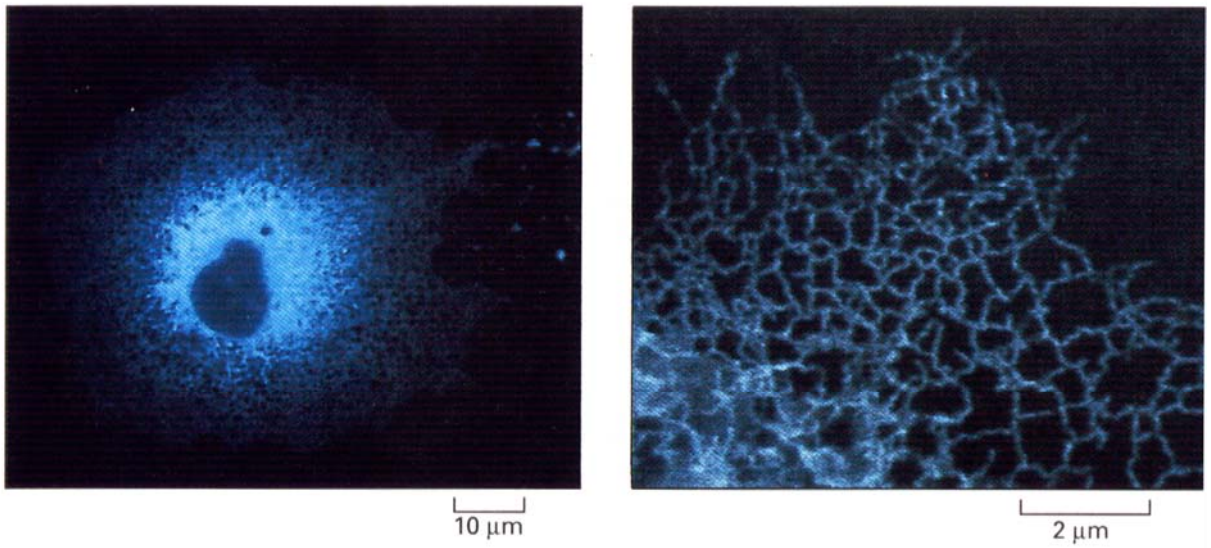


Organelle structure

histocytochemical evidences

Ergastoplasm –pancreatic acinar cells

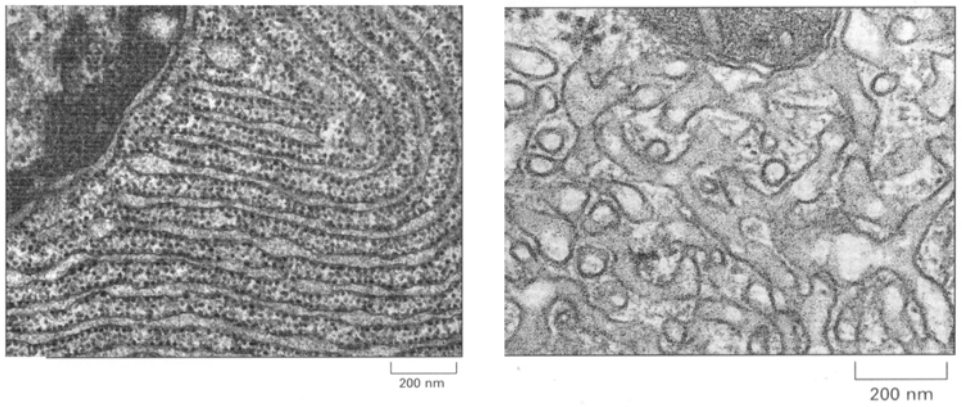
Organelle structure
immunocytochemical evidence



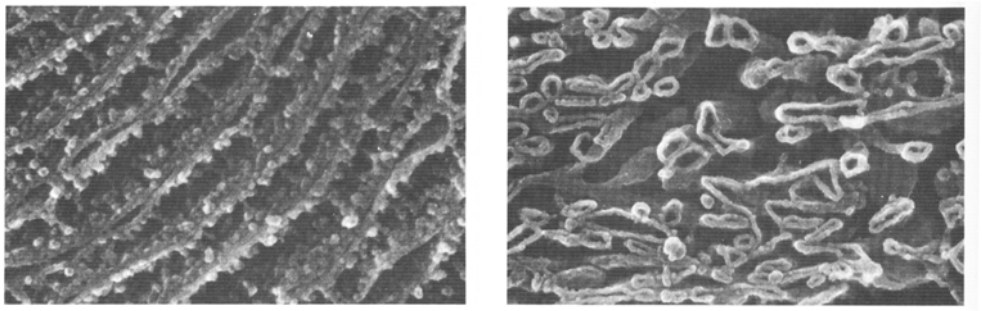
The significance of the organelle's name

Organelle ultrastructure
electron microscopy

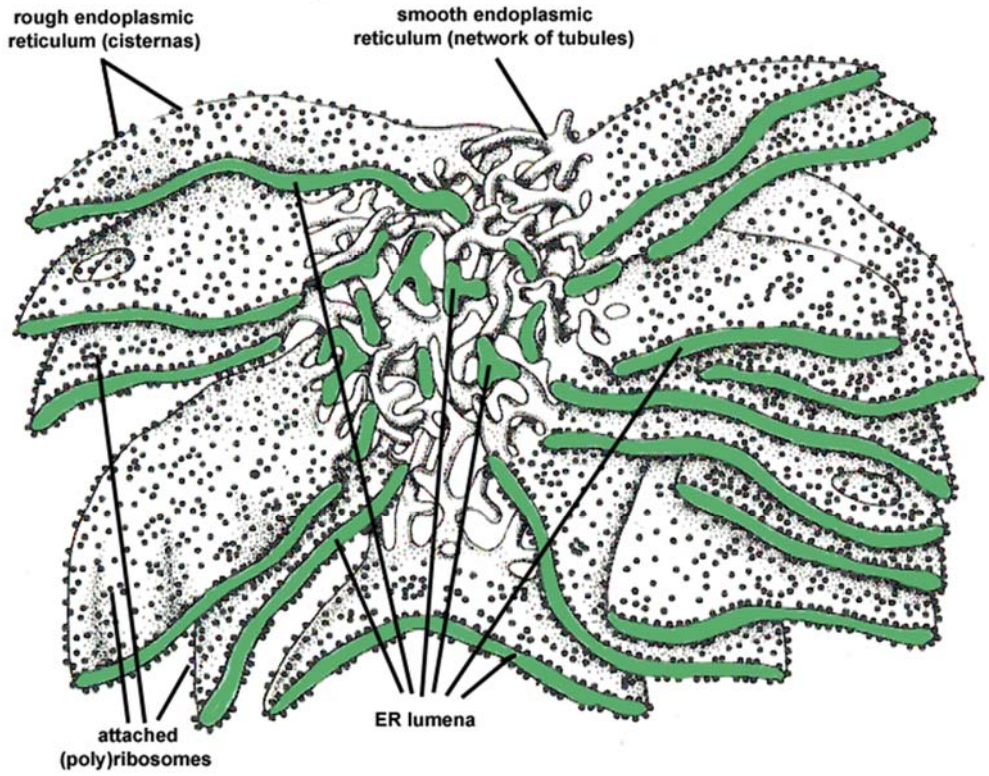
Transmission
Electron Microscopy



Scaning
Electron Microscopy



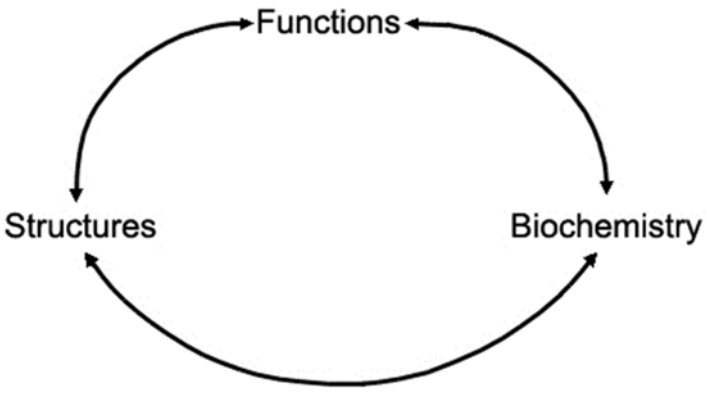
Organelle ultrastructure
3D organization of ER



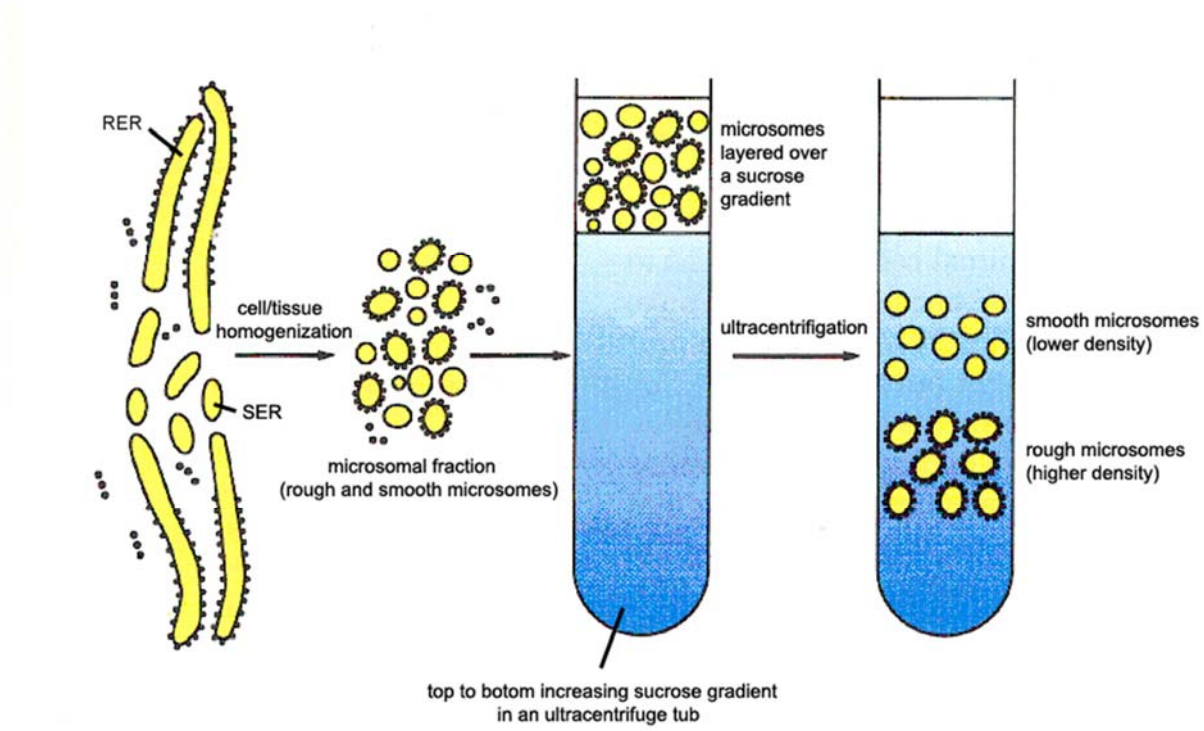
Roles of ER

Functions must be understood in terms of structures; structures must be understood in terms of chemistry.

(George Emil PALADE)



Method of ER study (main approach)

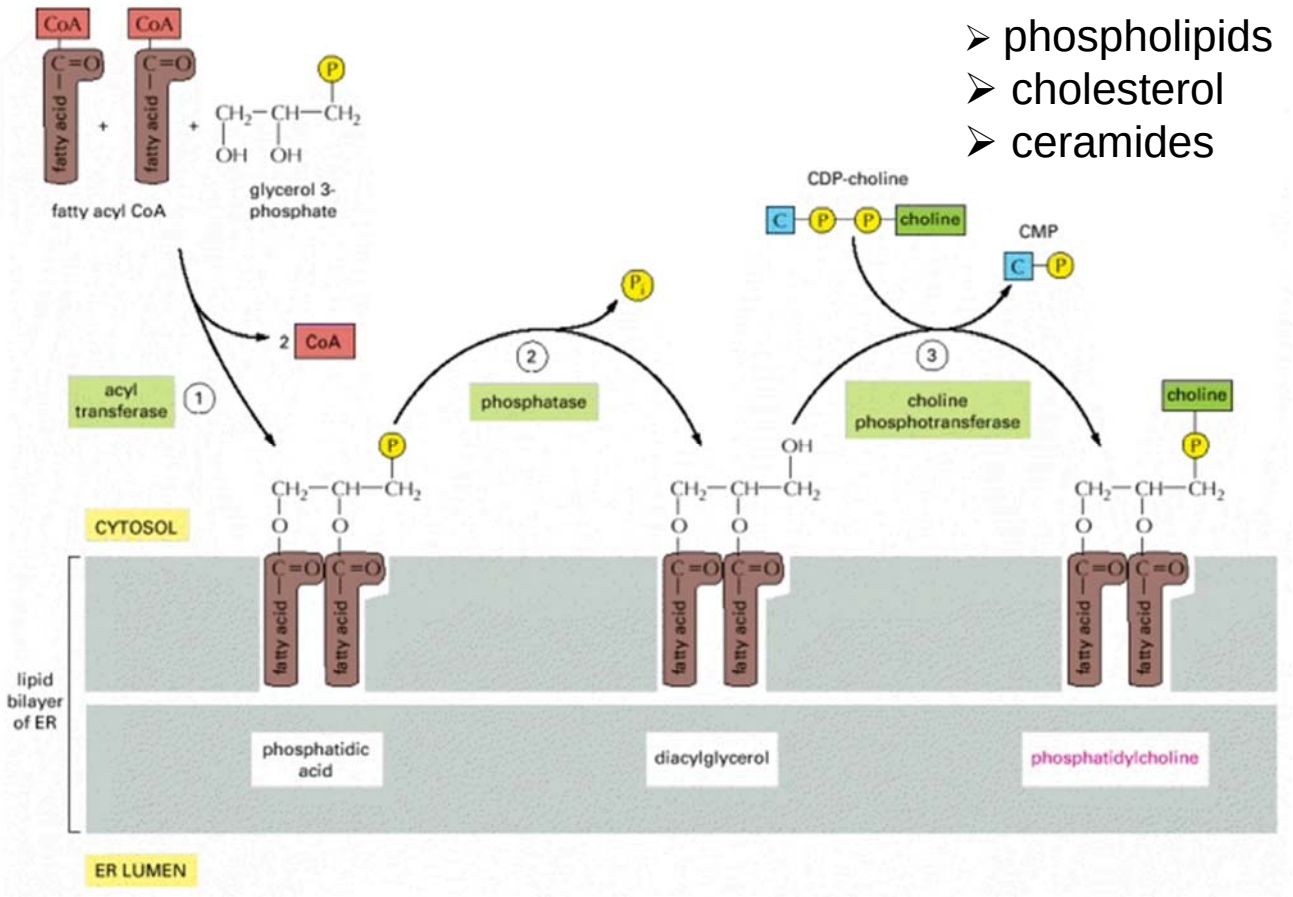


Functions of the Smooth Endoplasmic Reticulum

- **Metabolism of lipids;**
- **Cellular Detoxification;**
- **Special Functions.**

Biosynthesis of Membrane Lipids

- phospholipids
- cholesterol
- ceramides



Assembly of membrane lipids

into bilayer

- Production on the internal leaflet
- Asymmetrical distribution
- Phospholipid translocators
  - Flippases
  - Floppases
  - Scramblases

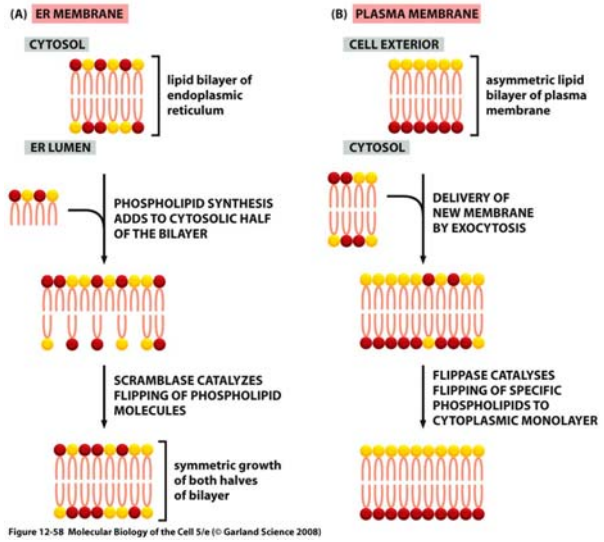
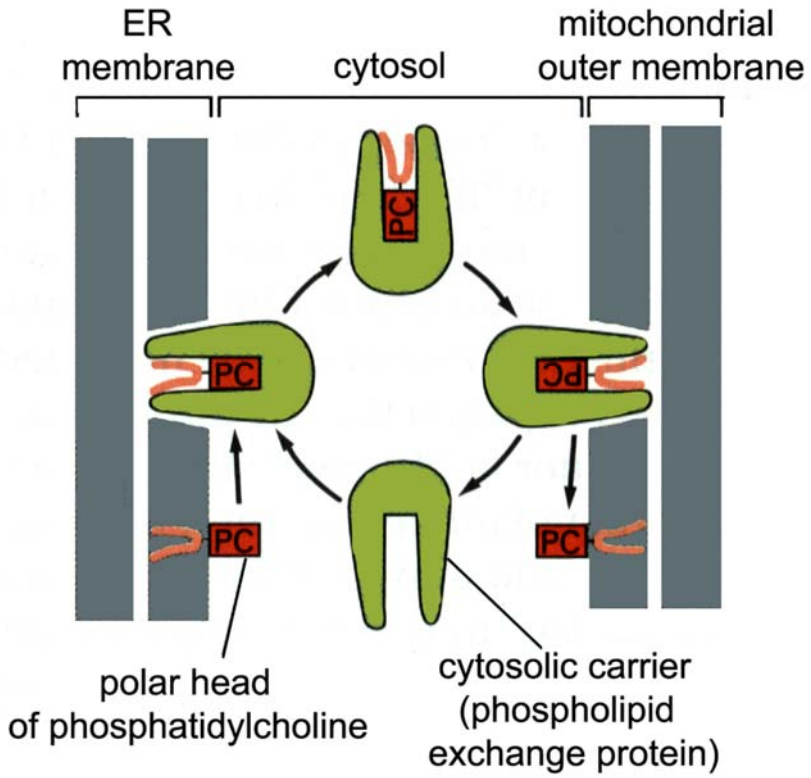
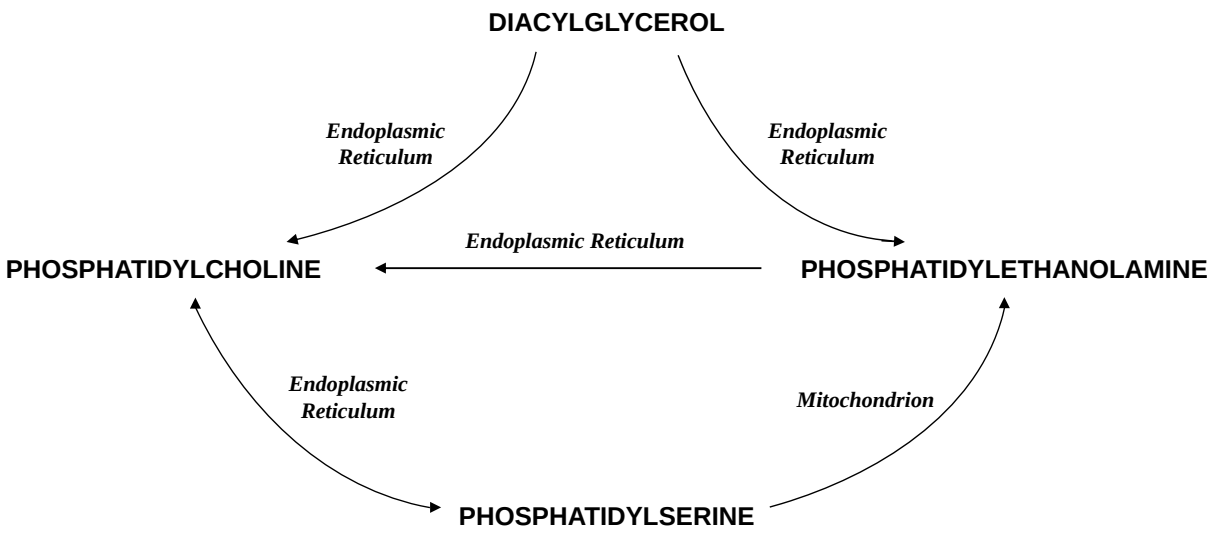


Figure 12-58 Molecular Biology of the Cell 5/e (© Garland Science 2008)

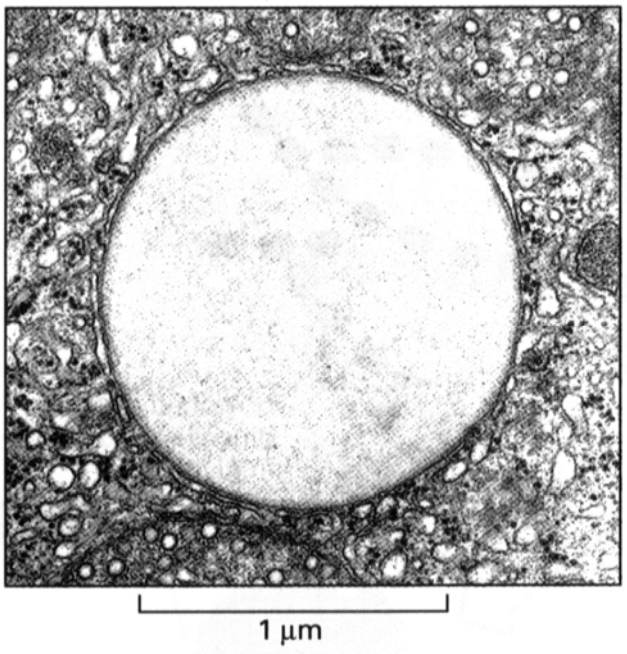
Phospholipid transfer toward mitochondria

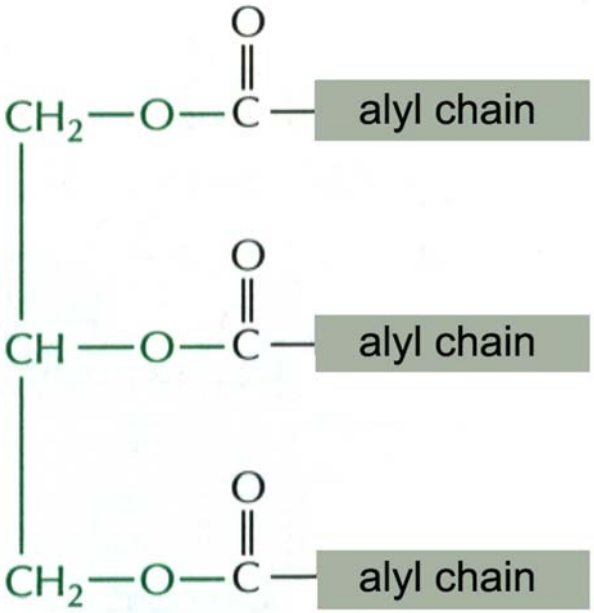


Inter-relationships among biosynthesis' pathways of phospholipids in mammalian cells



Triglyceride Metabolism





Fatty Acid Desaturation

Effector:

Enzyme complex containing fatty acid desaturase / cytochrome b_5 / NADH-cytochrome b_5 reductase

Effect:

Control and modulation of membrane fluidity

Cellular Detoxification

Effectors:

Enzyme complex containing cytochrome P450 / NADPH-cytochrome P450 reductase

Proved by:

Reversible hyperplasia of smooth endoplasmic reticulum in hepatocyte

Special Function

- Dynamic deposit for calcium ions
- Sarcoplasmic reticulum (striated muscle cell)
- Calsequestrin
- Calcium channels (ligand operated)
- Calcium pumps

Functions of the Rough Endoplasmic Reticulum

- Biosynthesis of some proteins
- Processing of biosynthetized proteins
  - Co/post-translational modification
  - Folding and assembly
- Sorting and transport to Golgi

Biosynthesis of some proteins

- ✓ **Membrane proteins** (except proteins of mitochondrial membranes and most proteins of peroxisomal membrane)
- ✓ **Proteins for lumina of ER, Golgi, lysosomes, and endosomal system**
- ✓ **Exported proteins (secretory proteins)**

A polyribosome

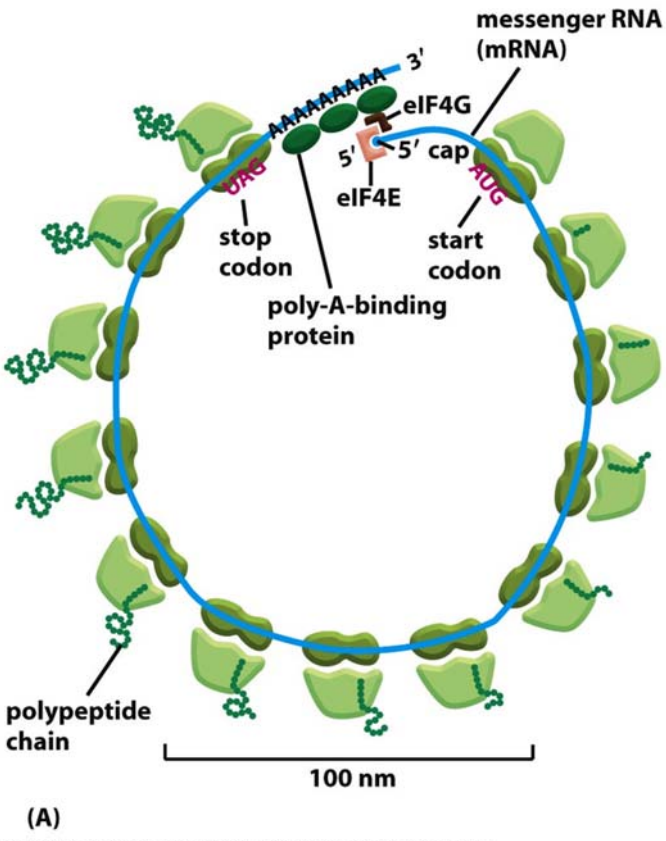
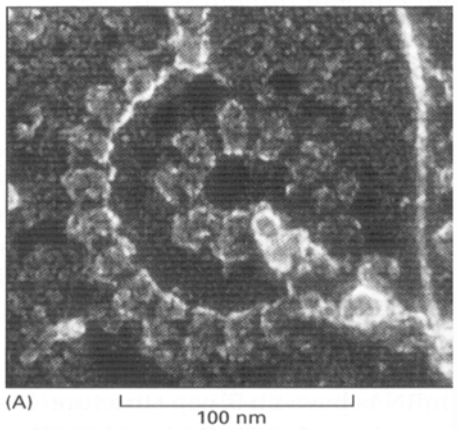
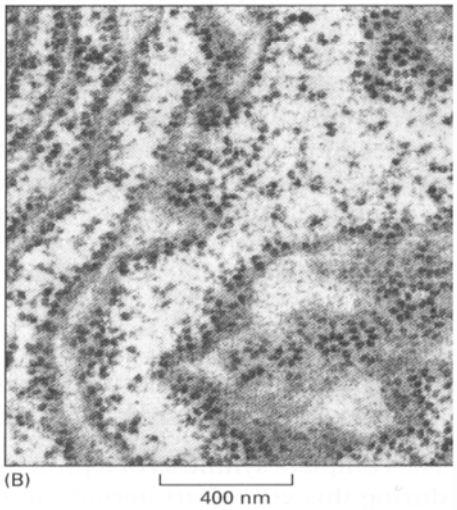


Figure 6-76 Molecular Biology of the Cell 5/e (© Garland Science 2008)





Signal Peptides

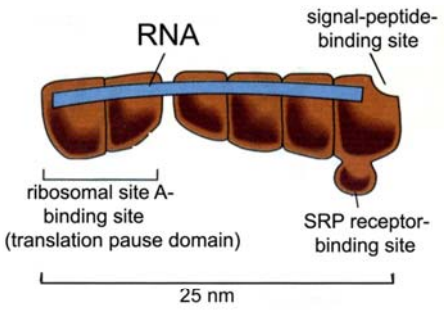
Table 12–3 Some Typical Signal Sequences

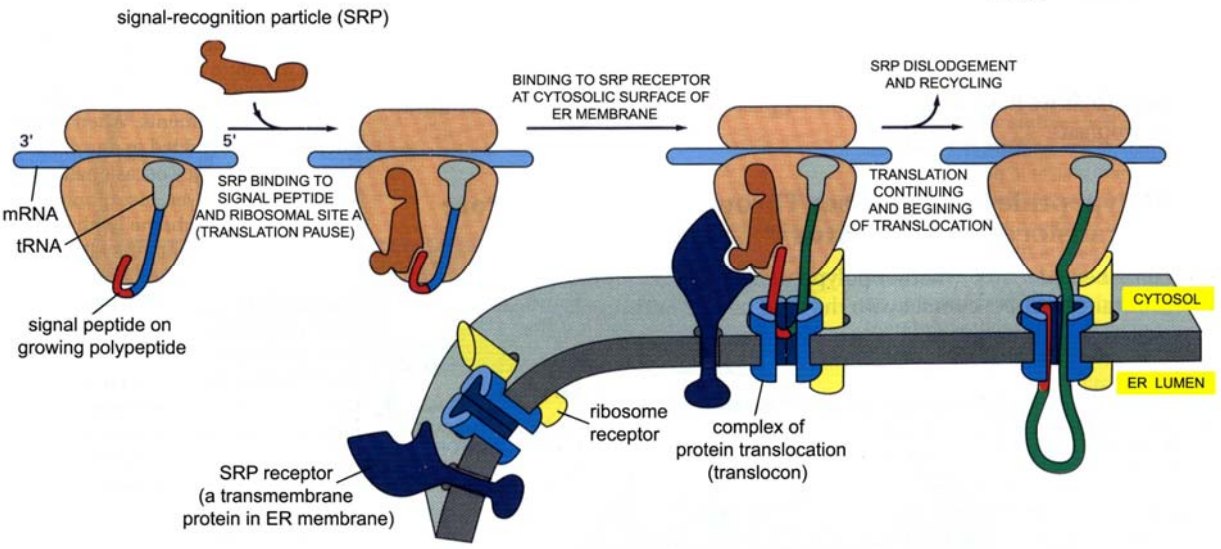
FUNCTION OF SIGNAL SEQUENCE	EXAMPLE OF SIGNAL SEQUENCE
Import into nucleus	-Pro-Pro-Lys-Lys-Lys-Arg-Lys-Val-
Export from nucleus	-Leu-Ala-Leu-Lys-Leu-Ala-Gly-Leu-Asp-Ile-
Import into mitochondria	$^{+}H_3N$-Met-Leu-Ser-Leu-Arg-Gln-Ser-Ile-Arg-Phe-Phe-Lys-Pro-Ala-Thr-Arg-Thr-Leu-Cys-Ser-Ser-Arg-Tyr-Leu-Leu-
Import into plastid	$^{+}H_3N$-Met-Val-Ala-Met-Ala-Met-Ala-Ser-Leu-Gln-Ser-Ser-Met-Ser-Ser-Leu-Ser-Leu-Ser-Ser-Asn-Ser-Phe-Leu-Gly-Gln-Pro-Leu-Ser-Pro-Ile-Thr-Leu-Ser-Pro-Phe-Leu-Gln-Gly-
Import into peroxisomes	-Ser-Lys-Leu-COO^{-}
Import into ER	$^{+}H_3N$-Met-Met-Ser-Phe-Val-Ser-Leu-Leu-Leu-Val-Gly-Ile-Leu-Phe-Trp-Ala-Thr-Glu-Ala-Glu-Gln-Leu-Thr-Lys-Cys-Glu-Val-Phe-Gln-
Return to ER	-Lys-Asp-Glu-Leu-COO^{-}

Some characteristic features of the different classes of signal sequences are highlighted in color. Where they are known to be important for the function of the signal sequence, positively charged amino acids are shown in *red* and negatively charged amino acids are shown in *green*. Similarly, important hydrophobic amino acids are shown in *white* and hydroxylated amino acids are shown in *blue*. $^{+}H_3N$ indicates the N-terminus of a protein; COO^{-} indicates the C-terminus.

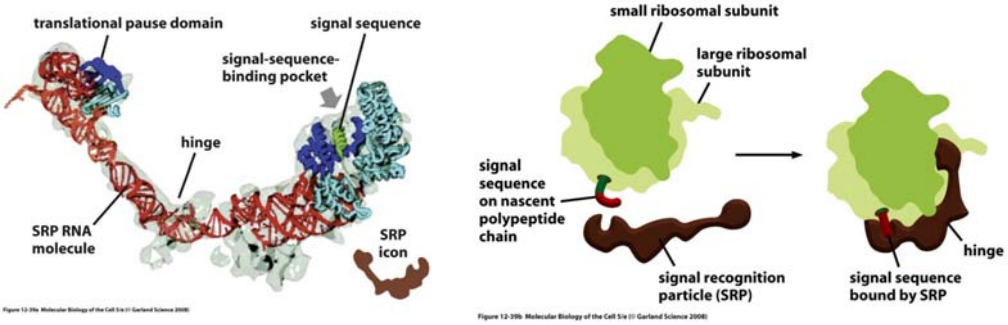
Table 12-3 Molecular Biology of the Cell 5/e (© Garland Science 2008)

Mechanism of protein import by ER and initializing of translocation through ER membrane





Protein recruitment by ER and initializing of translocation through membrane



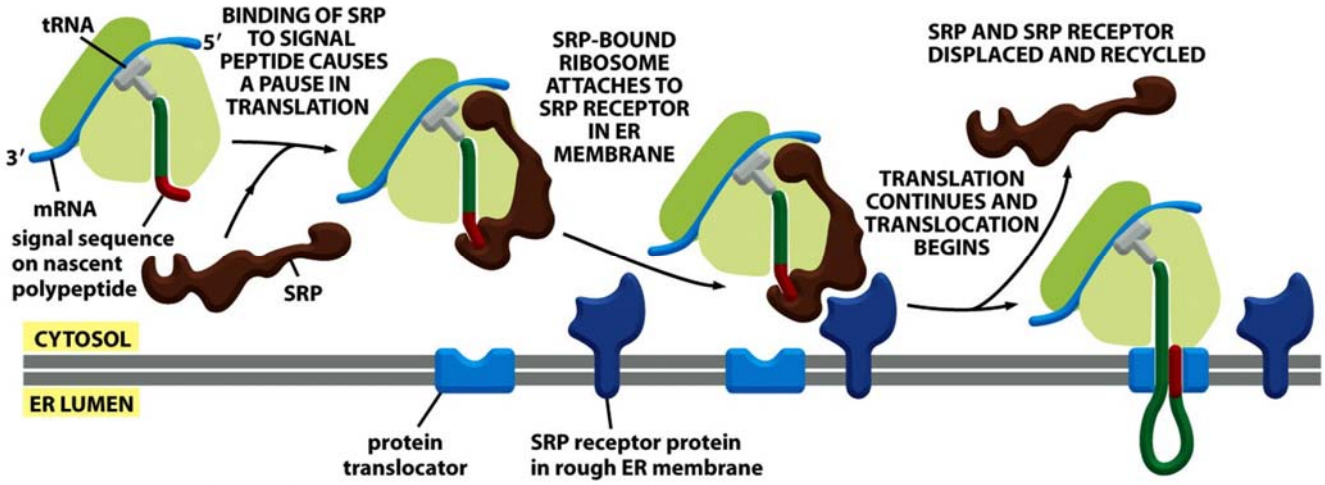


Figure 12-40 Molecular Biology of the Cell 5/e (© Garland Science 2008)

Biosynthesis of exported (secretory) proteins

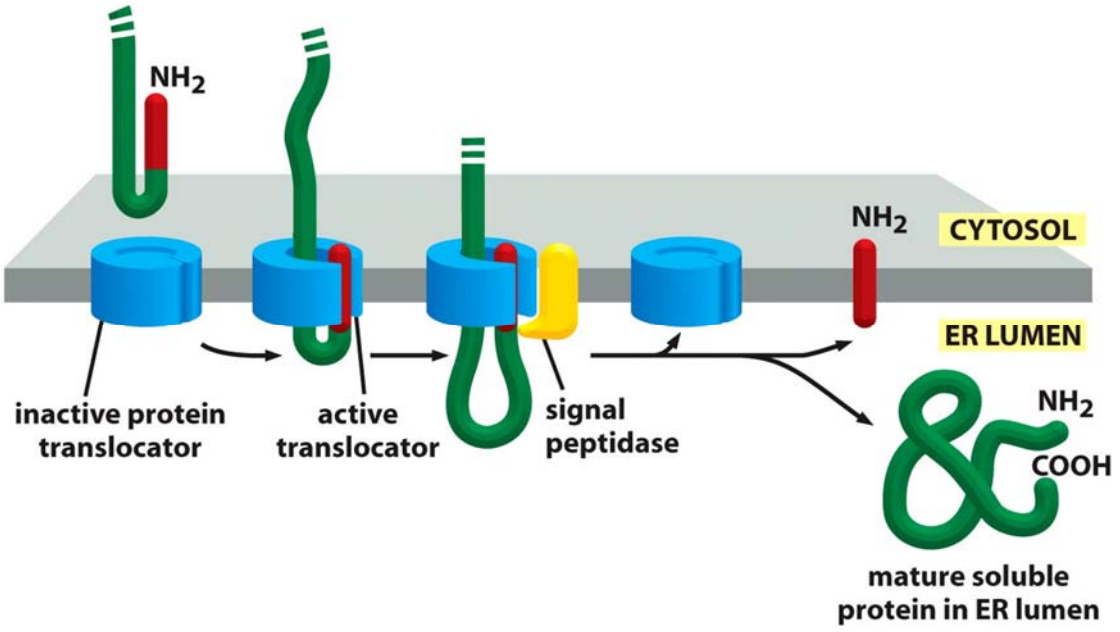


Figure 12-45 Molecular Biology of the Cell 5/e (© Garland Science 2008)

Biosynthesis of single-pass transmembrane proteins ***(i)***

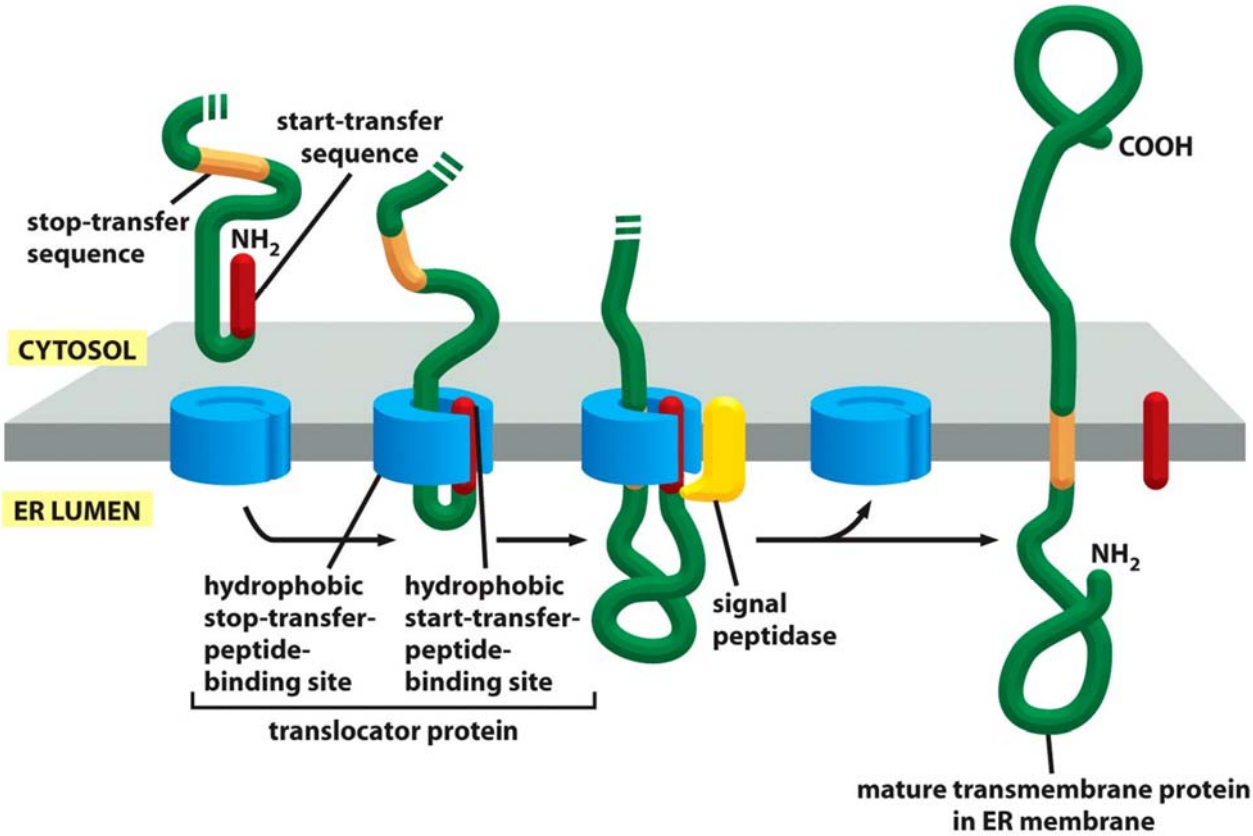


Figure 12-46 Molecular Biology of the Cell 5/e (© Garland Science 2008)

Biosynthesis of single-pass transmembrane proteins *(ii)*

normal insertion

Insertion into the translocon is determined by the electric charges of the polypeptide chain in the proximity of the signal peptide

reverse insertion

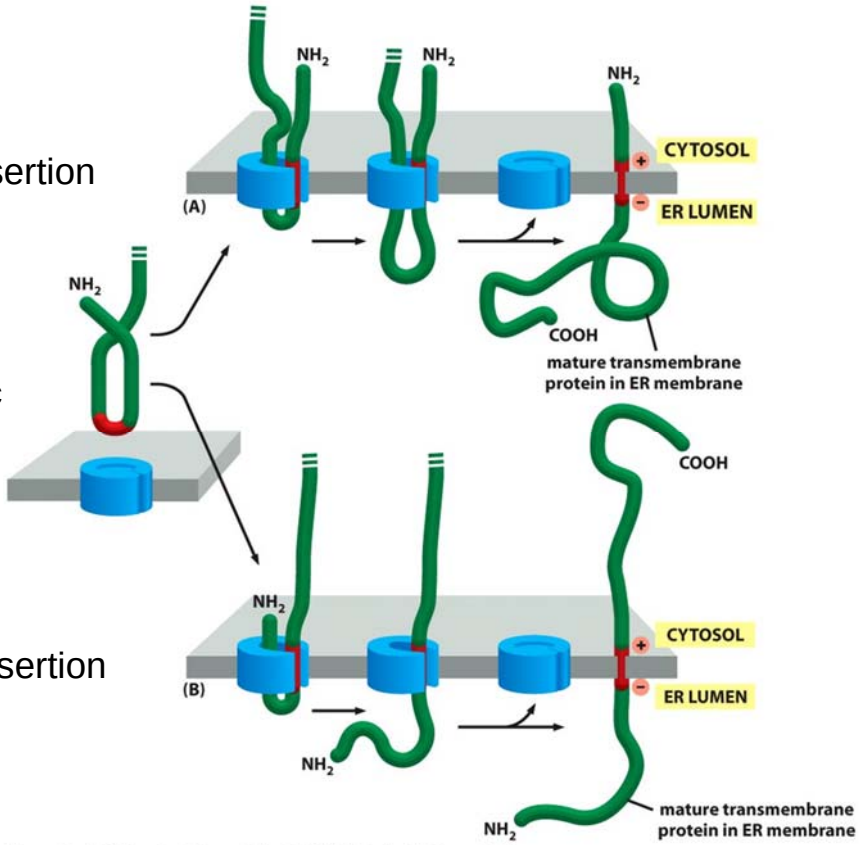


Figure 12-47 Molecular Biology of the Cell 5/e (© Garland Science 2008)

Biosynthesis of multi-pass transmembrane proteins

Multiple hydrophobic sequences in the polypeptide chain of nascent protein
Odd sequences – start transfer information
Even sequences – stop transfer information

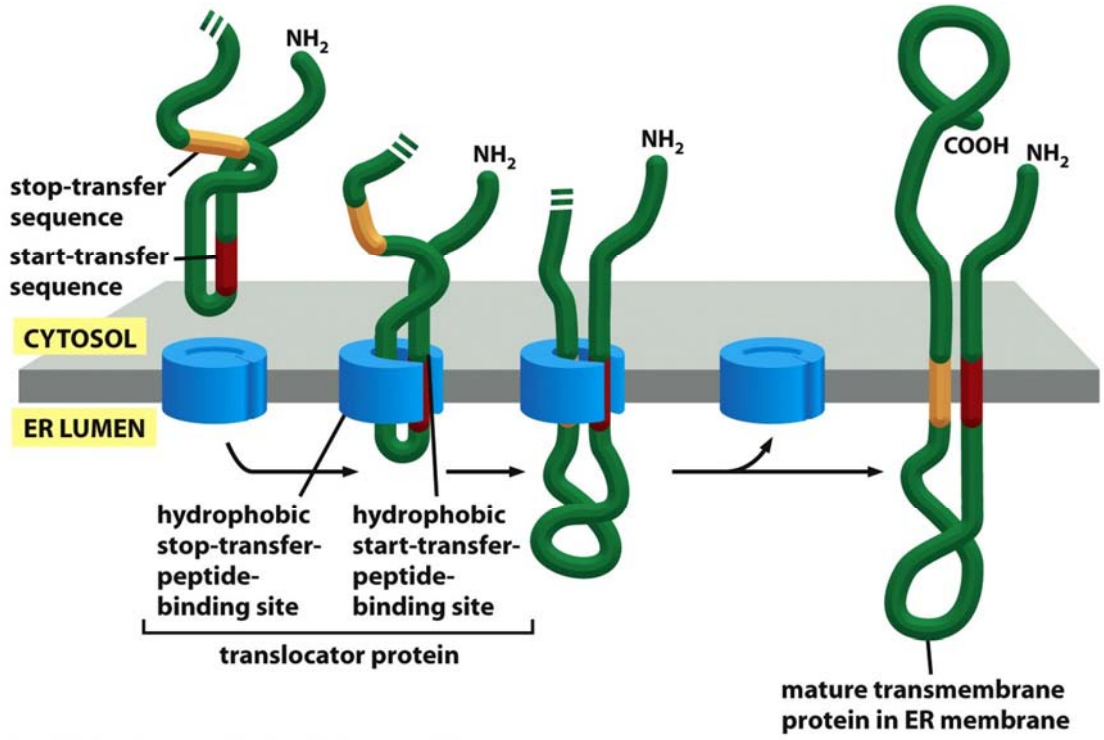


Figure 12-48 Molecular Biology of the Cell 5/e (© Garland Science 2008)

Protein processing

Glycosylation on asparagine (*N*-linked oligosaccharide formation)

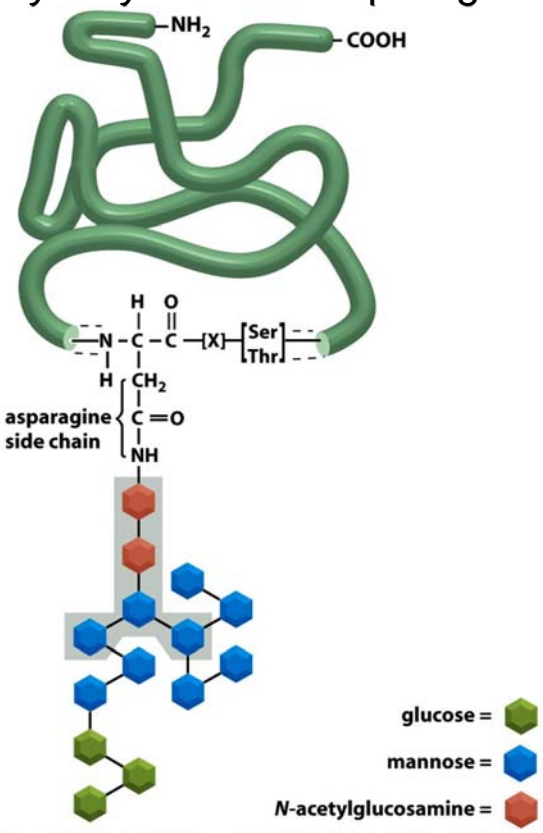


Figure 12-50 Molecular Biology of the Cell 5/e (© Garland Science 2008)

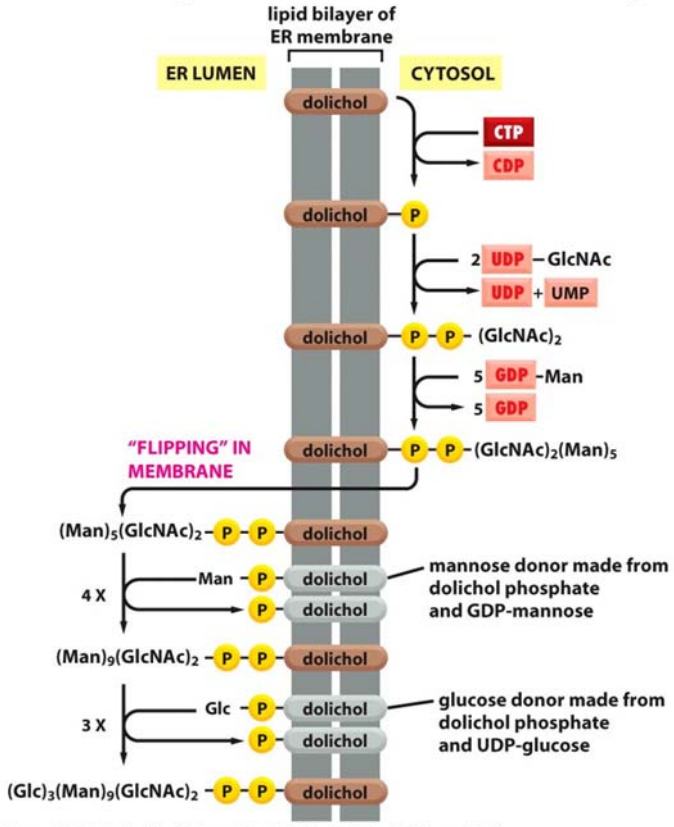


Figure 12-52 Molecular Biology of the Cell 5/e (© Garland Science 2008)

Protein processing

Glycosylation on asparagine (*N*-linked oligosaccharide formation)

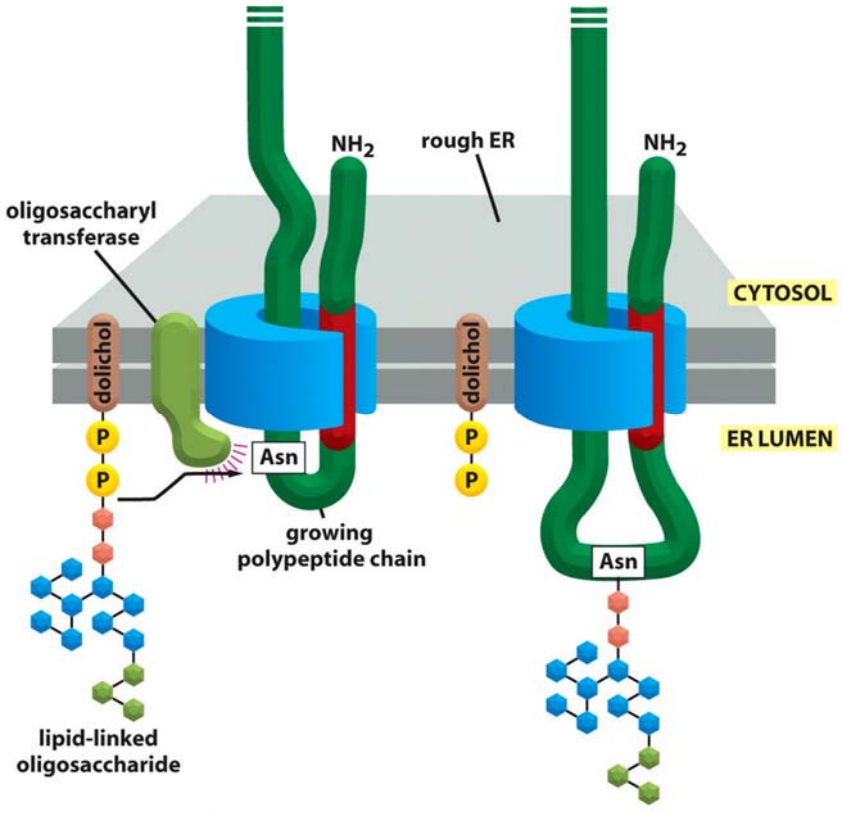


Figure 12-51 Molecular Biology of the Cell 5/e (© Garland Science 2008)

Polypeptide Chain Processing in ER

- Co-translational processing:
  - *(i)* starting protein glycosylation (*N*-glycosidic linkages);
  - *(ii)* proline (position 4) and lysine (position 5) hydroxylation; enzyme (prolyl-4-hydroxylase) is a heterotetramer $\alpha_2\beta_2$, with β subunit identical to PDI;
  - *(iii)* carboxylation of glutamic acid side chain (position γ); enzyme is a transmembrane protein with the active site at luminal surface.

- Post-translational processing:
  - *(i)* trimming of *N*-glycosidic oligosaccharides;
  - *(ii)* right protein folding and assembly (chaperons);
  - *(iii)* glypiation (glycosylphosphatidylinositol-anchor, GPI-anchor).

Protein processing

Trimming of N-linked oligosaccharide

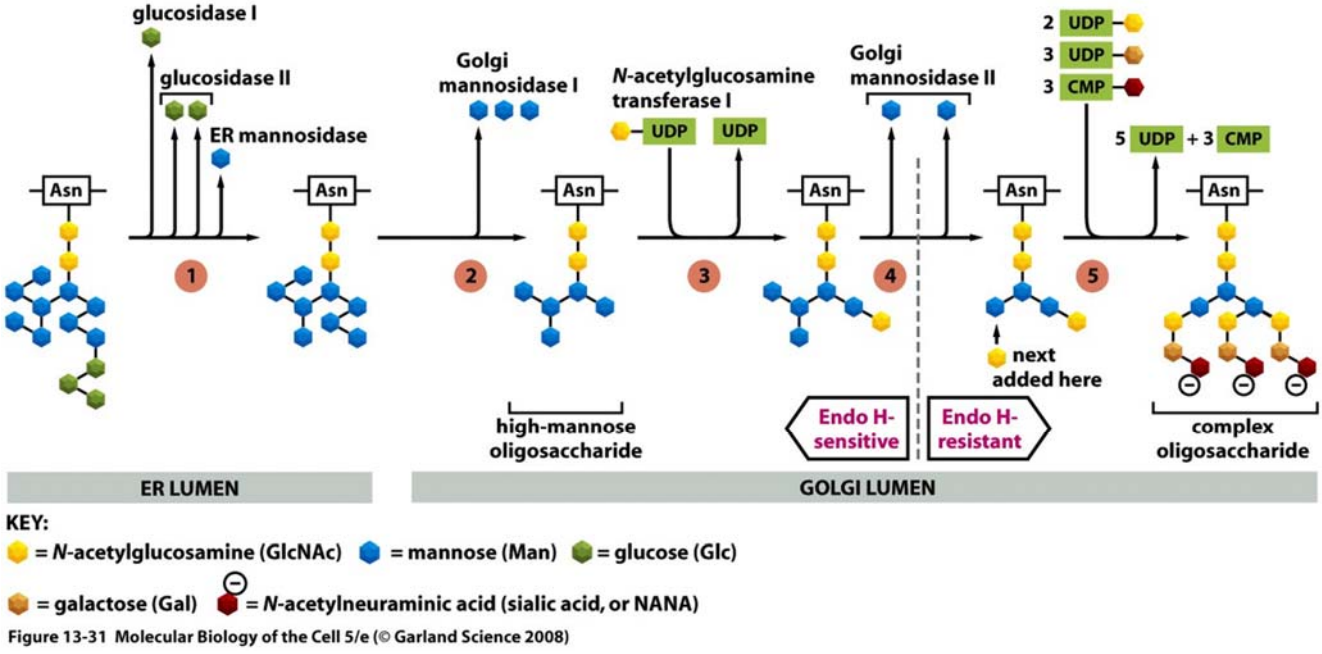


Figure 13-31 Molecular Biology of the Cell 5/e (© Garland Science 2008)

Protein processing

Assistance of glycoprotein folding

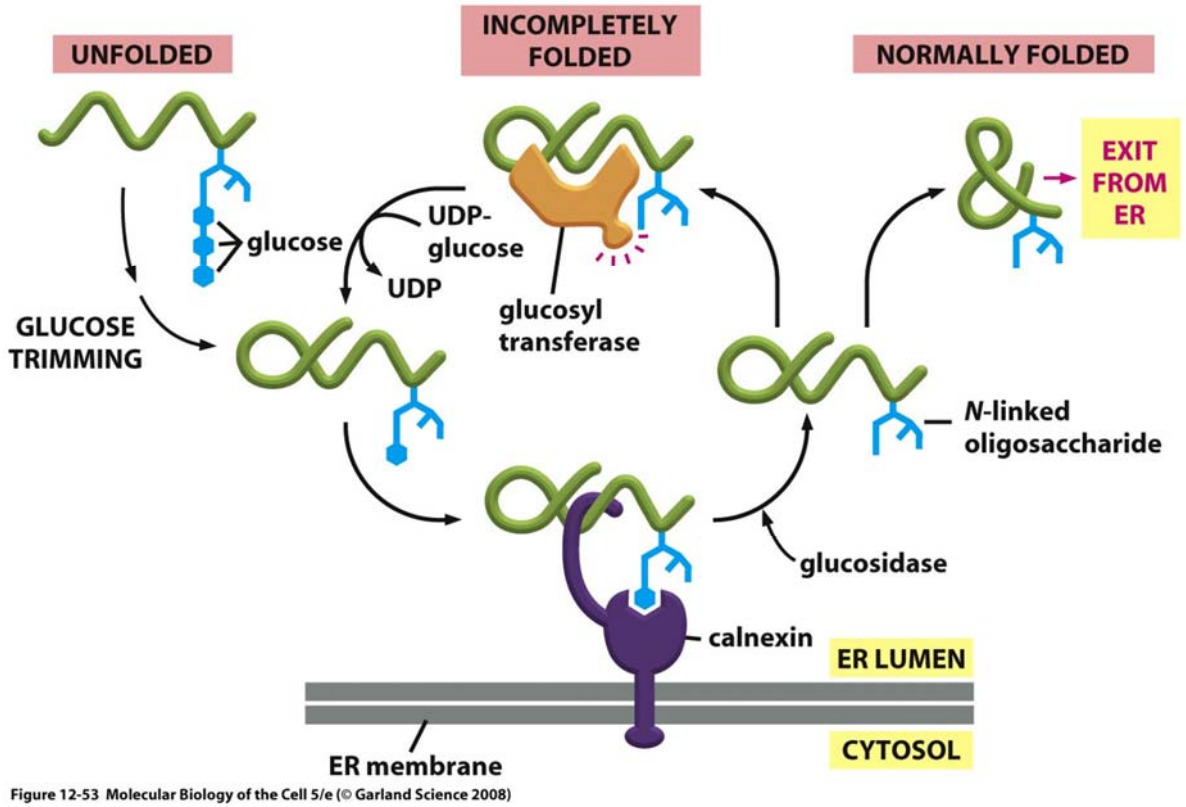
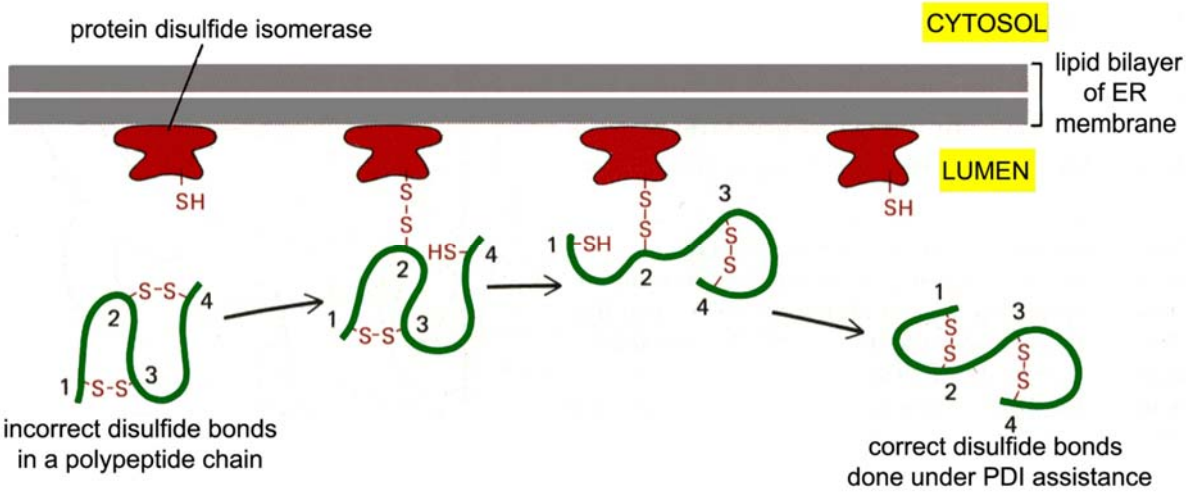


Figure 12-53 Molecular Biology of the Cell 5/e (© Garland Science 2008)

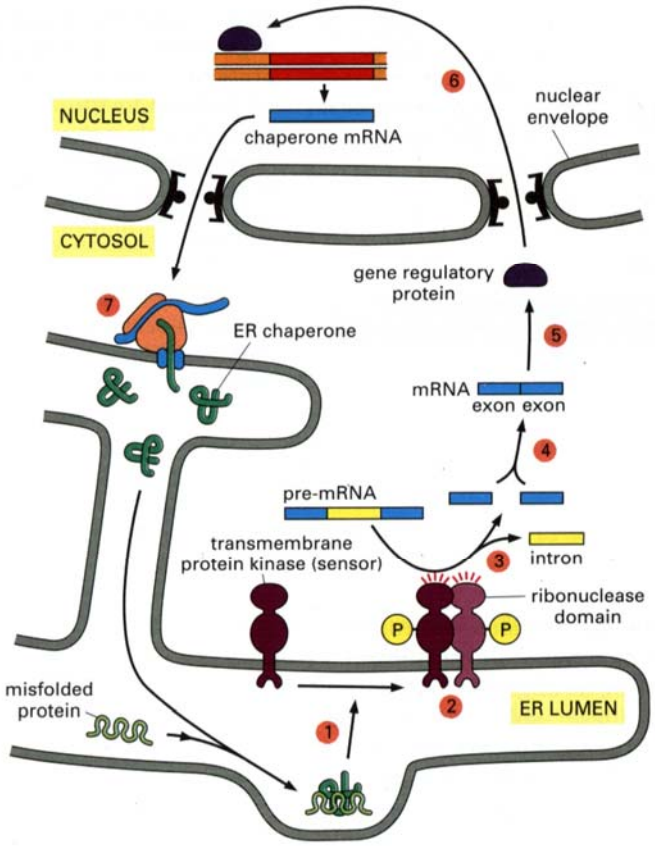
Chaperones in ER

- Calnexin – acts by a lectin-like mechanism
- Protein disulfide isomerase (PDI) – assists in forming of right disulfide bridges

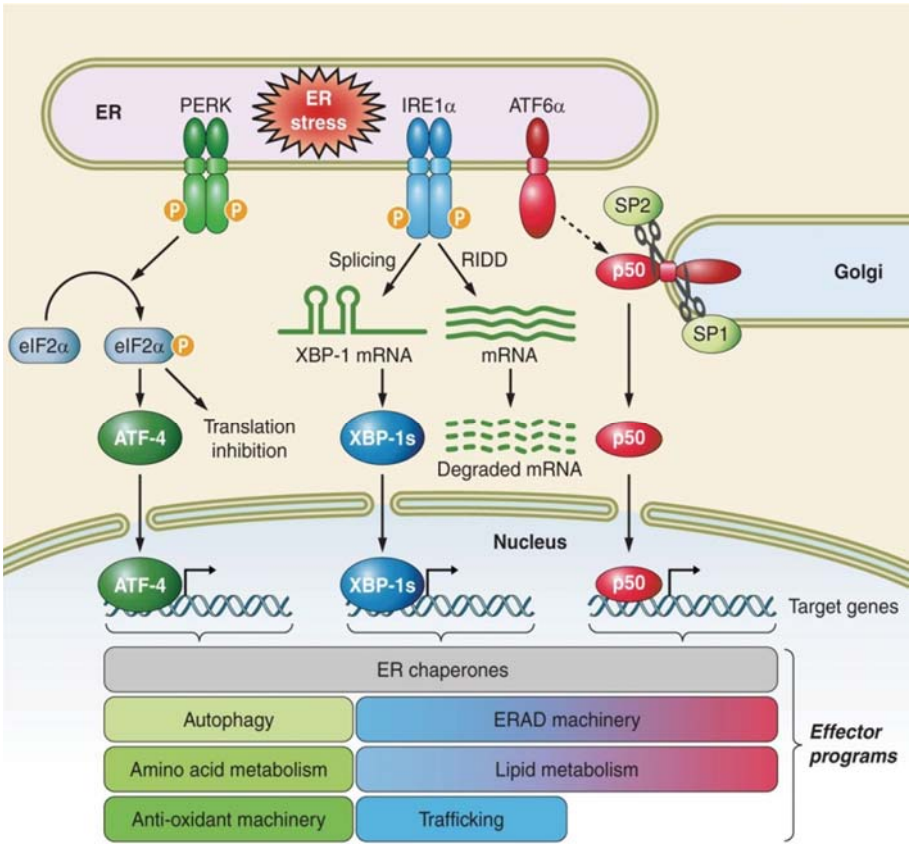


- Binding protein (BiP) – belonging to Hsp70 family of chaperones

Control of chaperone level in ER



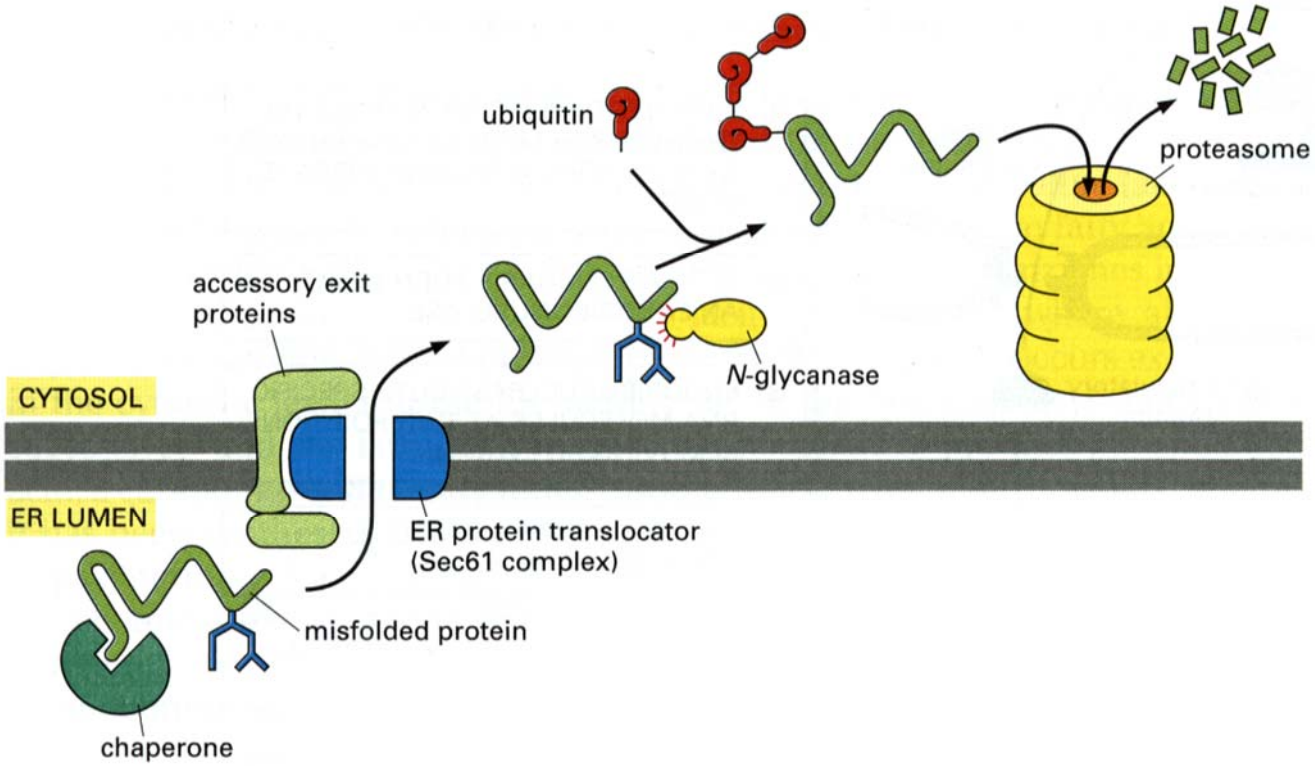
ER Stress



Legend:
PERK – „***P**ancreatic **ER** **K**inase*"
IRE1α – „***I**nositol-**R**equiring **E**nzyme **1α***"
ATF6α – „***A**ctivation **T**ranscription **F**actor **6α***"
eIF2α – „***e**ukaryotic **I**nitiation **F**actor **2α***"
RIDD – „**R**egulated **I**RE1-**D**ependent **D**egradation"
SP1(2) – „***S**ite-**1(2)** **P**rotease*"
XBP-1 – „***X**-**B**ox **P**rotein **1***"

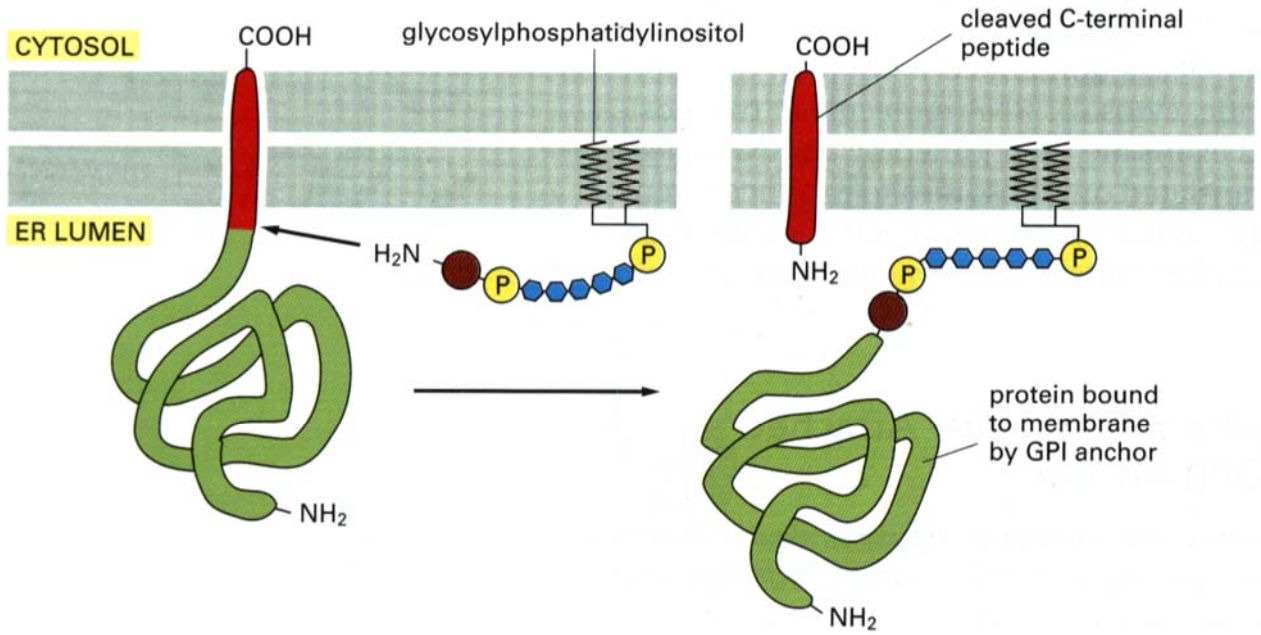
From: Pluquet O, Pourtier A, Abbadie C. *Am J Physiol Cell Physiol*. 2015; 308(6): C415-25. doi: 10.1152/ajpcell.00334.2014.

Retro-translocation and degradation of misfolded proteins

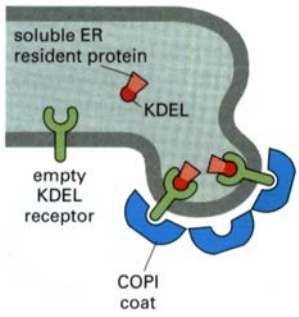


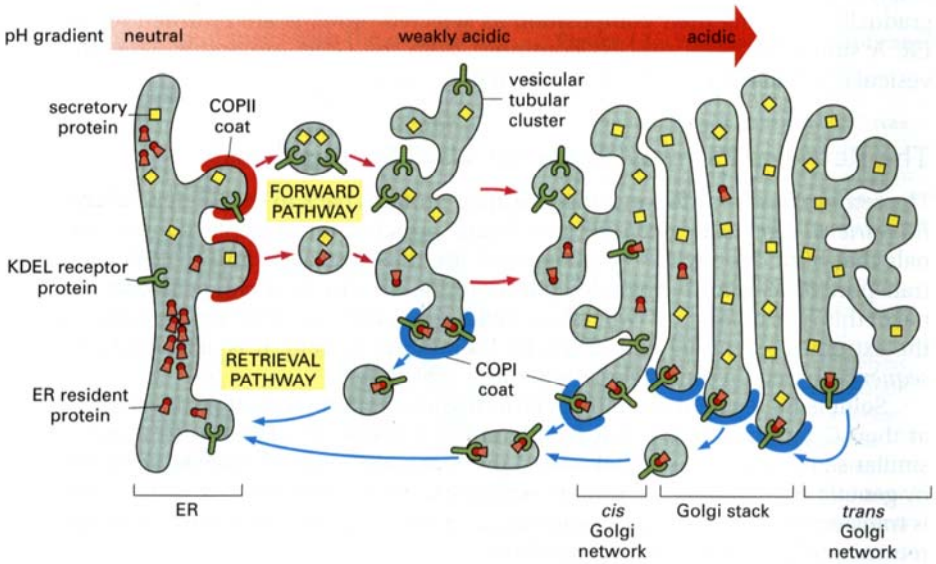
Processing of biosynthetized proteins

Glypiation (addition of glycosylphosphatidylinositol anchor)

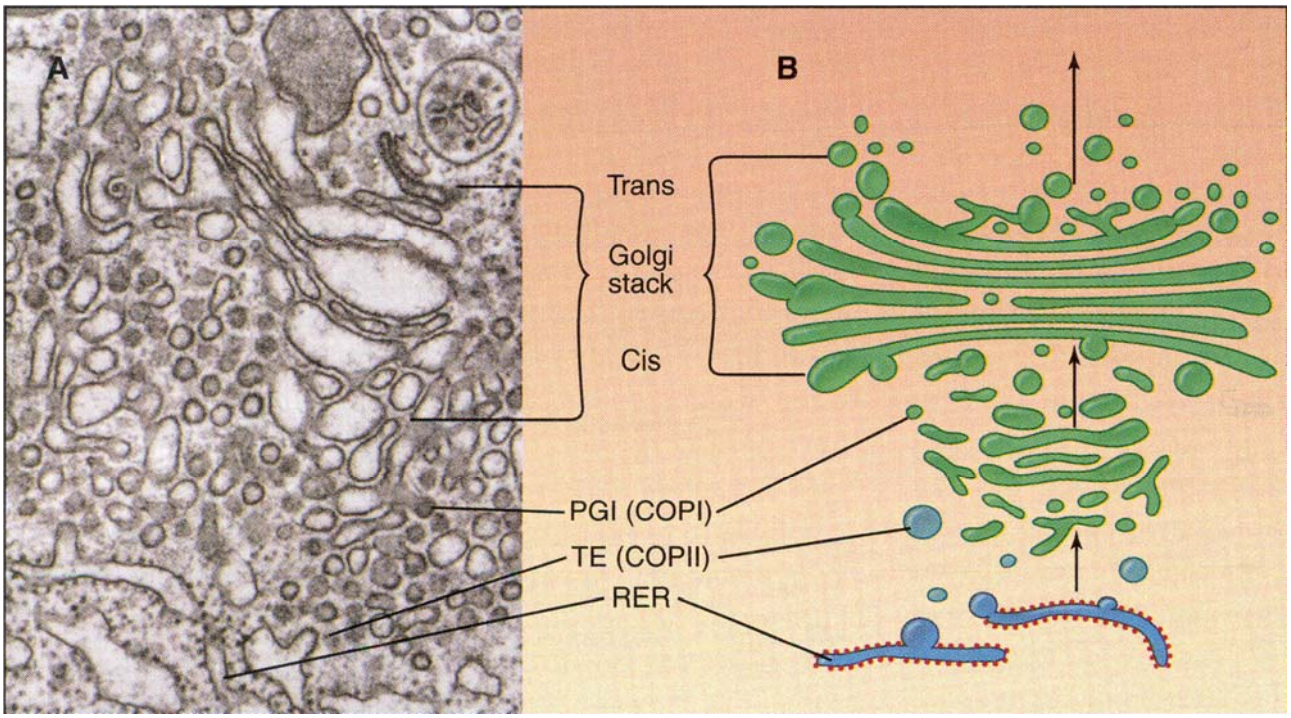


Protein sorting and transport to Golgi



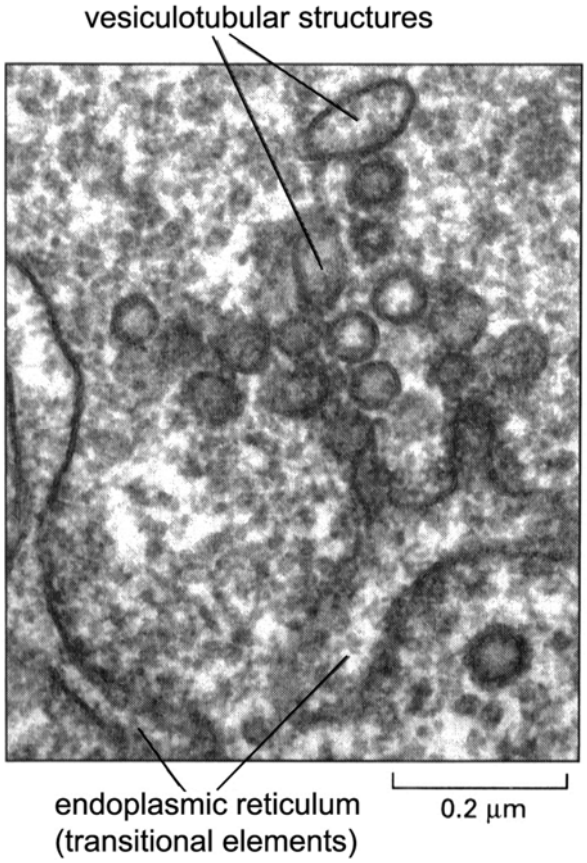


Relationships between ER and Golgi Complex



Add a new ultrastructural element of ER: transitional ER (represents ER exit sites, where vesicles are budding to travel toward Golgi complex)

Vesiculotubular Structures



Summary

- ER is an organelle acting in membrane biogenesis and intracellular membrane traffic;
- Main functions of ER are:
  - Lipid biosynthesis for SER
  - Protein biosynthesis (membrane proteins, secretory proteins, proteins of ER, Golgi and lysosome lumina) for RER
- ER acts in protein folding and assembly by chaperones;
- ER cooperates with both ribosomes and Golgi complex to effectively accomplish its functions.