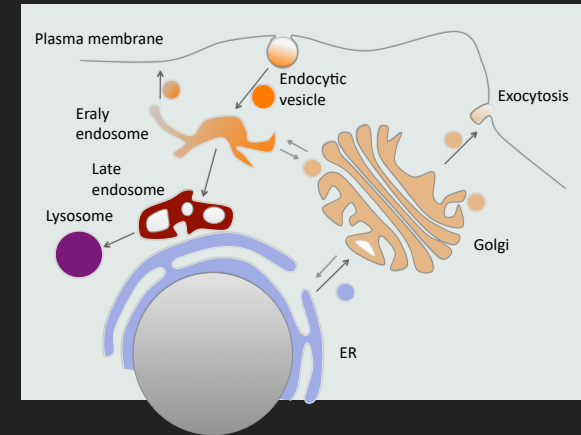


MEMBRANE TRAFFIC

PR CHRISTIAN POÜS 2020-2024

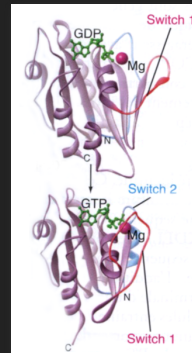
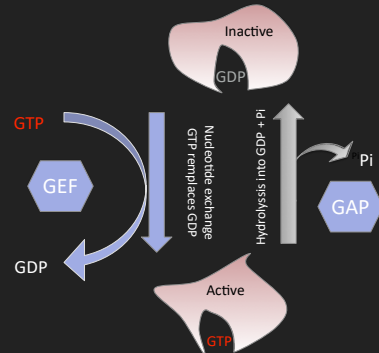
MEMBRANE TRAFFIC

GENERAL ORGANISATION



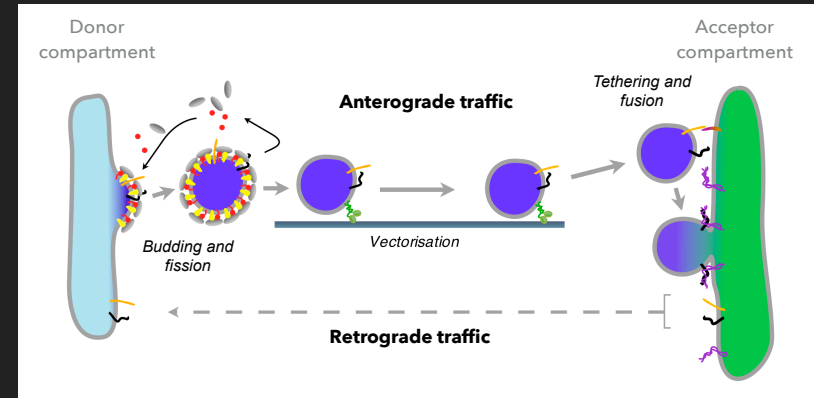
MEMBRANE TRAFFIC

GENERAL ORGANISATION



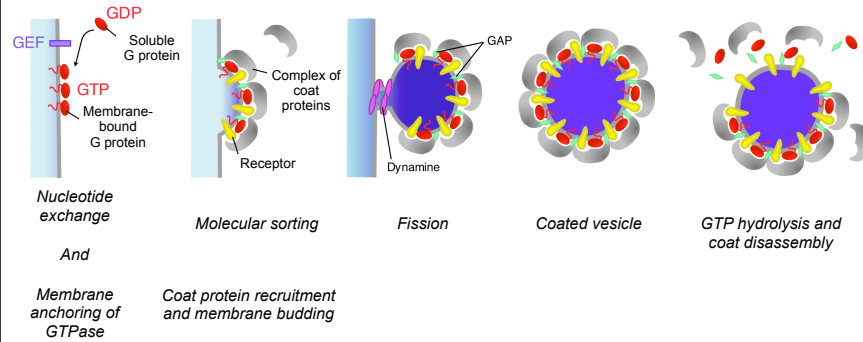
ARF1 = ADP-Ribosylation Factor 1

GEF = Guanine nucleotide Exchange Factor
GAP = GTPase Activating Protein



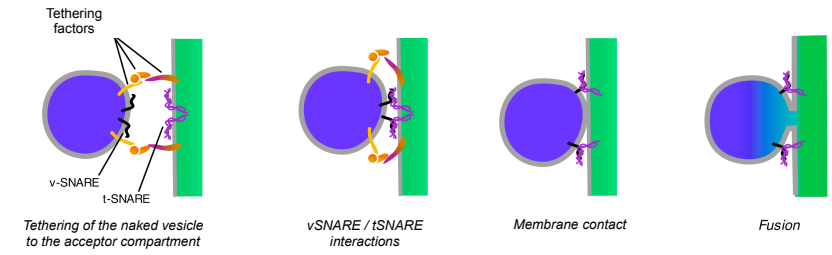
MEMBRANE TRAFFIC

GENERAL ORGANISATION



MEMBRANE TRAFFIC

GENERAL ORGANISATION



MEMBRANE TRAFFIC

MOLECULAR MACHINERY

GTPases OF THE ARF FAMILY

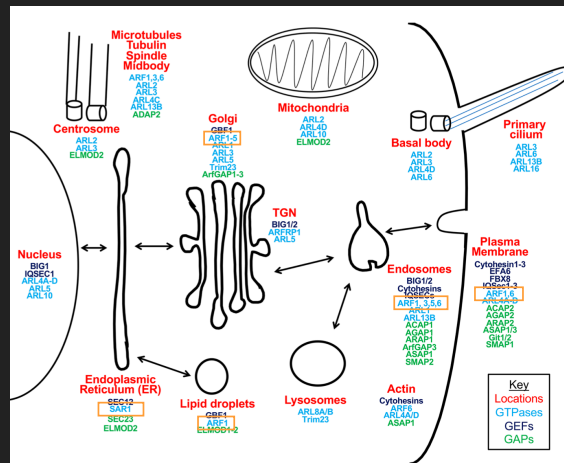
ARF 1 - 6

ARL 1 - 16

Sar1

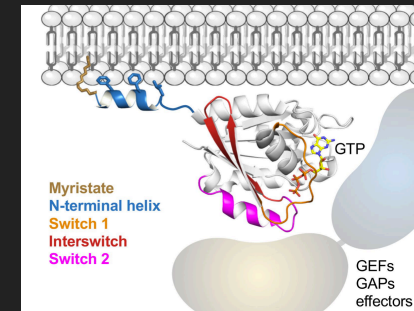
Redundant localizations

A same Arf can distribute at multiple locations



MEMBRANE TRAFFIC

MOLECULAR MACHINERY



GTPase activity :

- ARF-GTP no hydrolysis
- ARF-GTP + ARF-GAP : 1/1000 of Ras + Ras-GAP
- ARF-GTP + ARF-GAP + Coatomer = Ras + Ras-GAP

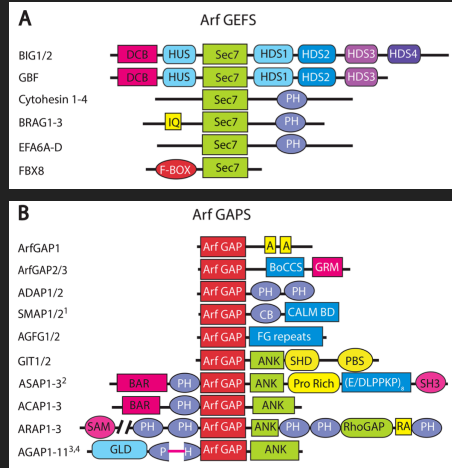
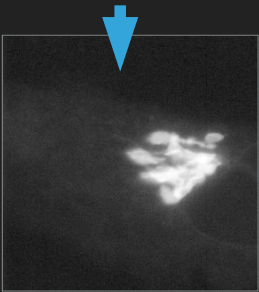
Hydrolysis blocked during coat assembly :

- Budding not perturbed by GTP-γS, but problems of molecular sorting

MEMBRANE TRAFFIC
MOLECULAR MACHINERY

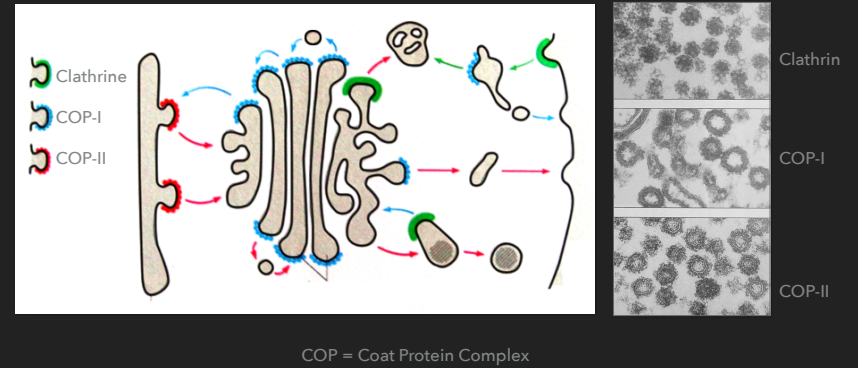
GEF & GAP

- ◆ Effector catalytic domains
- ◆ Sec7 target of Brefeldin A



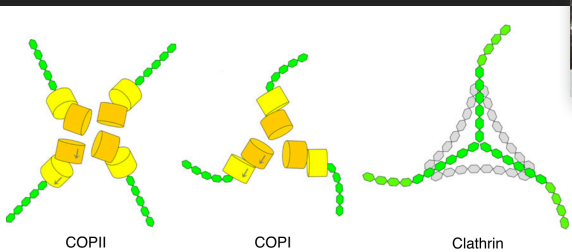
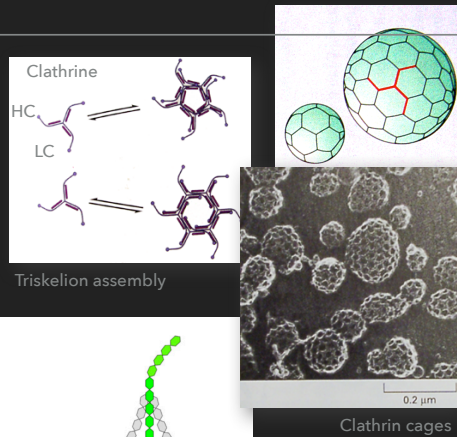
MEMBRANE TRAFFIC
MOLECULAR MACHINERY

COAT PROTEINS



MEMBRANE TRAFFIC
MOLECULAR MACHINERY

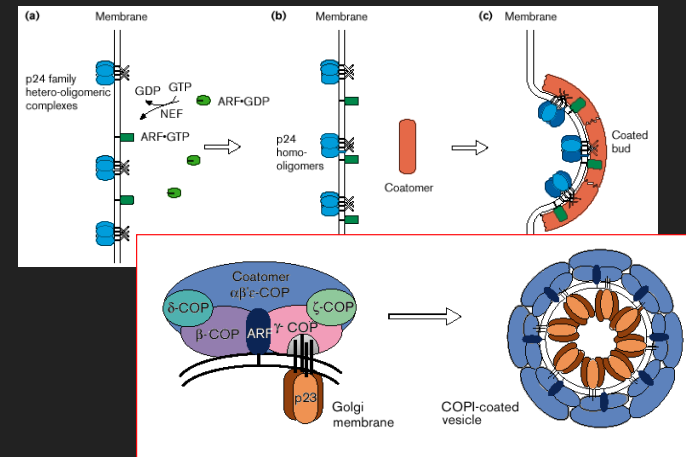
COAT PROTEINS



Lee & Goldberg, Cell, 2010, 142:123.

MEMBRANE TRAFFIC
MOLECULAR MACHINERY

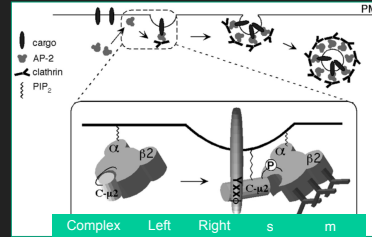
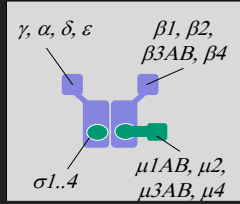
Molecular sorting upon ER exit



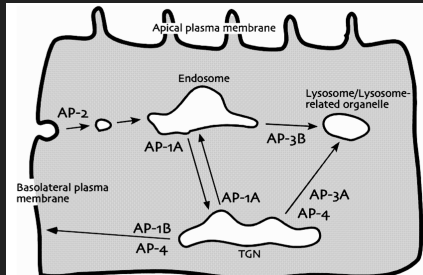
MEMBRANE TRAFFIC

MOLECULAR MACHINERY

Clathrin adapters



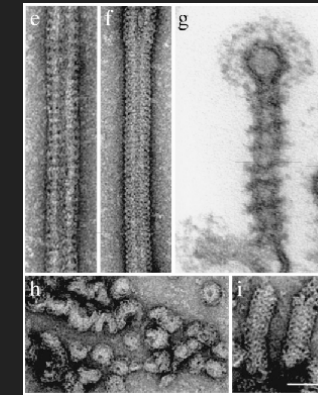
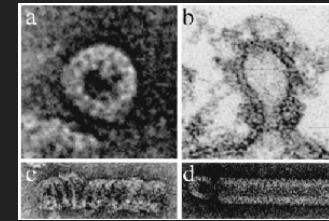
Complex	Left ear	Right ear	s	m
AP-1A	g	b1	s1	m1A
AP-1B	g	b1	s1	m1B
AP-2	a	b2	s2	m2
AP-3A	d	b3A	s3	m3A
AP-3B	d	b3B	s3	m3B
AP-4	e	b4	s4	m4



MEMBRANE TRAFFIC

MOLECULAR MACHINERY

FISSION

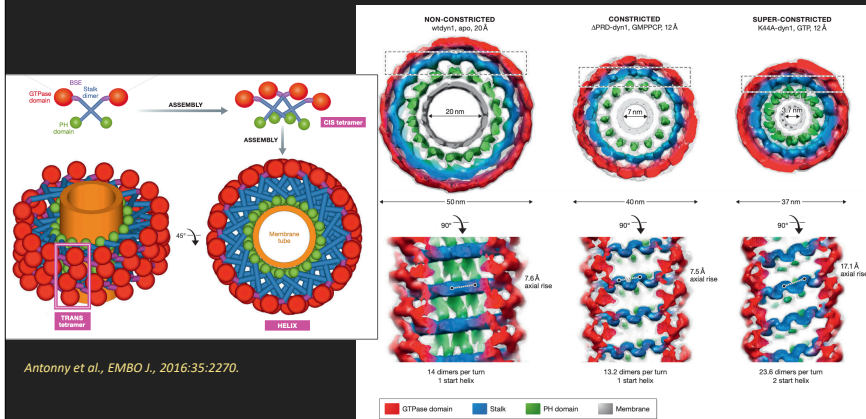


Dynamine
GTPase ~ 100 kDa

MEMBRANE TRAFFIC

MOLECULAR MACHINERY

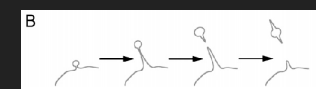
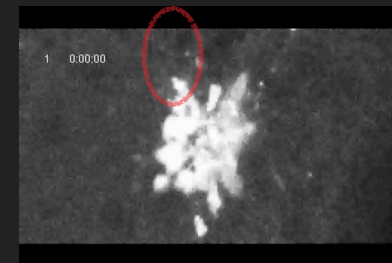
FISSION



MEMBRANE TRAFFIC

MOLECULAR MACHINERY

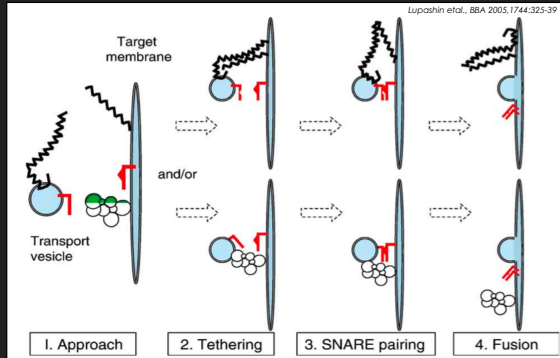
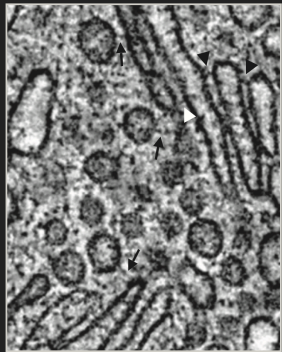
FISSION



Cooperation dynamine + molecular motor

MEMBRANE TRAFFIC
MOLECULAR MACHINERY

TETHERING FACTORS

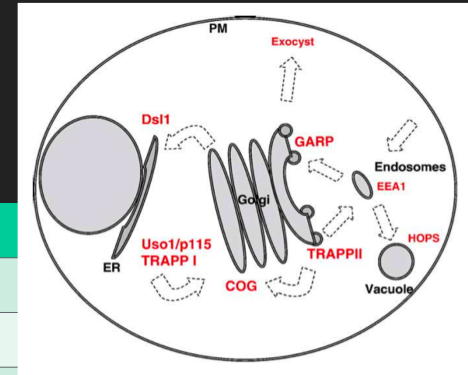


Sztul & Lupashin, Am. J. Physiol. Cell Physiol., 2006, 290: C11

MEMBRANE TRAFFIC
MOLECULAR MACHINERY

TETHERING FACTORS

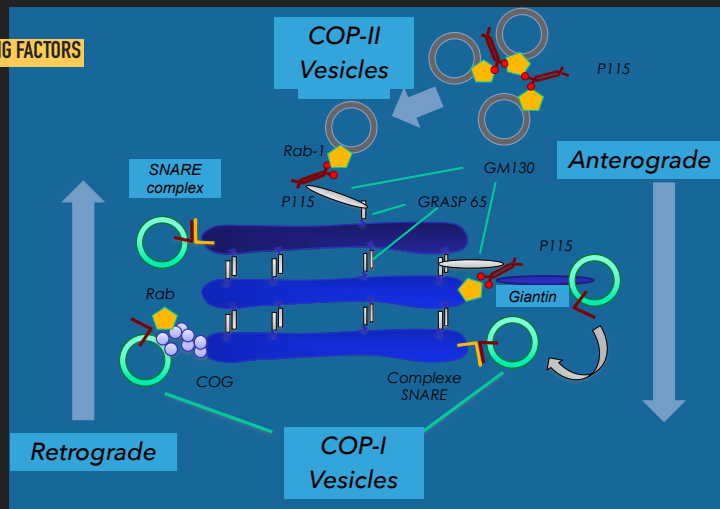
Traffic step	Tethering factor
ER-Golgi	Uso1, TRAPP I
Intra-Golgi	Golgins, COG
Golgi-Plasma membrane	Exocyst
Plasma membrane - Endosome	EEA1, Vac1p



Sztul & Lupashin, Am. J. Physiol. Cell Physiol., 2006, 290: C11

MEMBRANE TRAFFIC
MOLECULAR MACHINERY

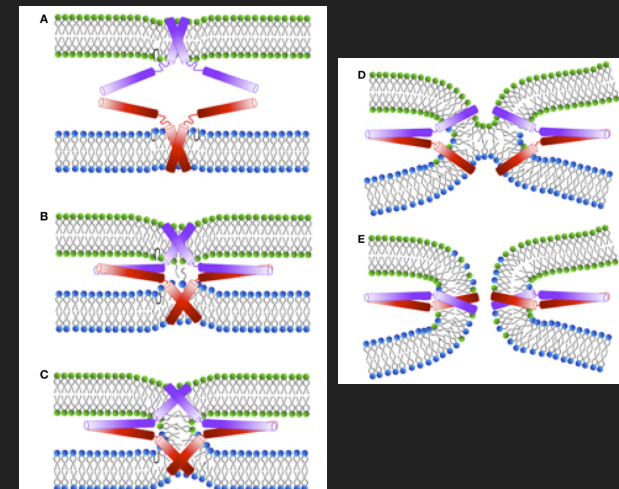
TETHERING FACTORS



MEMBRANE TRAFFIC
MOLECULAR MACHINERY

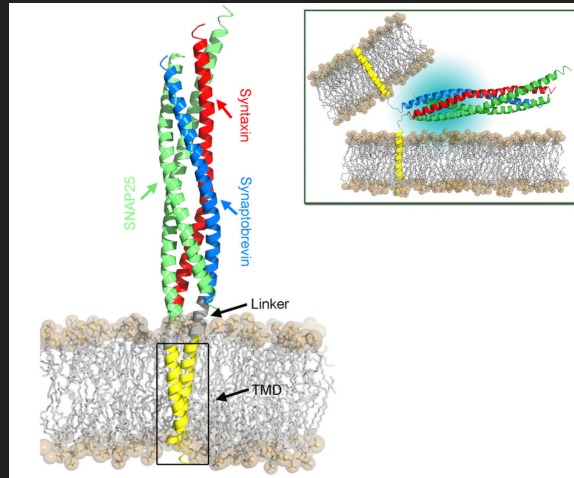
FUSION

Reorganization of SNAREs and membrane lipids



MEMBRANE TRAFFIC
MOLECULAR MACHINERY

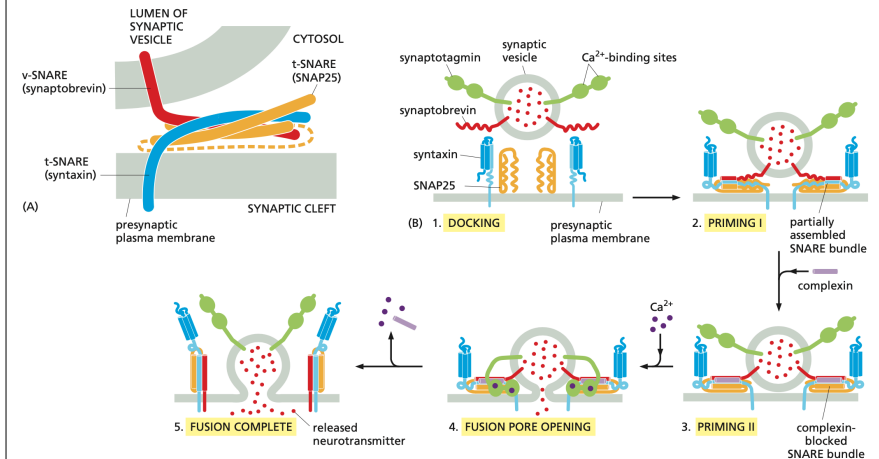
FUSION



MEMBRANE TRAFFIC
MOLECULAR MACHINERY

FUSION

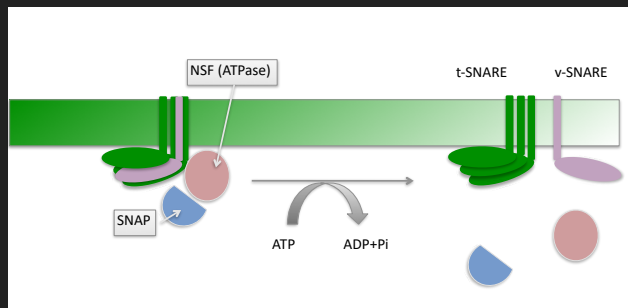
Example of synaptic vesicle exocytosis



Alberts et al., in Molecular Biology of the Cell, Garland Science.

MEMBRANE TRAFFIC
MOLECULAR MACHINERY

SNARE recycling



NSF = N-ethylmaleimide-Sensitive Fusion protein

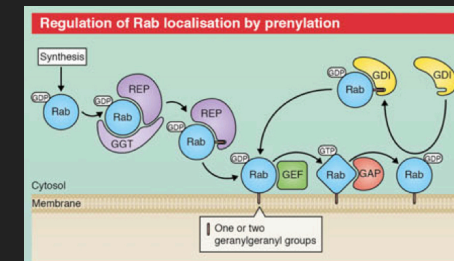
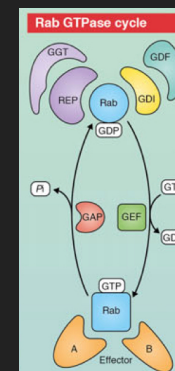
SNAP = Soluble NFS Attachment Protein

SNARE = SNAP Receptor

MEMBRANE TRAFFIC
MOLECULAR MACHINERY

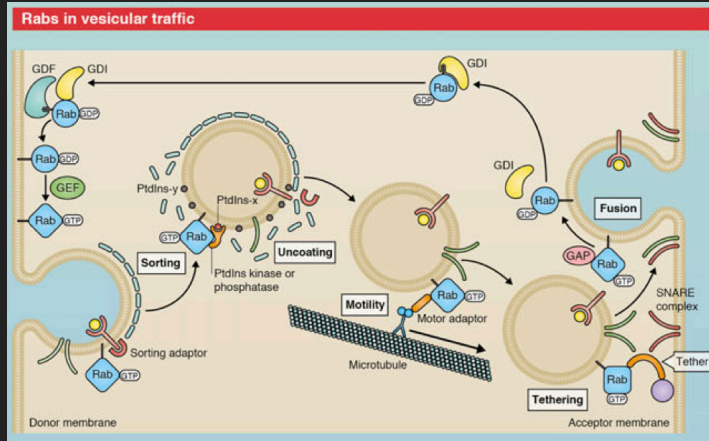
REGULATION BY Rab GTPases

REP = Rab Escort Protein
GGT = Geranyl-Geranyl Transferase
GDI = GDP Dissociation Inhibitor
GDF = GDI Dissociation Factor



Zhen & Stenmark, J Cell Sci, 2015, 128:3171.

MEMBRANE TRAFFIC
MOLECULAR MACHINERY



Zhen & Stenmark, *J Cell Sci*, 2015, 128:3171.

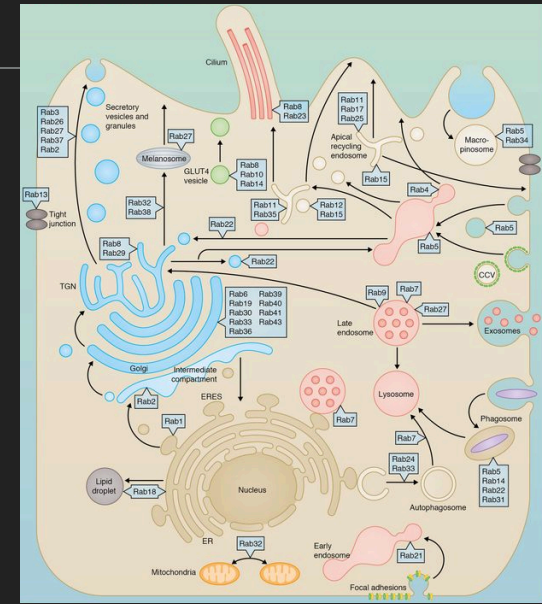
MEMBRANE TRAFFIC
MOLECULAR MACHINERY

~70 Rabs in humans

Control of specificity and directionality of the traffic

With Rab GEFs and Rab GAPs

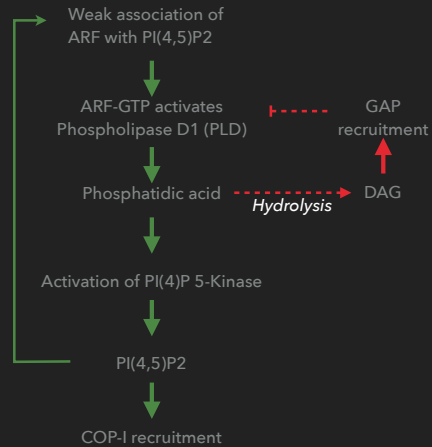
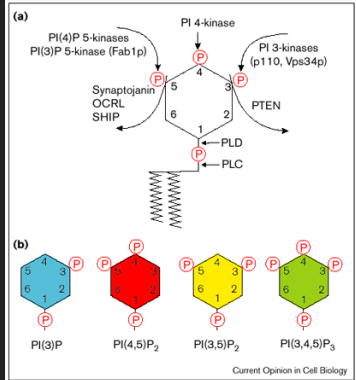
Variety of effectors



Zhen & Stenmark, *J Cell Sci*, 2015, 128:3171.

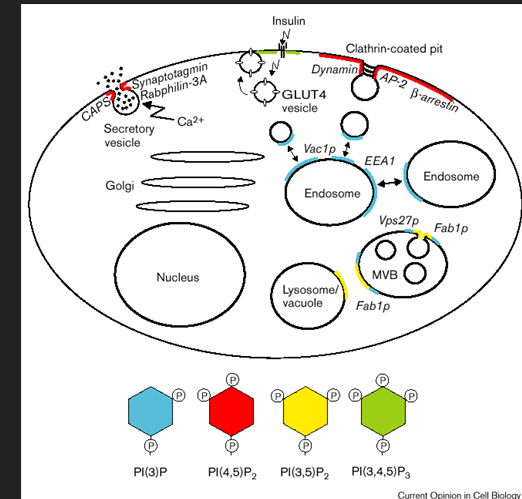
MEMBRANE TRAFFIC
MOLECULAR MACHINERY

REGULATION BY LIPIDS



Current Opinion in Cell Biology

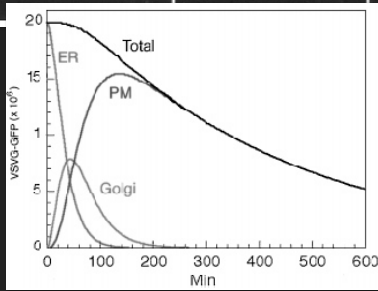
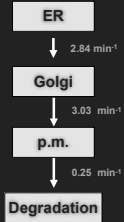
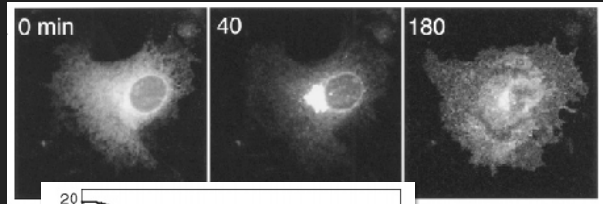
MEMBRANE TRAFFIC
MOLECULAR MACHINERY



Current Opinion in Cell Biology

MEMBRANE TRAFFIC

ER-TO-GOLGI TRAFFIC



Hirschberg et al., 1998, *J. Cell Biol.*, 143:1485-98

MEMBRANE TRAFFIC

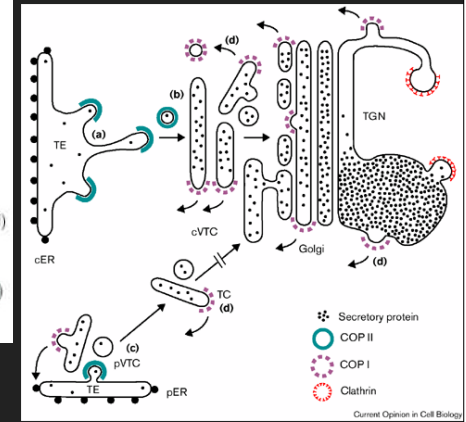
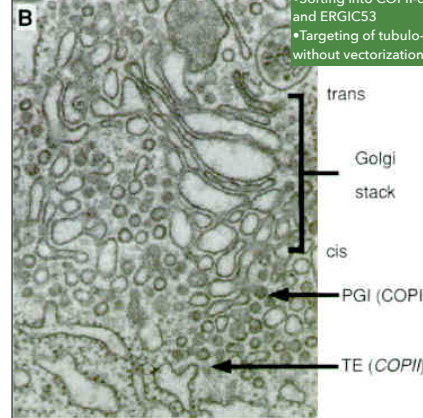
ER-TO-GOLGI TRAFFIC

Cell center:

- Transition elements (TE) = ER exit sites = Smooth regions of the rough ER
- Sorting into COPII-coated vesicles via p24 and ERGIC53
- Targeting of tubulo-vesicular clusters without vectorization

Cell periphery:

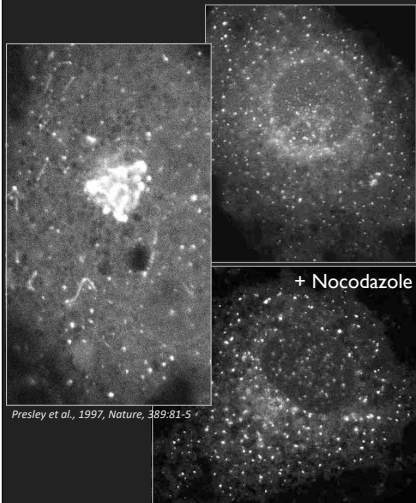
- Vesiculo-tubular clusters (VTC) close to ER exit sites
- VTC transport along microtubules towards the cis-Golgi



Current Opinion in Cell Biology

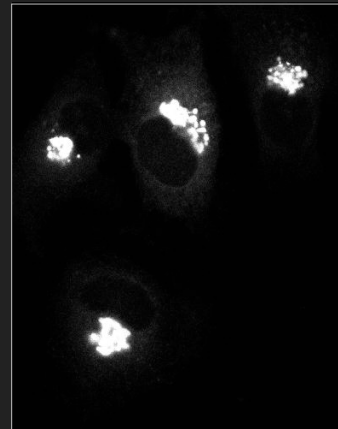
MEMBRANE TRAFFIC

ER-TO-GOLGI TRAFFIC



Presley et al., 1997, *Nature*, 389:81-5

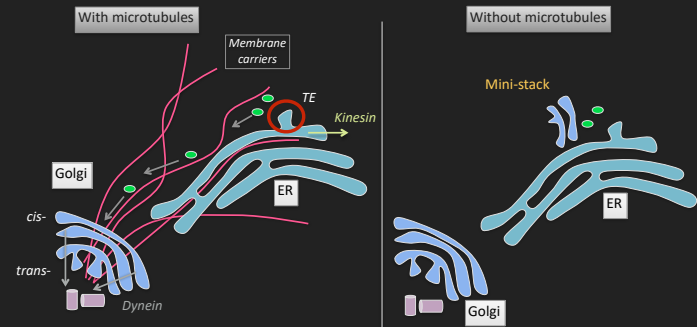
GT + Nocodazole



Sciaky et al., 1997, *J. Cell Biol.*, 139:1137-55

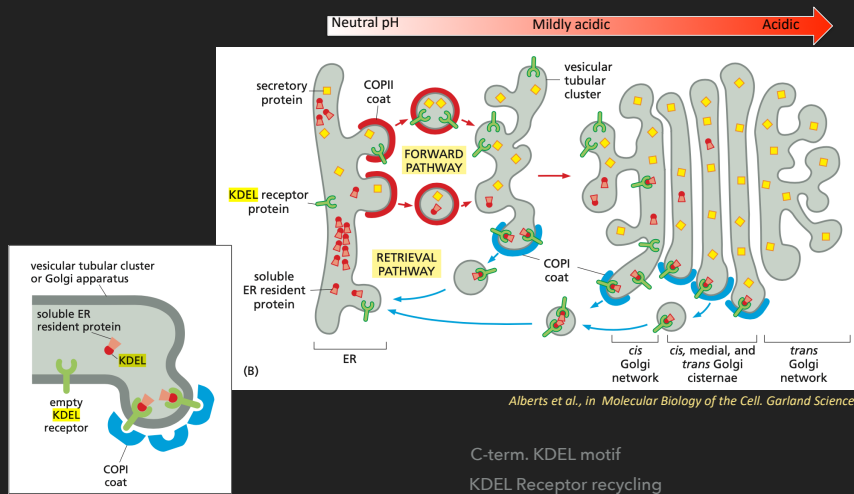
MEMBRANE TRAFFIC

ER-TO-GOLGI TRAFFIC & GOLGI MAINTENANCE



MEMBRANE TRAFFIC

BACK TO THE ER



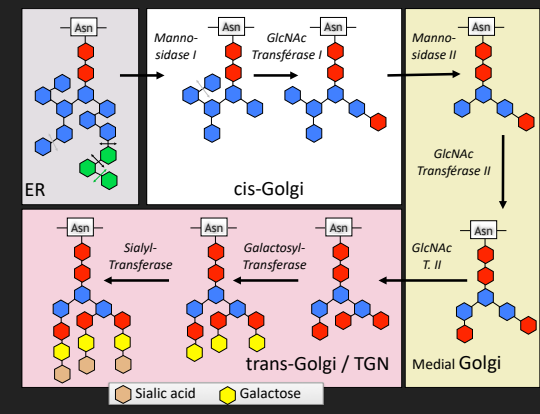
MEMBRANE TRAFFIC

GOLGI FUNCTIONS

N-glycan maturation + many others:

Protein O-glycosylation, lipid glycosylation, phosphate, sulfate additions etc...

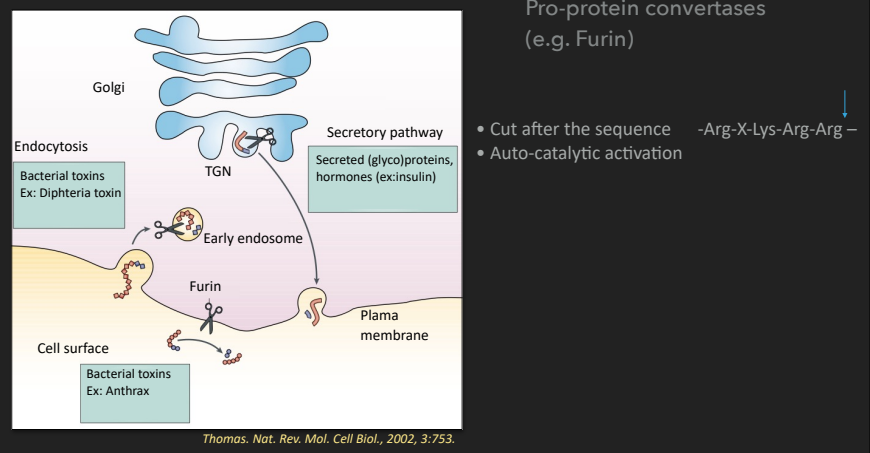
GLYCOSYLATION



MEMBRANE TRAFFIC

GOLGI FUNCTIONS

PROTEOLYTIC MATURATION

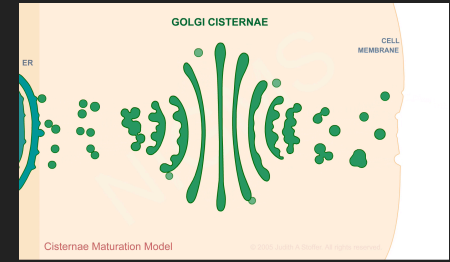
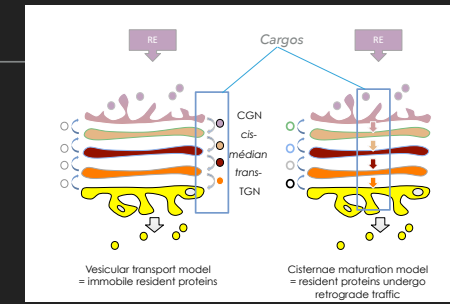
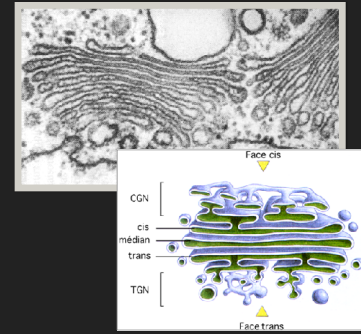


MEMBRANE TRAFFIC

GOLGI MAINTENANCE

ER ~50 to 70% of a cell's membranes
Golgi apparatus < 10% of a cell's membranes

Up to ~10-times higher protein concentration in the Golgi compared to the ER



Alberts et al., in *Molecular Biology of the Cell*. Garland Science.

MEMBRANE TRAFFIC

POST-GOLGI TRAFFIC

SORTING BETWEEN RESIDENT AND EXPORTED PROTEINS IN THE TGN

Glyco-Sphingolipids and Sphingomyelin synthesis in the Golgi lumen

↓

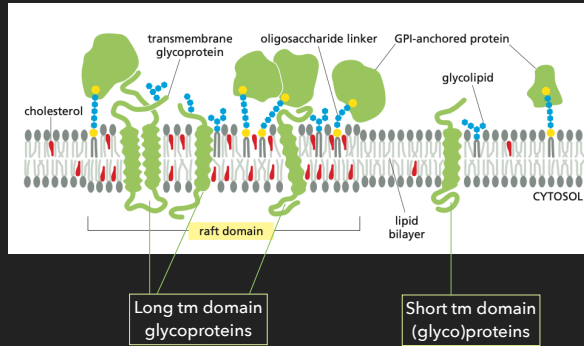
Aggregation with Cholesterol

↓

Phase separation of membrane lipids

↓

Protein segregation according to the length of their transmembrane domains

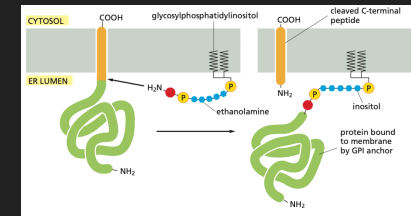


Alberts et al., in *Molecular Biology of the Cell*. Garland Science.

MEMBRANE TRAFFIC

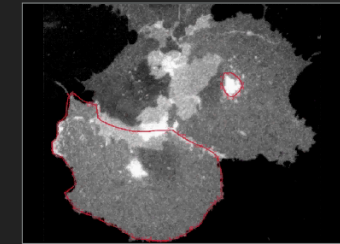
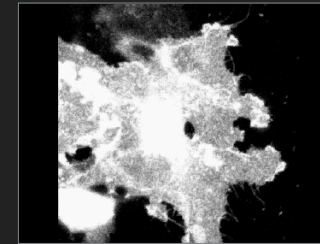
POST-GOLGI TRAFFIC

GPI-anchored glycoproteins are built in the ER



Alberts et al., in *Molecular Biology of the Cell*. Garland Science.

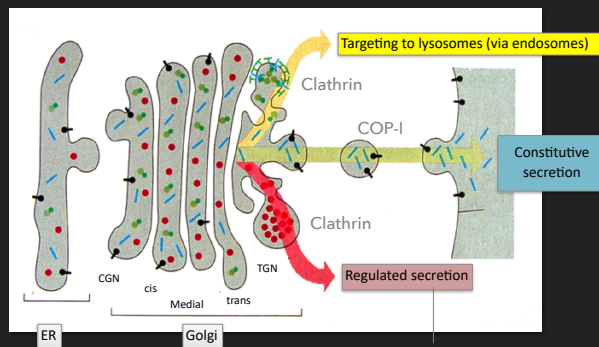
GPI-GFP traffic = cycling between the Golgi and the plasma membrane



Nichols et al., *J. Cell Biol*, 2001, 153:529.

MEMBRANE TRAFFIC

POST-GOLGI TRAFFIC



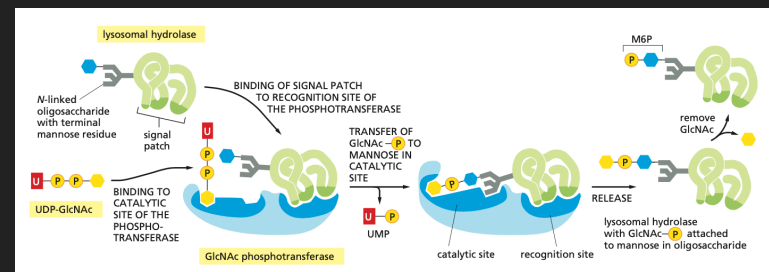
Alberts et al., in *Molecular Biology of the Cell*. Garland Science.

Storage of fusion-ready vesicles
Exocytosis upon signalling.
ex: synaptic vesicle exocytosis

MEMBRANE TRAFFIC

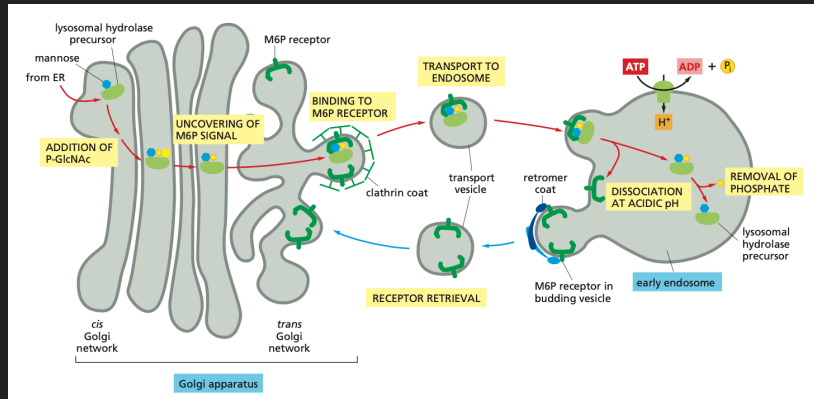
POST-GOLGI TRAFFIC

MAN-6P SYNTHESIS IN THE CGN



Alberts et al., in *Molecular Biology of the Cell*. Garland Science.

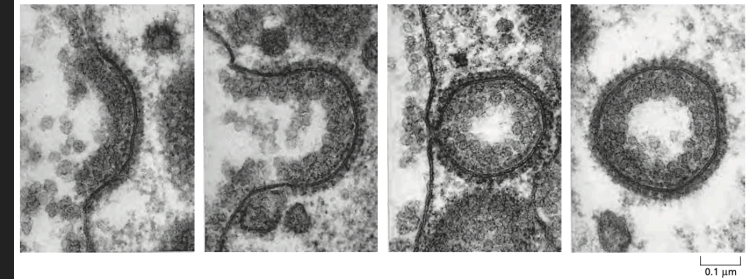
MEMBRANE TRAFFIC
POST-GOLGI TRAFFIC



Alberts et al., in *Molecular Biology of the Cell*, Garland Science.

MEMBRANE TRAFFIC
ENDOCYTOSIS

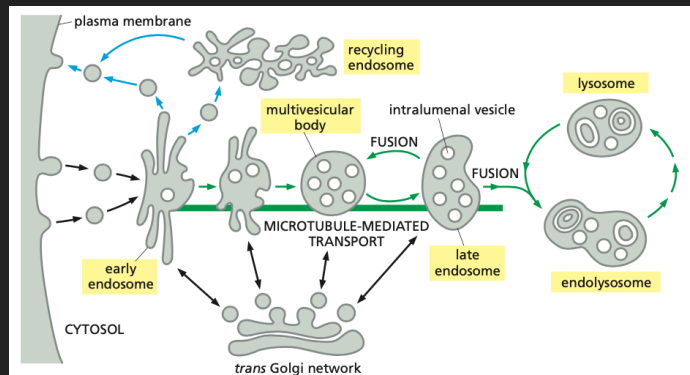
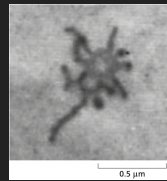
Clathrin-dependent endocytosis



Alberts et al., in *Molecular Biology of the Cell*, Garland Science.

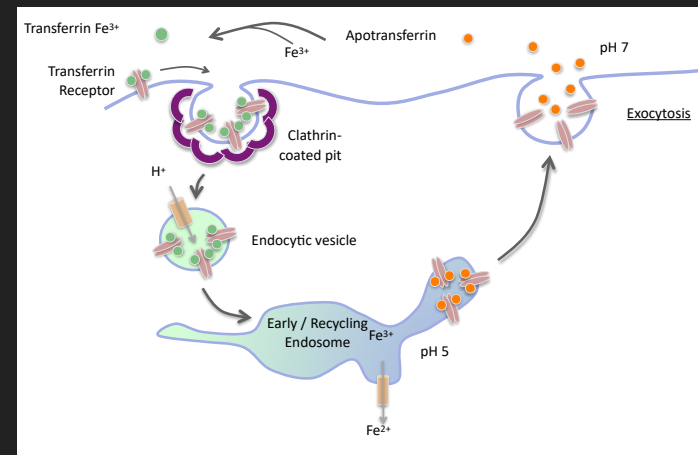
Internalization of selected extracellular ligands

MEMBRANE TRAFFIC
ENDOCYTOSIS



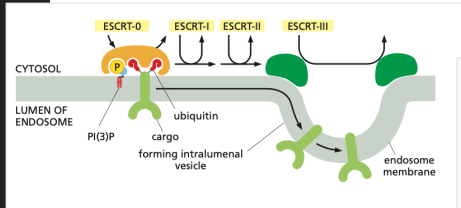
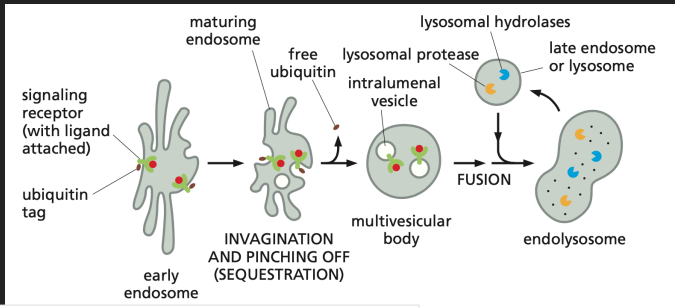
Alberts et al., in *Molecular Biology of the Cell*, Garland Science.

MEMBRANE TRAFFIC
ENDOCYTOSIS: RECYCLING



MEMBRANE TRAFFIC

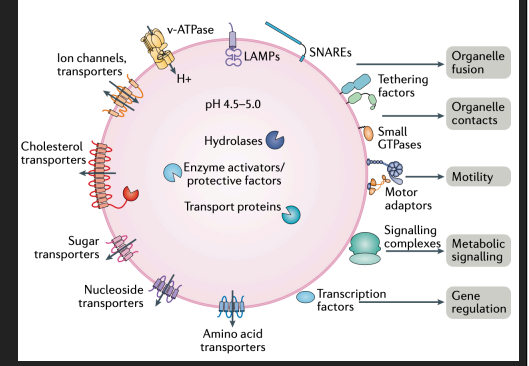
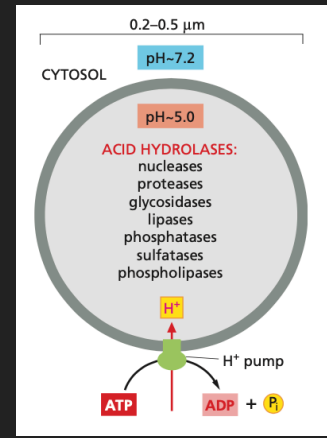
ENDOCYTOSIS: DEGRADATION



Alberts et al., in *Molecular Biology of the Cell*, Garland Science.

MEMBRANE TRAFFIC

LYSOSOMES: DEGRADATION



Ballabio & Bonifacio, *Nat. Rev. Mol. Cell Biol.*, 2020, 21:101.

Alberts et al., in *Molecular Biology of the Cell*, Garland Science.