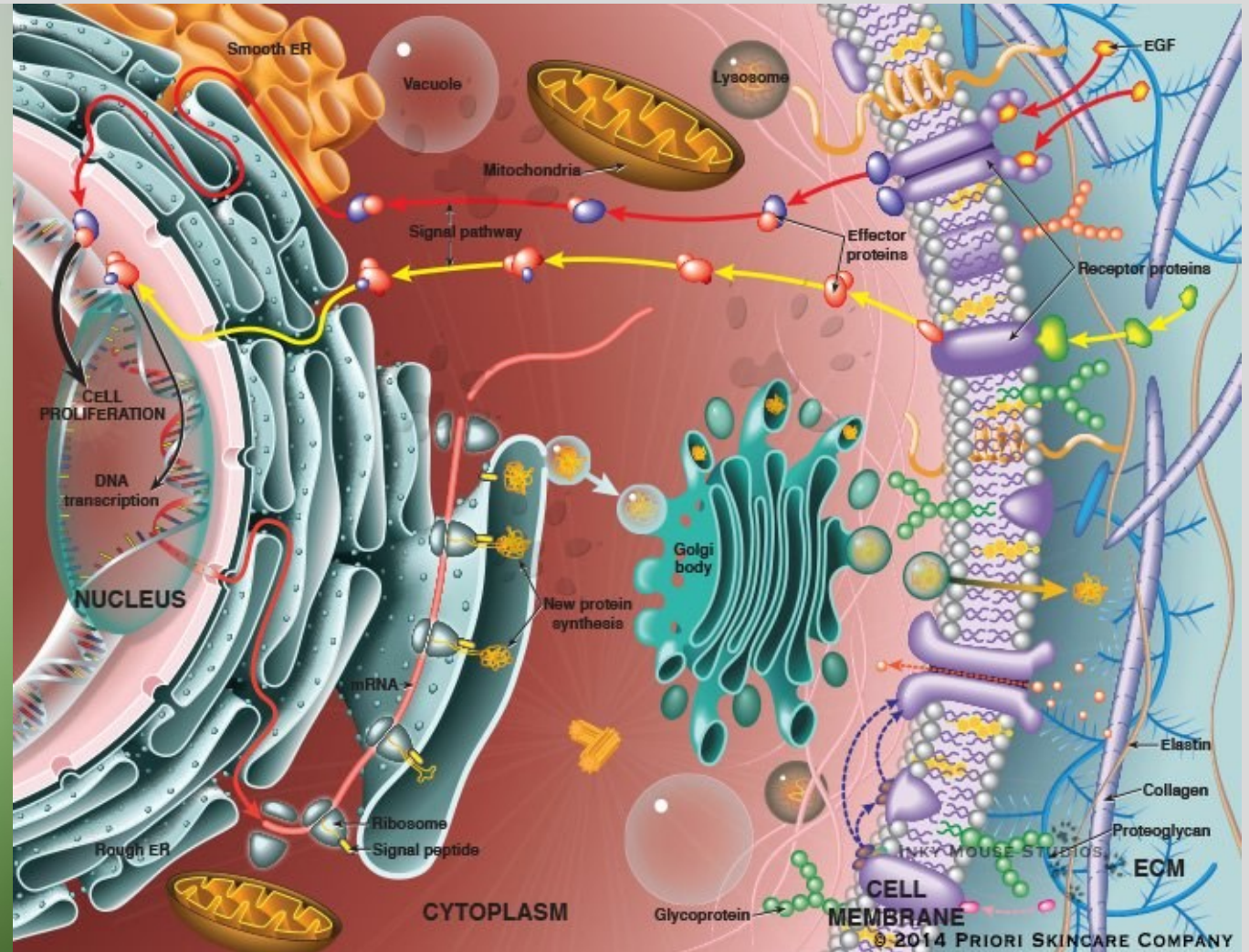
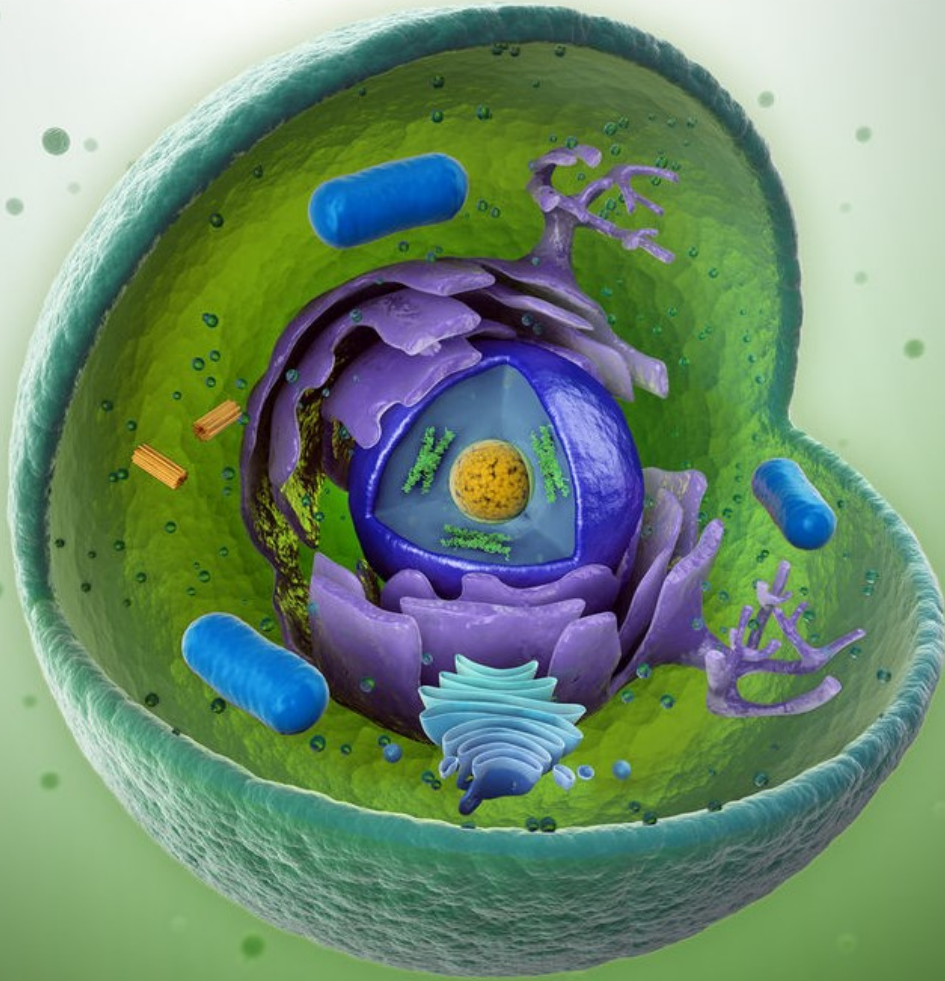
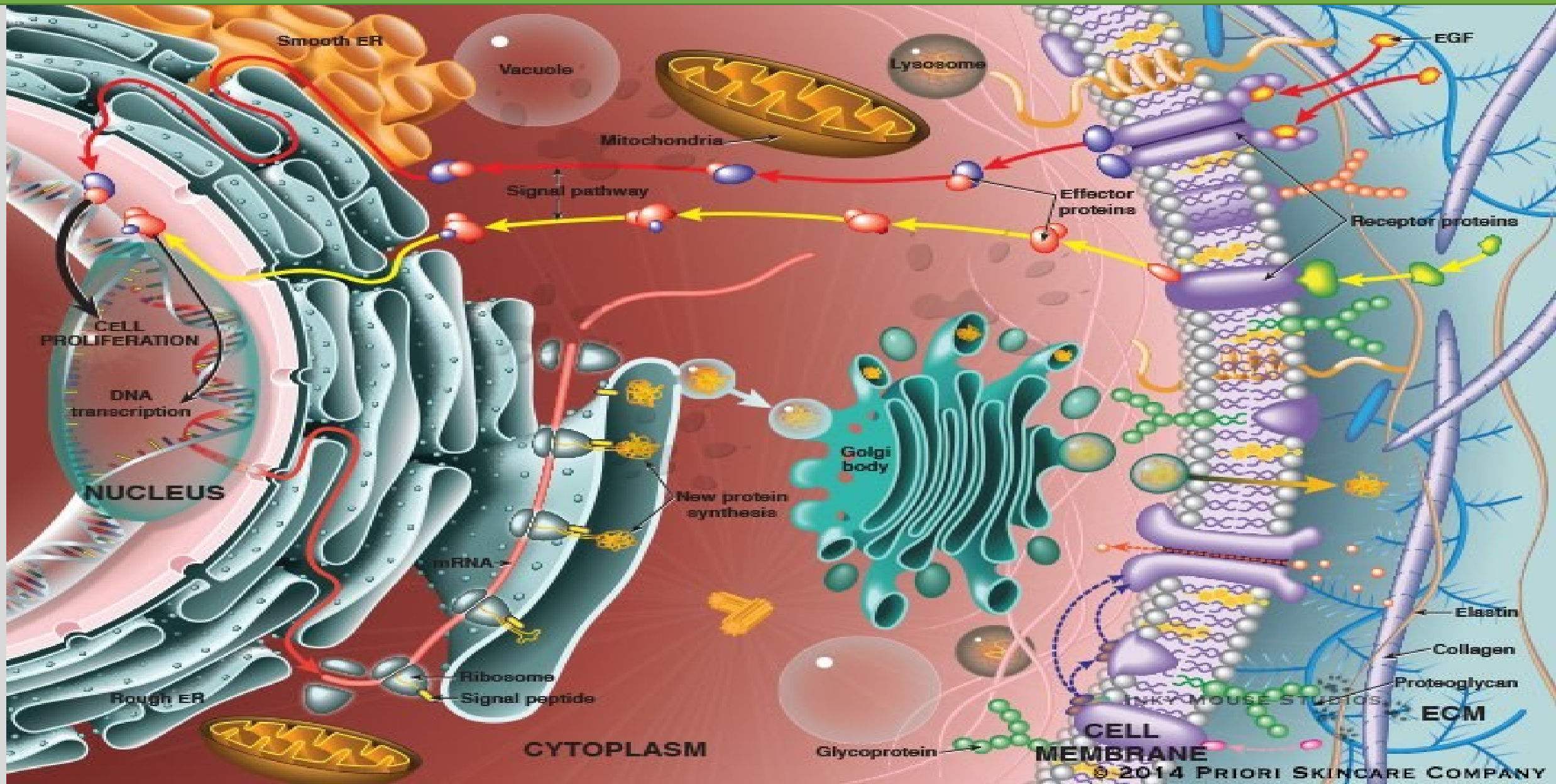


ZOO 529 Animal Cell Biology

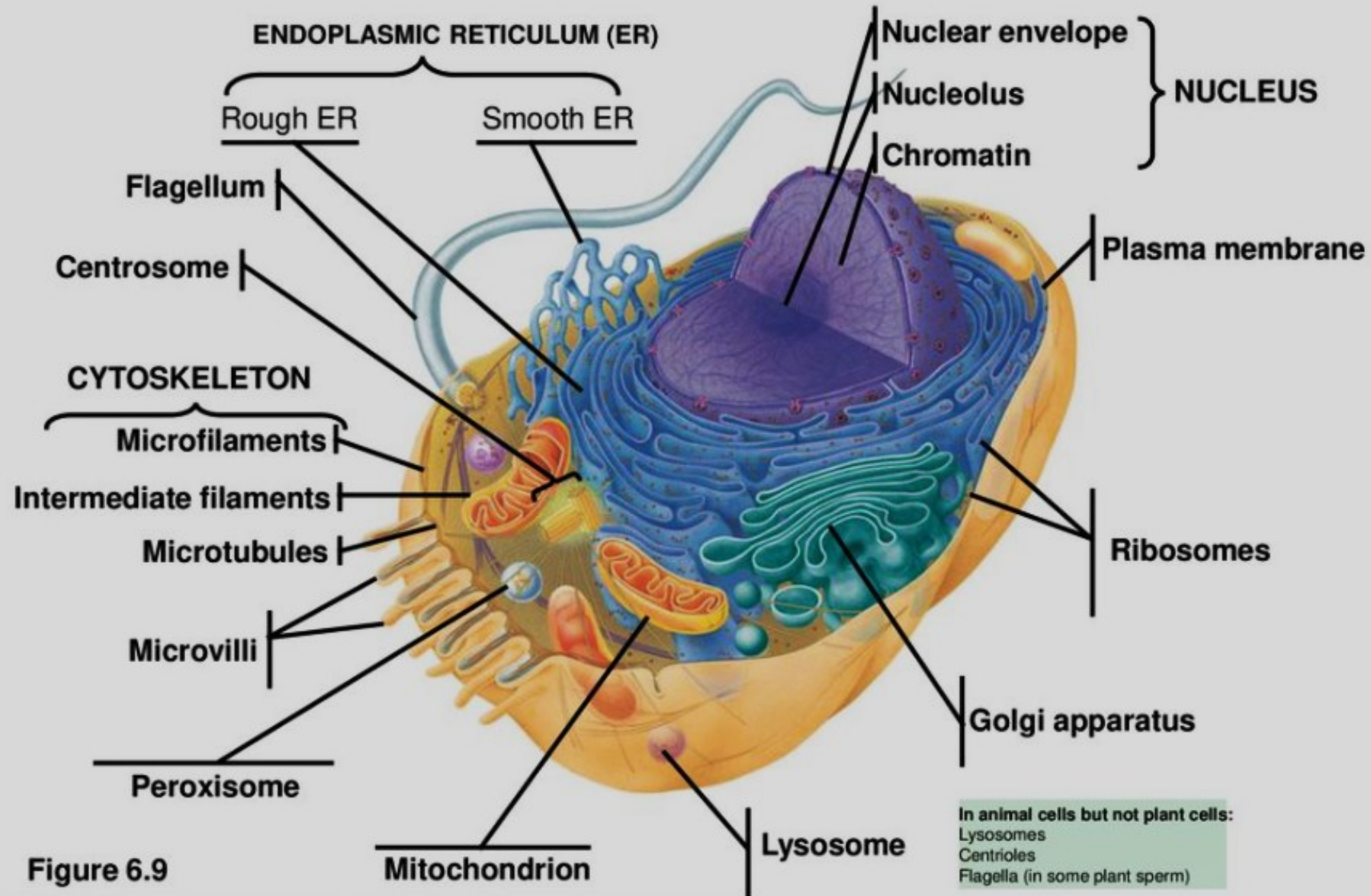
Lecture 16-17



Endomembrane system:



Endomembrane system (EMS)



Endomembrane: Review

- The endomembrane system
 - Is a complex and dynamic player in the cell's compartmental organization
- Relationships among organelles of the endomembrane system:

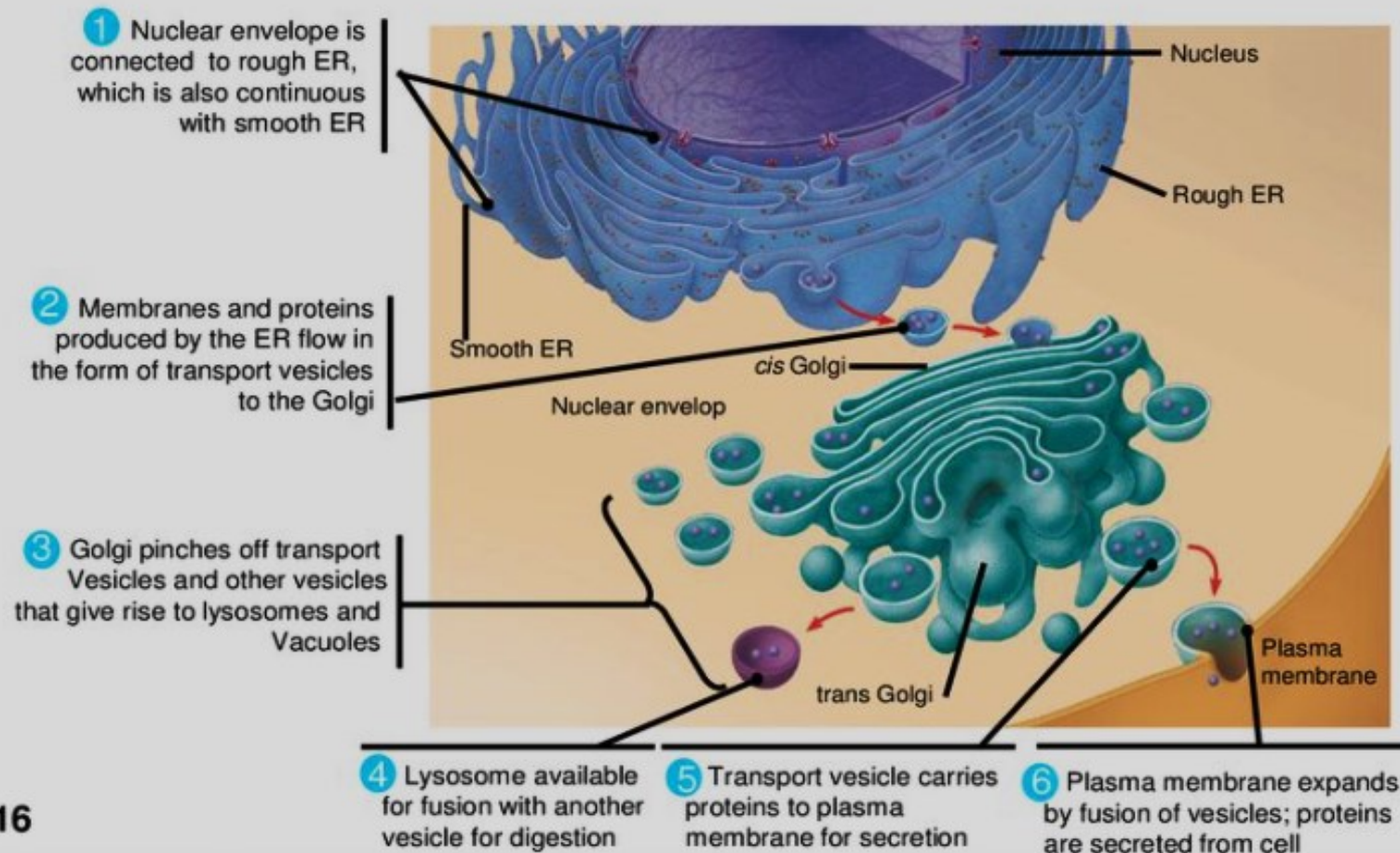
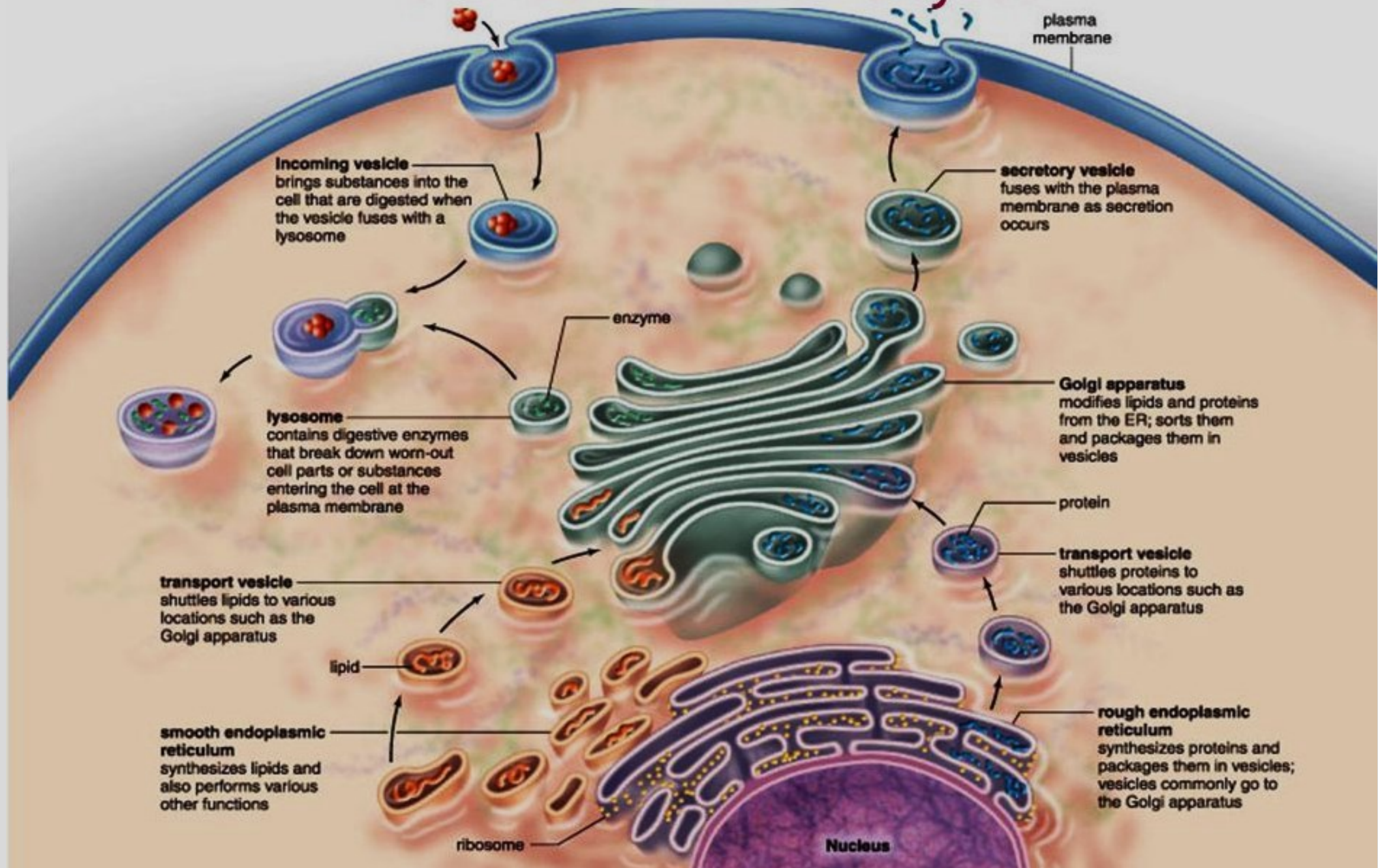


Figure 6.16

The Endomembrane system



What is Endomembrane system?

The endomembrane (endo means within) system is a set of membranes in eukaryotic cells spreading from the nuclear membrane to the plasma membrane.

These membranes divide the cell into different functional and structural compartments or organelles.

The components of the system work together to modify, package and transport lipids and proteins.

The system is defined as a set of membranes that give rise to a single functional and developmental unit, either directly connected or exchanging material through vesicular transport.

Functions of EMS

The endomembrane system is an internal membrane system within the cell that carries out a variety of functions such as:

- ✓ Synthesis of proteins and their transport
- ✓ Metabolism and movement of lipids
- ✓ The detoxification of poison

Here, the membranes are either directly in contact with each other or can communicate through the formation of vesicles.

Components of EMS

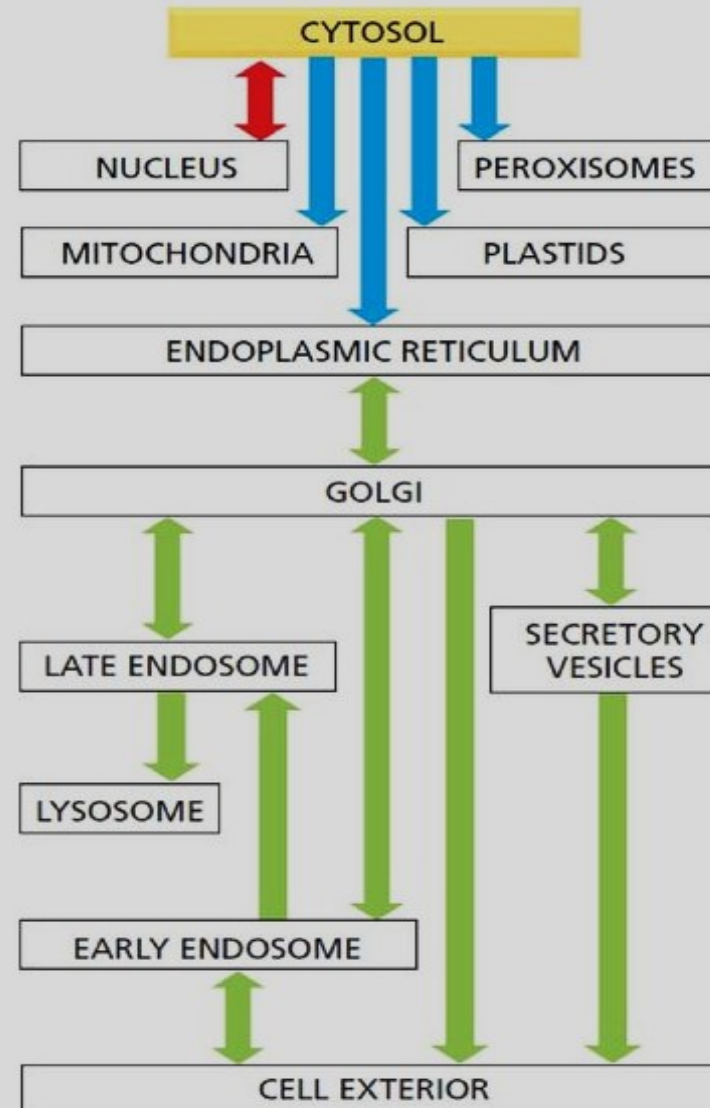
The endomembrane system comprises seven different parts or components of a typical **eukaryotic cell**. They are sequentially listed below in the order of protein and lipid secretion in a cell.

- 1 The Nuclear Envelope
- 2 The Endoplasmic Reticulum
- 3 The Golgi Apparatus
- 4 Lysosomes
- 5 Endosomes
- 6 Transport Vesicles
- 7 The Plasma Membrane

In the subsequent section below, we will deal with each of them and explain how they help regulate proteins and lipids.

Main Vesicular transport pathways

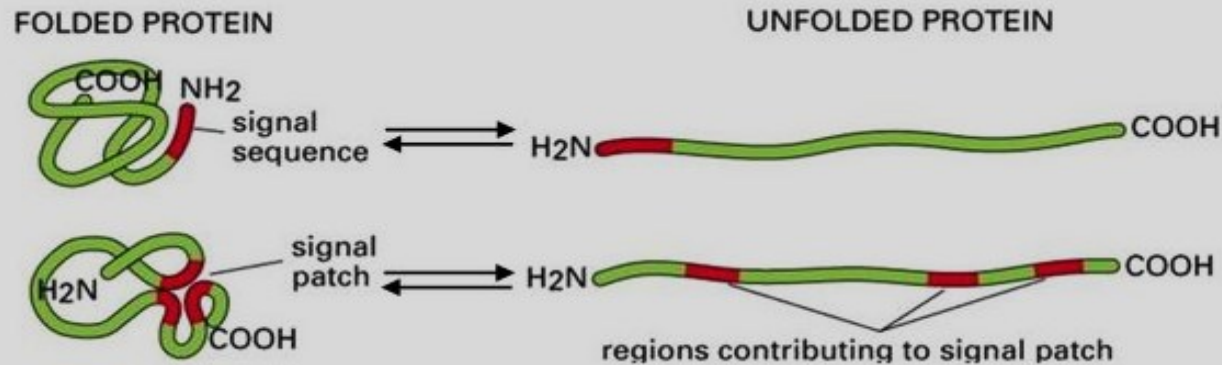
“Roadmap” of protein traffic



Main Vesicular transport pathways

Key components of the protein transport

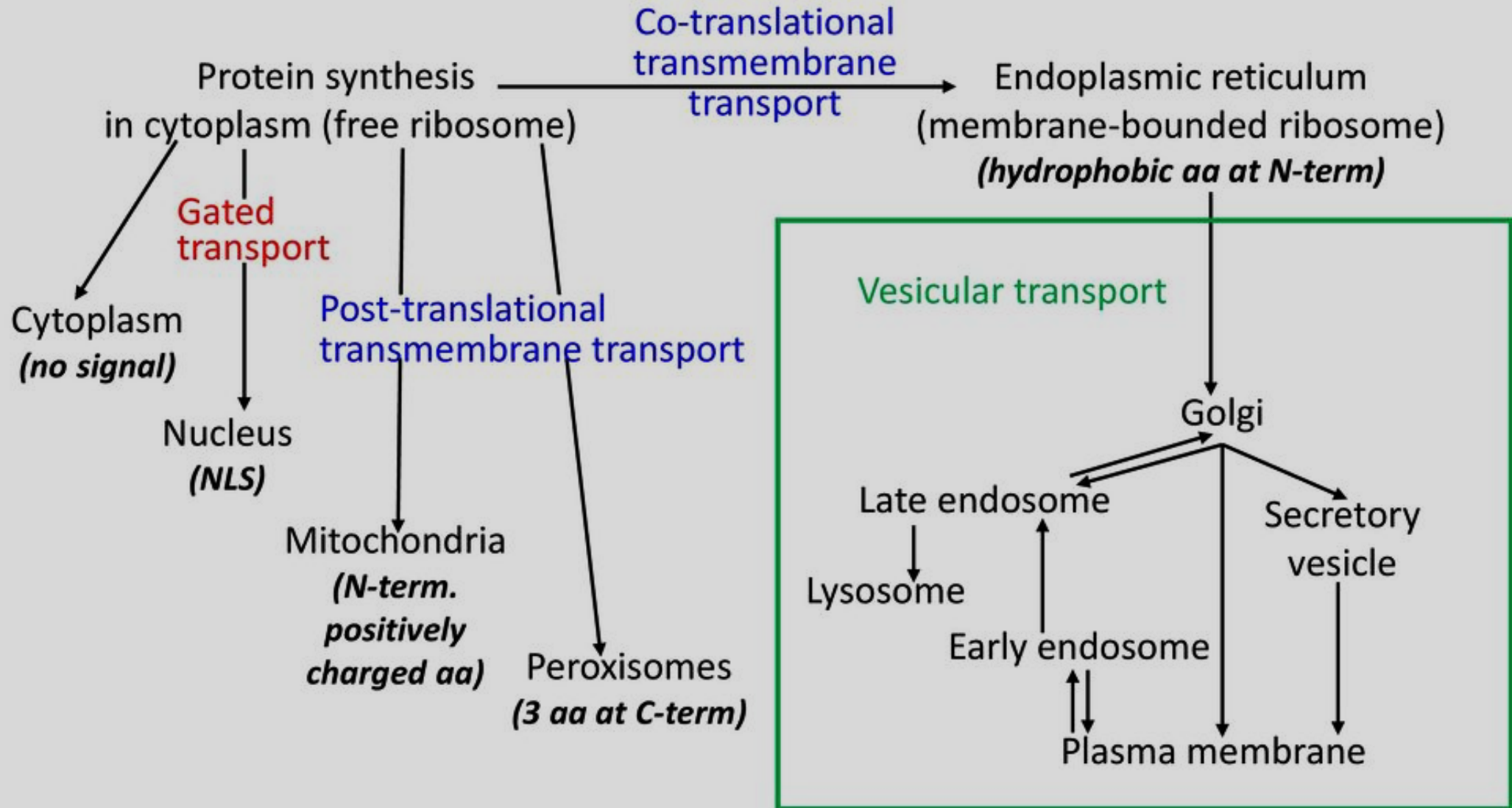
1. Sorting signal



2. Receptors: recognize sorting signal and guide proteins to their appropriate destination
3. Way of protein transfer
 - Gated transport through the nuclear pore
 - Translocation across the membrane (transmembrane protein translocator = translocon)
 - Vesicular transport
4. Energy

Main Vesicular transport pathways

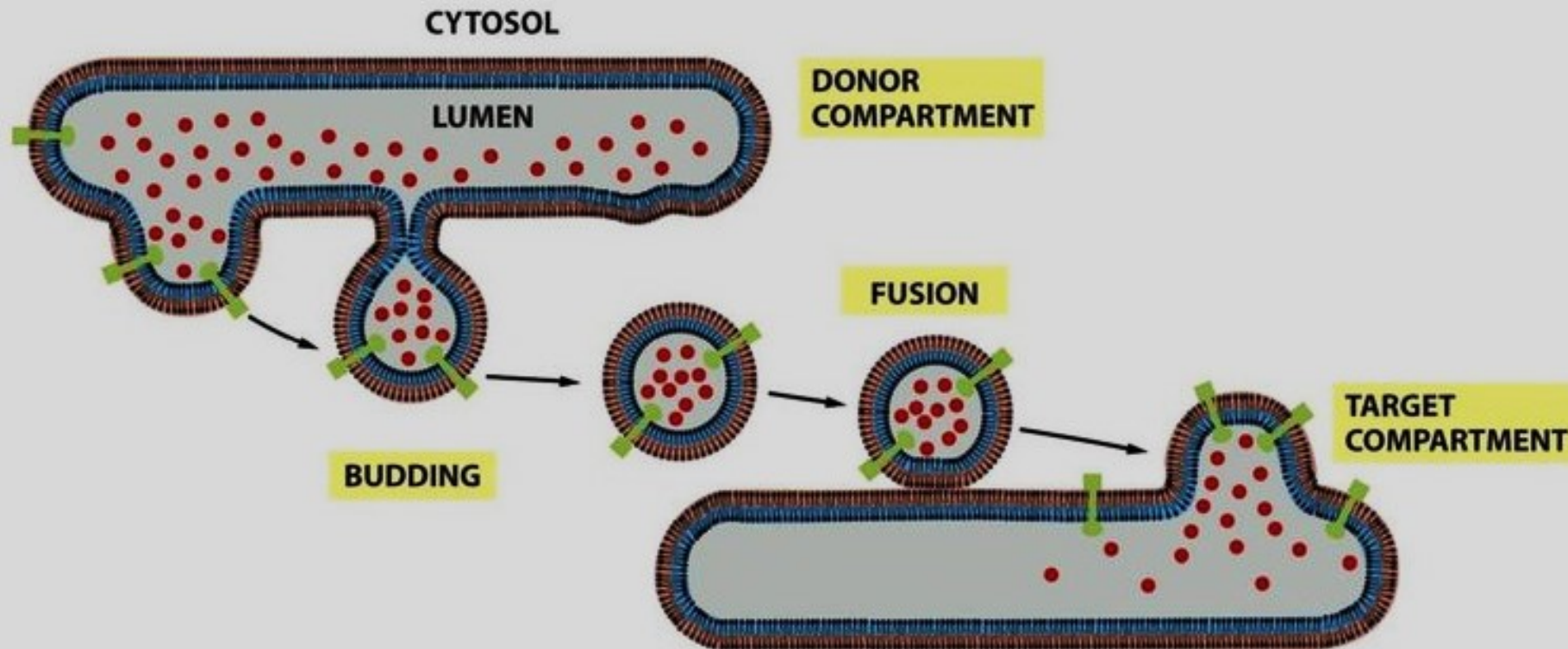
Protein sorting



Main Vesicular transport pathways

Vesicular transport

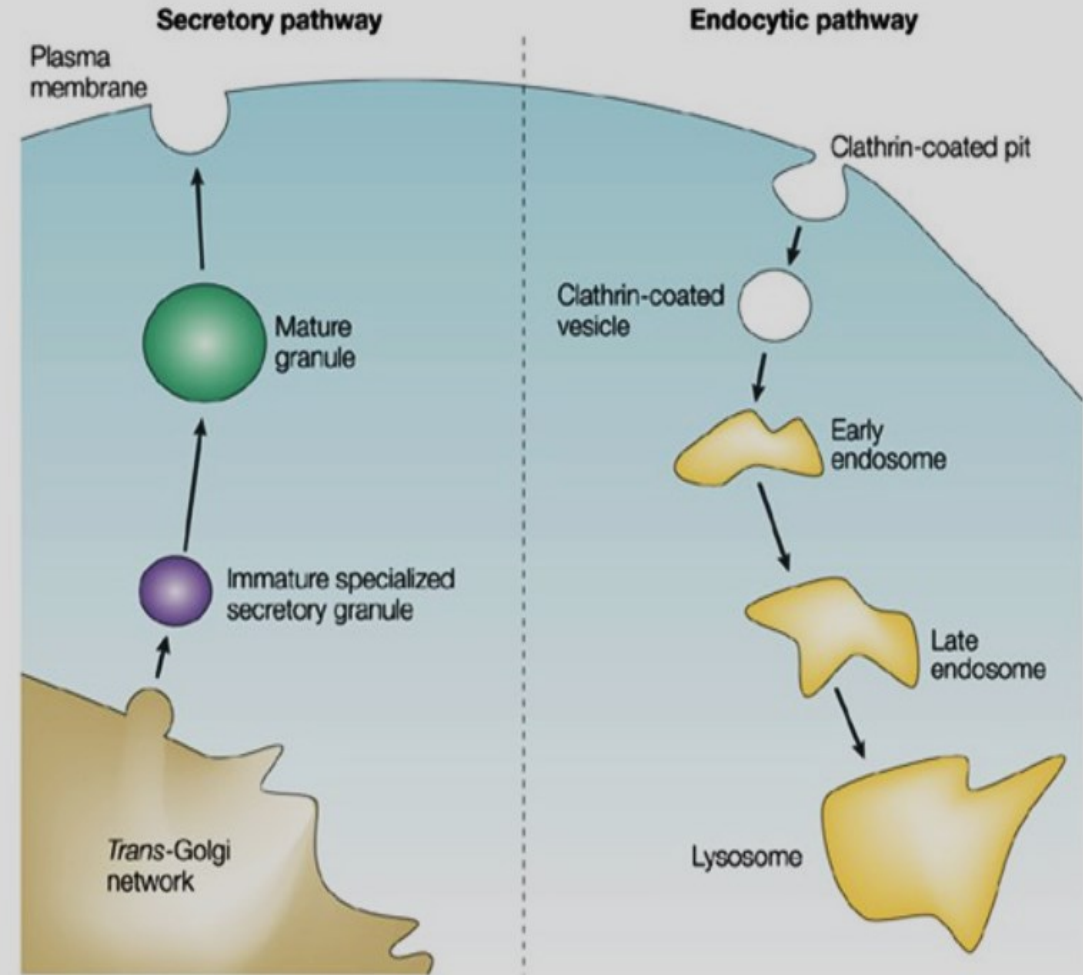
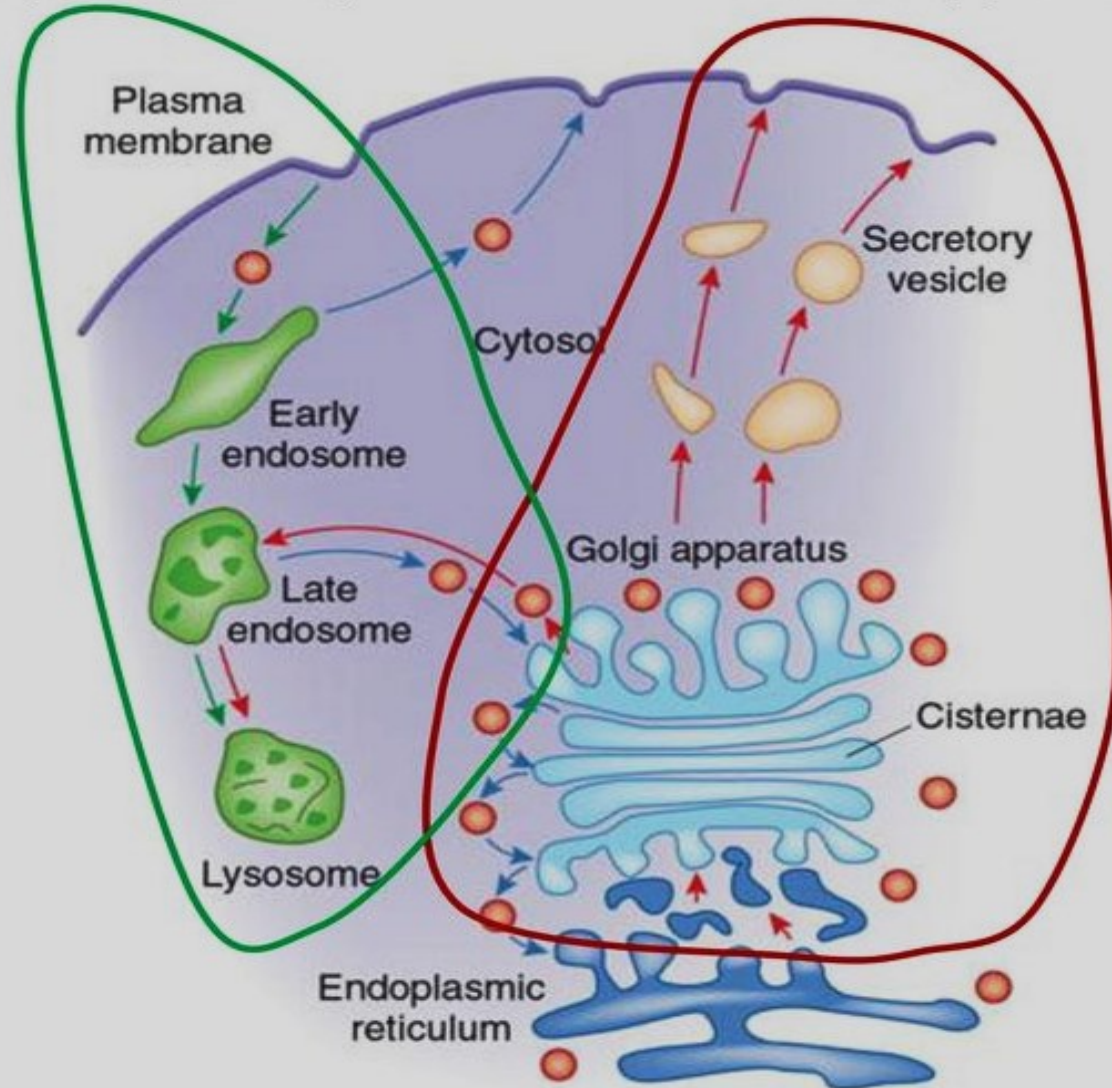
- transport between membrane-enclosed compartments
- transport of macromolecules (soluble and membrane-bound) from the donor compartment to the target compartment



Main Vesicular transport pathways

Inward transport
Endocytotic pathway

Outward transport
Secretory pathway



Main Vesicular transport pathways

Golgi apparatus



Camillo Golgi
(1843-1926)

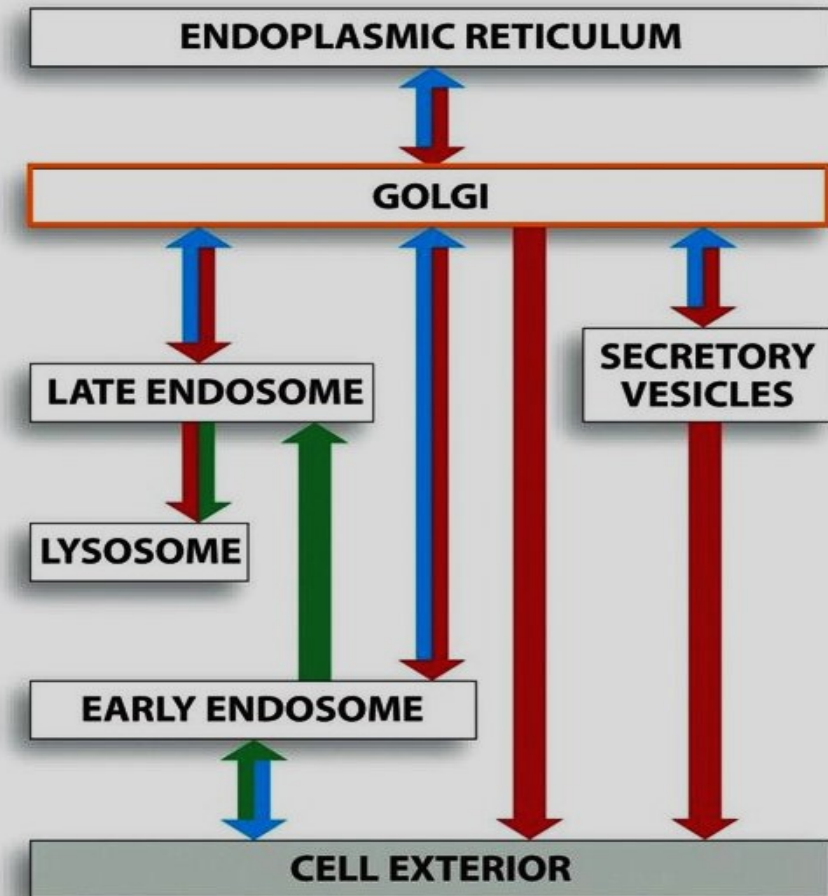
Nobel prize 1906

"internal reticular apparatus"



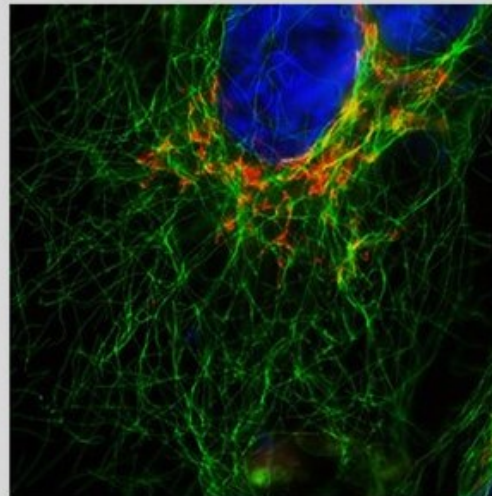
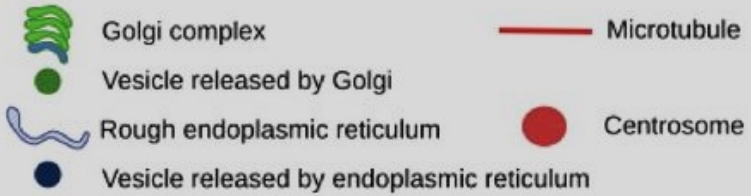
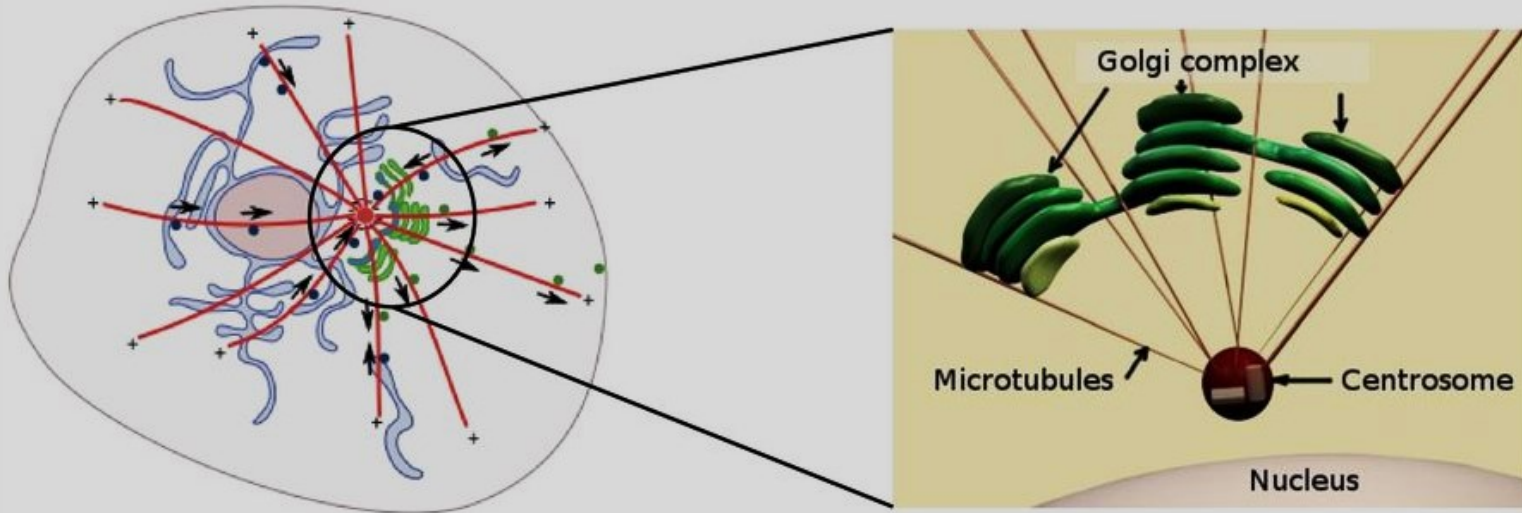
Metal impregnation

Golgi – „Traffic manager” of the cell



Golgi location

Position

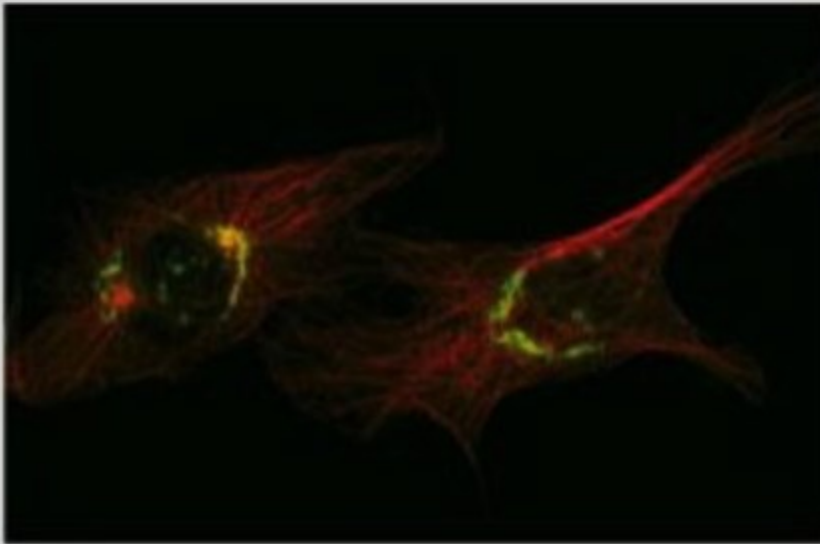


Nucleus
Microtubule
Golgi

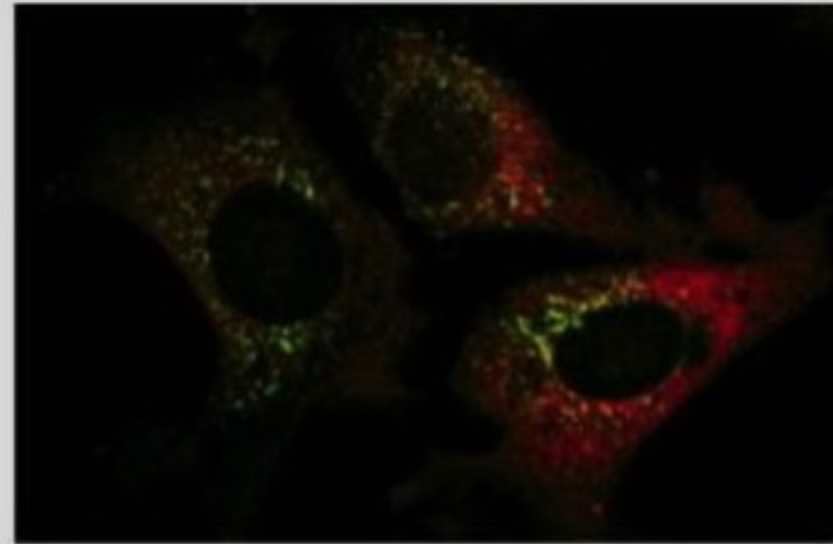
Golgi location

Role of microtubules in maintenance of Golgi structure

Intact microtubules



Disintegration of microtubules



Golgi- green

Microtubules - red

2002 by Bruce Alberts, Alexander Johnson, Julian Lewis, Martin Raff, Keith Roberts, and Peter Walter.

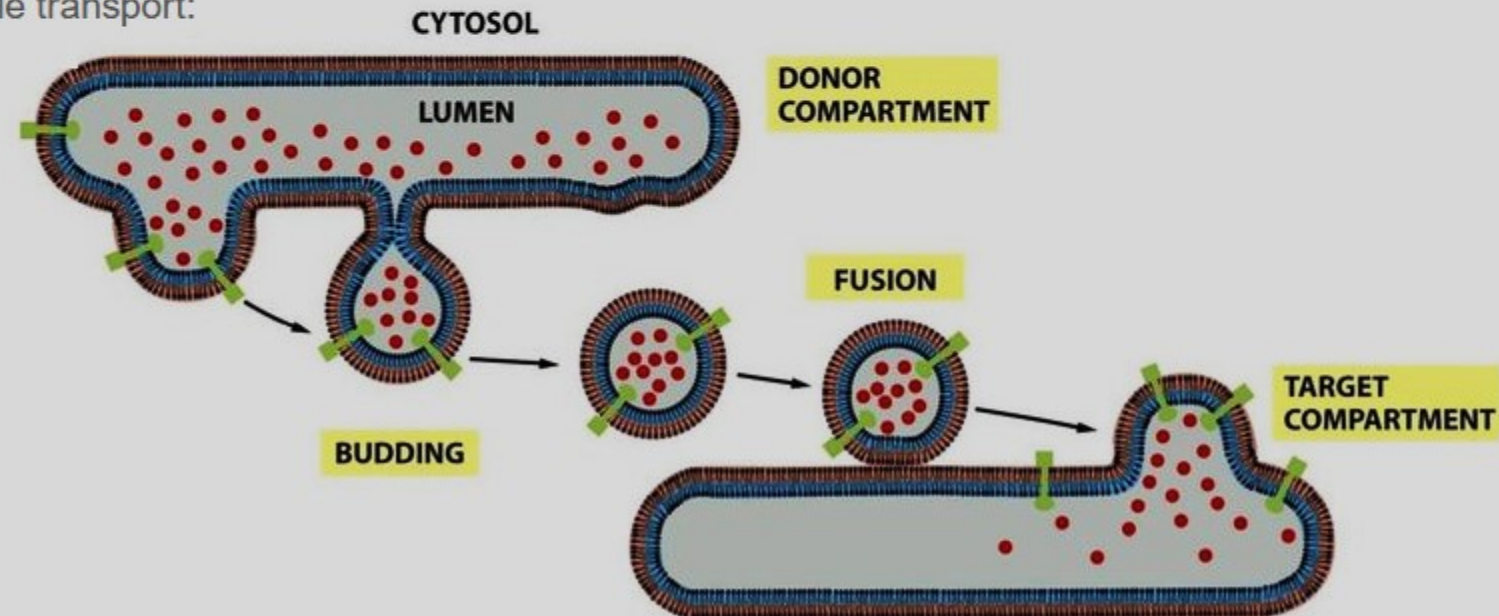
Main Vesicular transport pathways

Vesicular transport

- Process in which membrane-enclosed transport vesicles transport proteins from one membrane-enclosed compartment to another.
- Shape: spherical, larger irregular-shaped vesicles.
- Proteins do not move across the lipid bilayer of any membranes. But only move between topologically equivalent compartments (eg. Lumen of ER to lumen of Golgi to exterior of the cell).

Overview of vesicle transport:

- Budding
- Uncoating
- Transport
- Docking
- Fusion

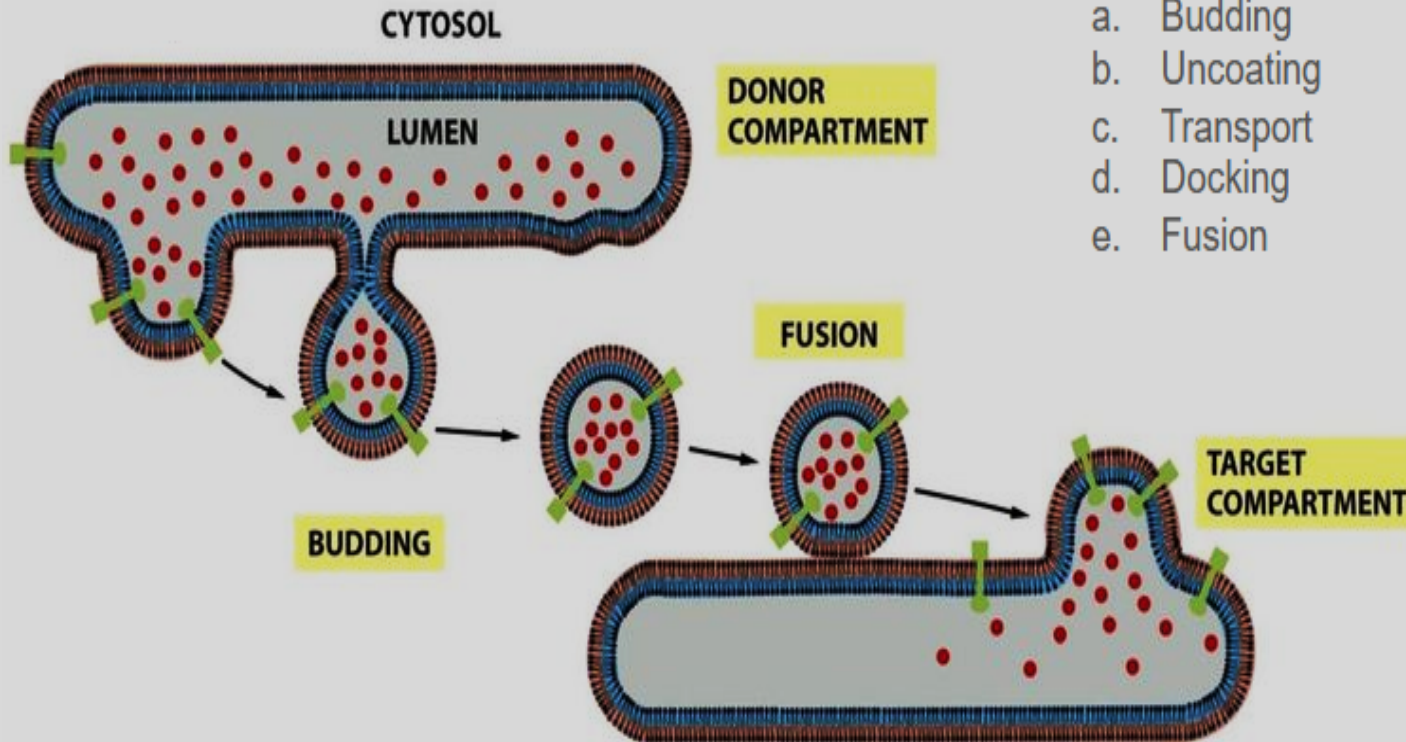


Main Vesicular transport pathways

Principles of vesicular transport

Overview of vesicle transport:

- Budding
- Uncoating
- Transport
- Docking
- Fusion



1. A protein-coated membrane-enclosed transport vesicle **buds** off from the membrane of donor compartment carrying a variety of specifically selected cargo molecules.
2. Transport vesicle binds to the target compartment and **fuse** with the membrane of the target compartment.
3. Cargo molecules transfer into lumen of the target compartment and inserting the vesicular membrane components into the target compartment membrane.

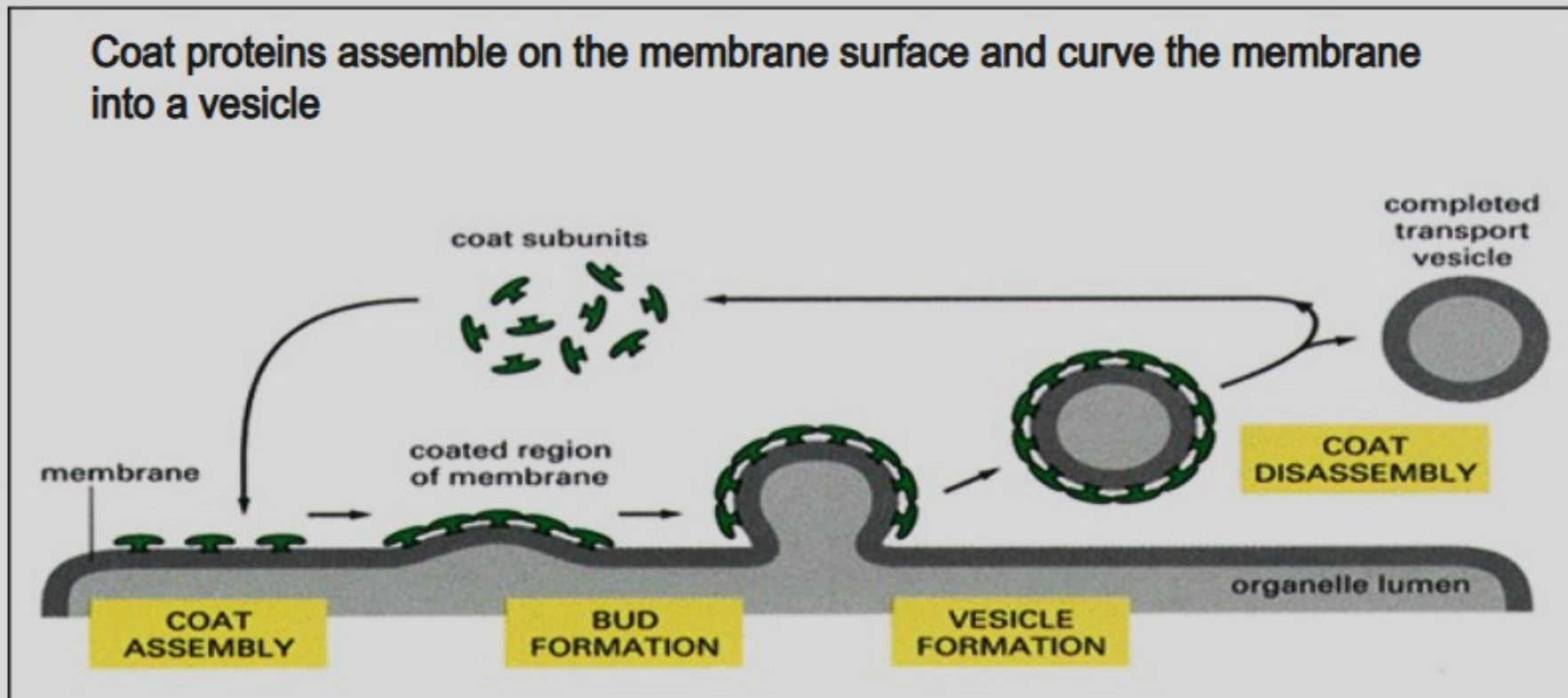
Main Vesicular transport pathways

Question 1: **How do transport vesicles form?**

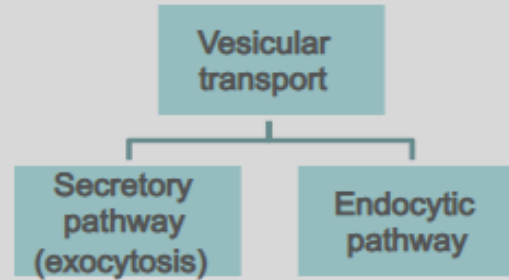
(Budding)

Question 2: **What deforms the membrane to cause a vesicle form?**

(A planar phospholipid lipid bilayer wants to remain flat. But, the small transport vesicles that are seen in cells are small and highly curved. It is this protein coat that causes the membrane to deform and form a transport vesicle.)



Vesicular transport



- new synthesized ER molecules are sorted and delivered to either other organelles or the cell's plasma membrane

- molecules from exterior of the cell are taken up into the cell and trafficked to an appropriate intracellular compartment

- exchange of membrane material and vesicular luminal contents, each organelle maintains its own highly specialized characteristics.

Protein-coated vesicles (bud off donor compartment)
-two functions

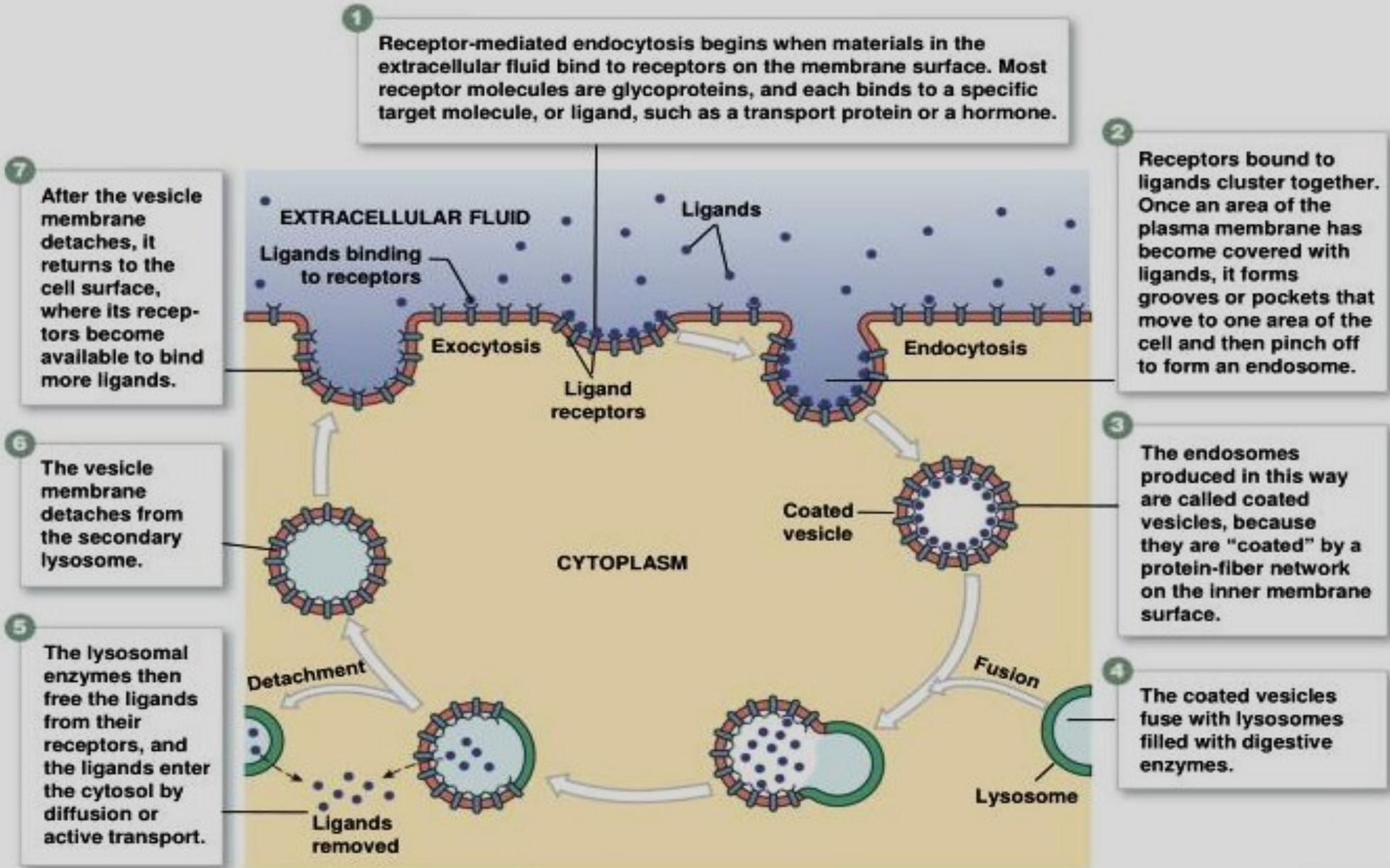
1. Helps select the cargo by concentrating specific membrane proteins into specialized membrane patches that give rise to the vesicle membrane.

2. Assembly of the coat proteins into curved basket-like lattices deforms the membrane in a manner that helps form vesicles of uniform size

Vesicular transport

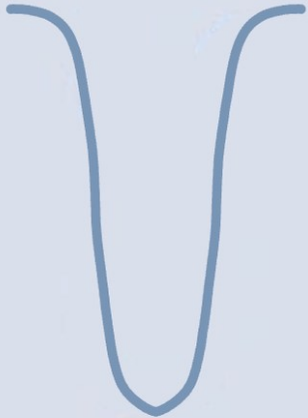
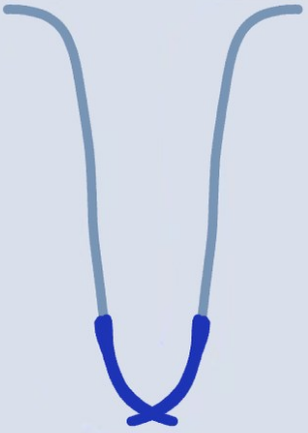
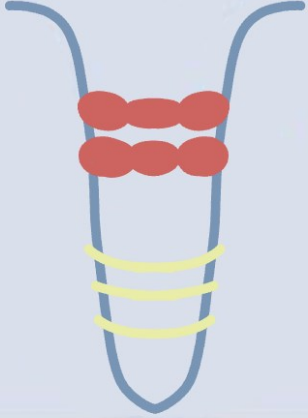
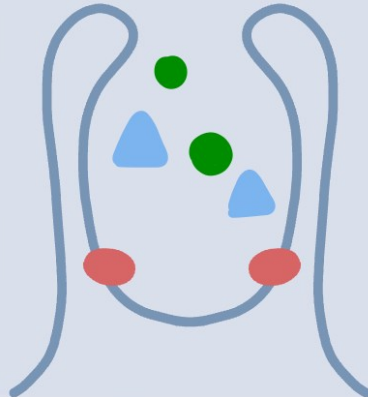
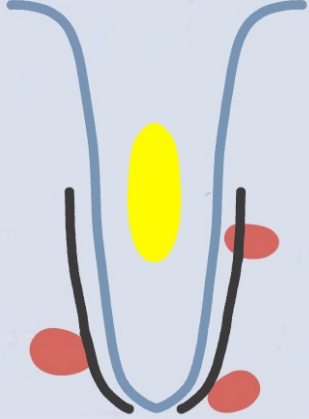
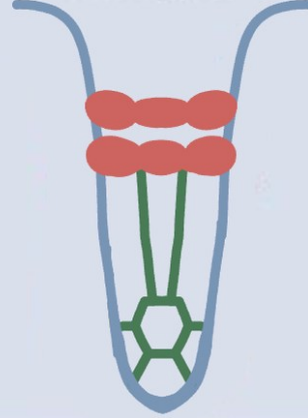
- Two major types (both require ATP)
 1. **Endocytosis** (into cell using **endosomes**)
 - a. **Receptor-mediated endocytosis**
 - 1) **Ligand** binds to receptor
 - 2) Plasma membrane folds around receptors bound to ligands
 - 3) **Coated vesicle** forms
 - 4) Vesicle fuses with lysosomes
 - 5) Ligands freed and enter cytosol
 - 6) Lysosome detaches from vesicle
 - 7) Vesicle fuses with plasma membrane again

Receptor-mediated Endocytosis



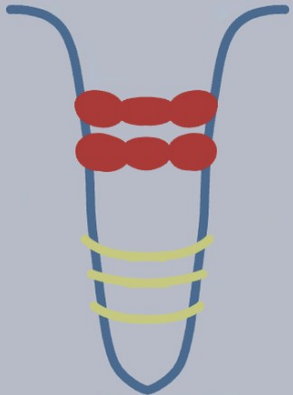
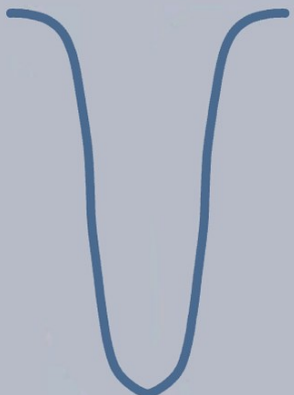
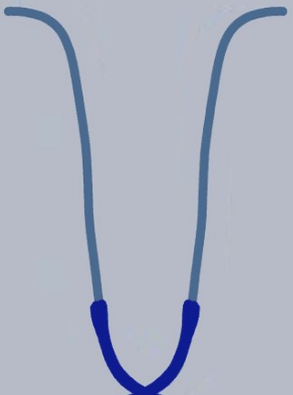
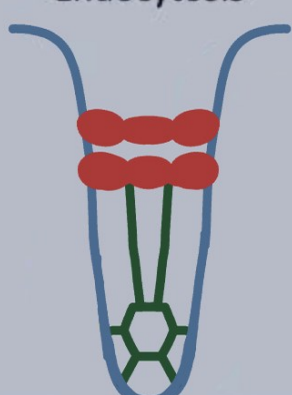
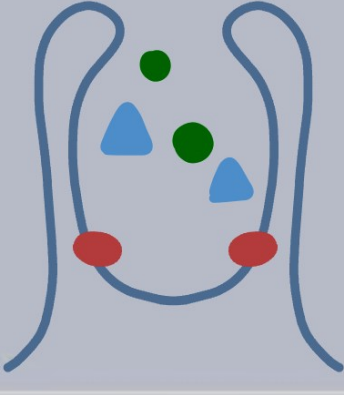
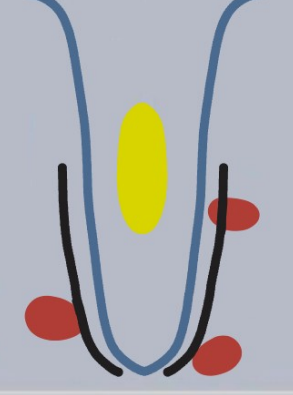
Vesicular transport

Classification of endocytosis pathways based on clathrin and dynamin-involvement

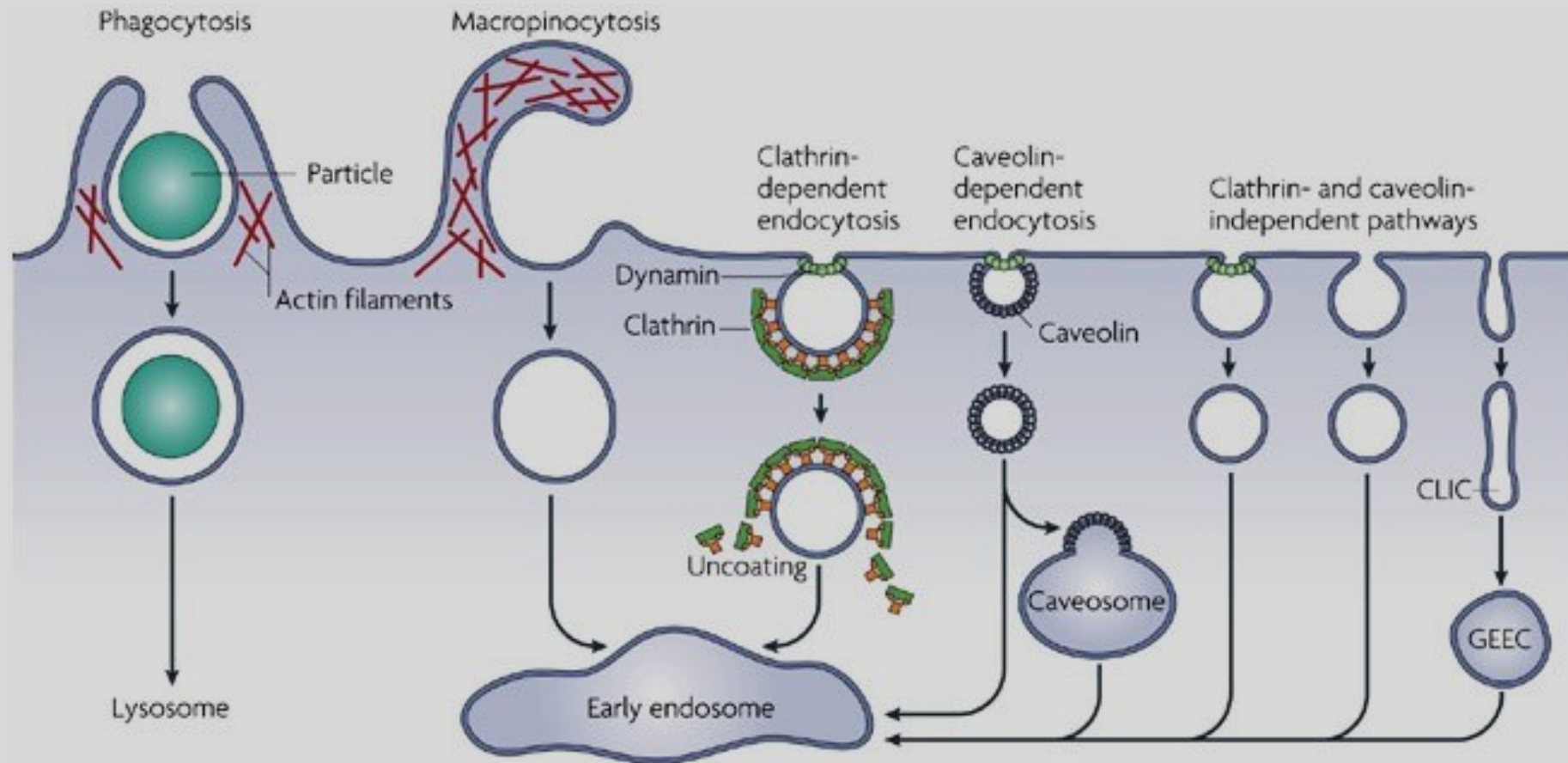
Clathrin and dynamin independent		Clathrin-independent and dynamin -dependent			Clathrin and dynamin dependent
CLIC/GEEC pathway Endocytosis	Flotillin pathway Endocytosis	Caveolae-dependent endocytosis	Micropinocytosis	Phagocytosis	Clathrin-mediated endocytosis
					
Main regulators: Cdc42 Actin	Main regulators: Flotillin Actin	Main regulators: Caveolin Dynamin Cavins Actin	Main regulators: Dyn-2 Actin BARs Kinases	Main regulators: Dyn-2 Actin	Main regulators: Clathrin Dynamin AP2 Eps15 Epsin Myosin Actin

Vesicular transport

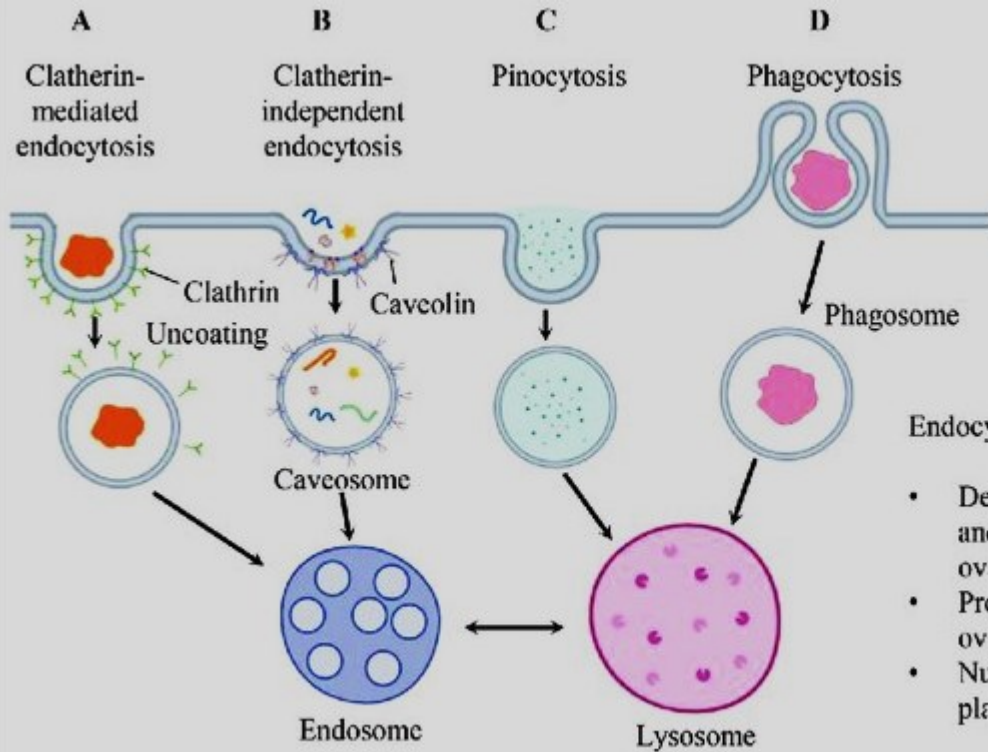
Lipid-rafts based classification of endocytosis pathways

In lipid-rafts membrane domain				Non-lipid rafts membrane domain	Mixed membrane domains	
Caveolae-dependent Endocytosis	CLIC/GEEC pathway Endocytosis	Flotillin pathway Endocytosis	Other Endocytosis pathways Pathways dependent on small GTPases	Clathrin-mediated Endocytosis	Micropinocytosis	Phagocytosis
						
Main regulators: Caveolin Dynamin Cavins Actin	Main regulators: Cdc42 Actin	Main regulators: Flotillin Actin	Main regulators: Cdc42 Arf1 Arf6 RhoA	Main regulators: Clathrin Dynamin AP2 Eps15 Epsin Myosin Actin	Main regulators: Dyn-2 Actin BARs Kinases	Main regulators: Dyn-2 Actin

Vesicular transport

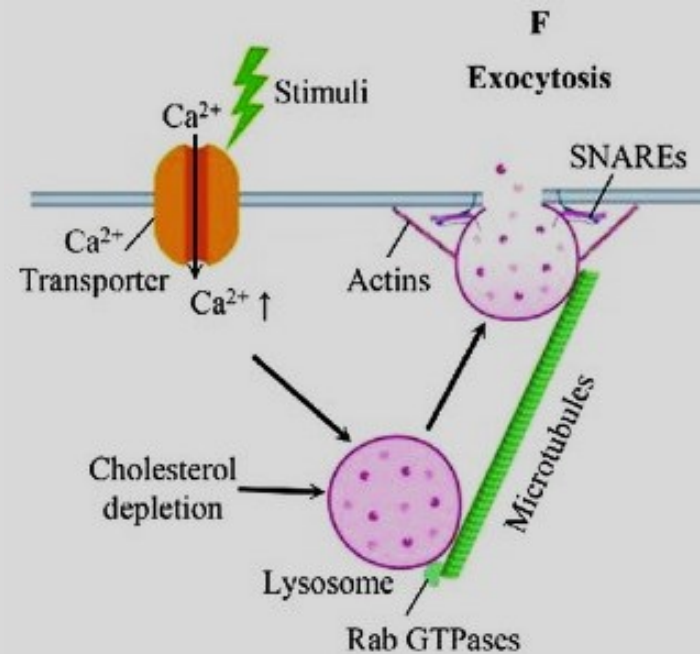


Vesicular transport



E Endocytosis involved in:

- Degradation of hormones and their receptors in the ovary
- Proteins uptake in the oviduct and uterus
- Nutrient transport in the placenta



G Exocytosis involved in:

- Ovulation
- Apical secretion of lysosomal proteins in the oviduct and uterus
- Secretion of extracellular vesicles to uterine lumen

Four main types of endocytosis and a proposed exocytosis involving lysosomes. (A) Clathrin-mediated endocytosis. (B) Clathrin-independent endocytosis. (C) Pinocytosis. (D) Phagocytosis. (E) A few events involving endocytosis in the female reproductive system. (F) A proposed lysosomal exocytosis. (G) A few events potentially involving exocytosis in the oviduct and uterus.

Vesicular transport

The sequence of these steps is similar in yeast and eukaryotic cells, but the requirement for key proteins differs.

In all cells, the process initiates by

- 1. the adaptor and accessory protein-mediated coordination of clathrin at nucleation sites on the plasma membrane to form the clathrin-coated pits (CCPs)**
- 2. polymerization of clathrin and/or actin into curved lattice structures;**
- 3. followed by formation of vesicular necks.**
- 4. Further on, the necks are constricted to bring distant membrane regions nearby.**
- 5. In mammals, the scission protein dynamin, a large GTPase, forms a helical polymer around the neck and mediates the release of the vesicle from the plasma membrane. This process requires GTP hydrolysis, is irreversible and triggers the release of clathrin from the vesicle.**
- 6. In yeast, actin only is essential for scission. While clathrin is required for vesicle formation in mammalian cells, actin and dynamin are not.**
- 7. In contrast, the endocytic process in yeast requires actin for vesicle formation, while clathrin is nonessential**
- 8. Also, dynamin is essential for vesicle release from the plasma membrane in mammalian cells, while it is not essential in yeast.**

Vesicular transport-reference slide

Two major types (both require ATP)

1. Endocytosis (into cell using **endosomes**) (continued)

b. Pinocytosis (“cell drinking”)

- Formation of endosomes with ECF
- No receptor proteins involved

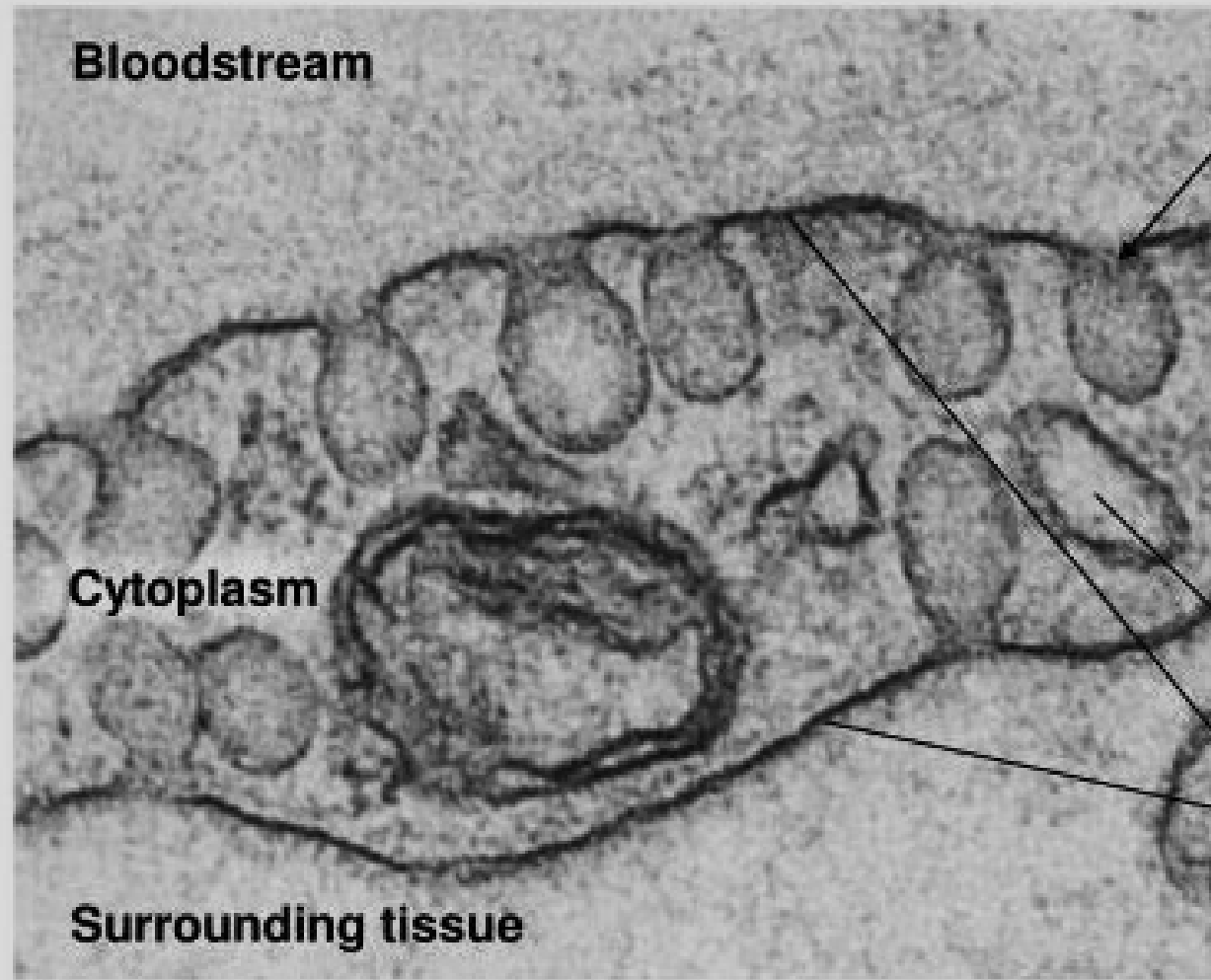
c. Phagocytosis (“cell eating”)

- Produces **phagosomes** containing solids
- **Phagocytes** or **macrophages** perform phagocytosis

2. Exocytosis

- Vesicle discharges materials into ECF

Vesicular transport: Cell Drinking-reference slide



Pinocytosis

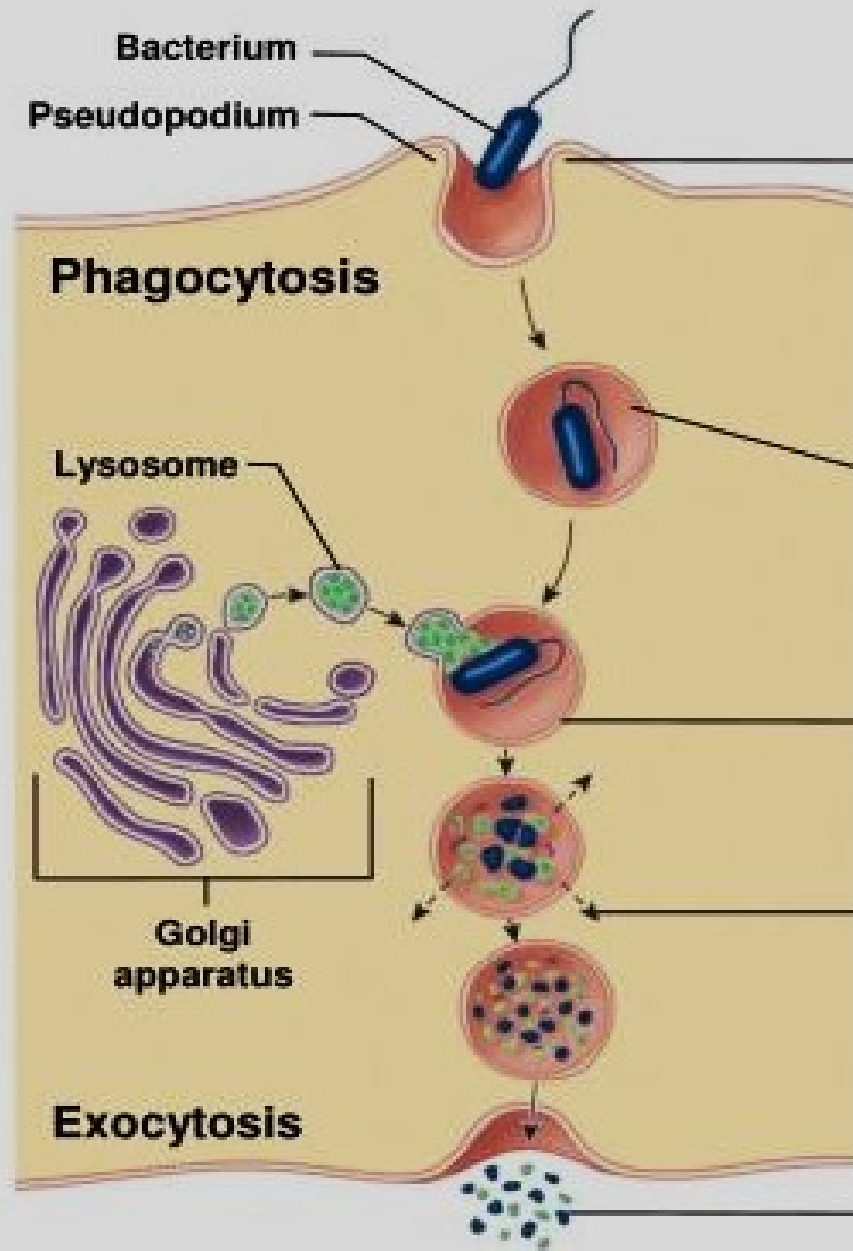
TEM × 20,000

Pinocytosis begins with the formation of deep grooves or pockets that then pinch off and enter the cytoplasm. The steps are similar to those of receptor-mediated endocytosis, but they occur in the absence of ligand binding.

Endosome

Plasma membrane

Vesicular transport: Cell Eating-reference slide



The vesicular events linking phagocytosis and exocytosis

Phagocytosis begins when cytoplasmic extensions called pseudopodia (soo-dō-PŌ-dē-ah; *pseudo*-, false *podon*, foot; singular *pseudopodium*) surround the object.

The pseudopodia then fuse at their tips to form a phagosome containing the targeted material.

This vesicle then fuses with many lysosomes, whereupon its contents are digested by lysosomal enzymes.

Released nutrients diffuse into the surrounding cytoplasm.

The residue is then ejected from the cell through exocytosis.

<https://www.youtube.com/watch?v=owdlv8M5wjA>
<https://www.youtube.com/watch?v=4sWnK7OqK-k>