Lipidne mikrodomene

funkcija

Cellular processes involving lipid rafts

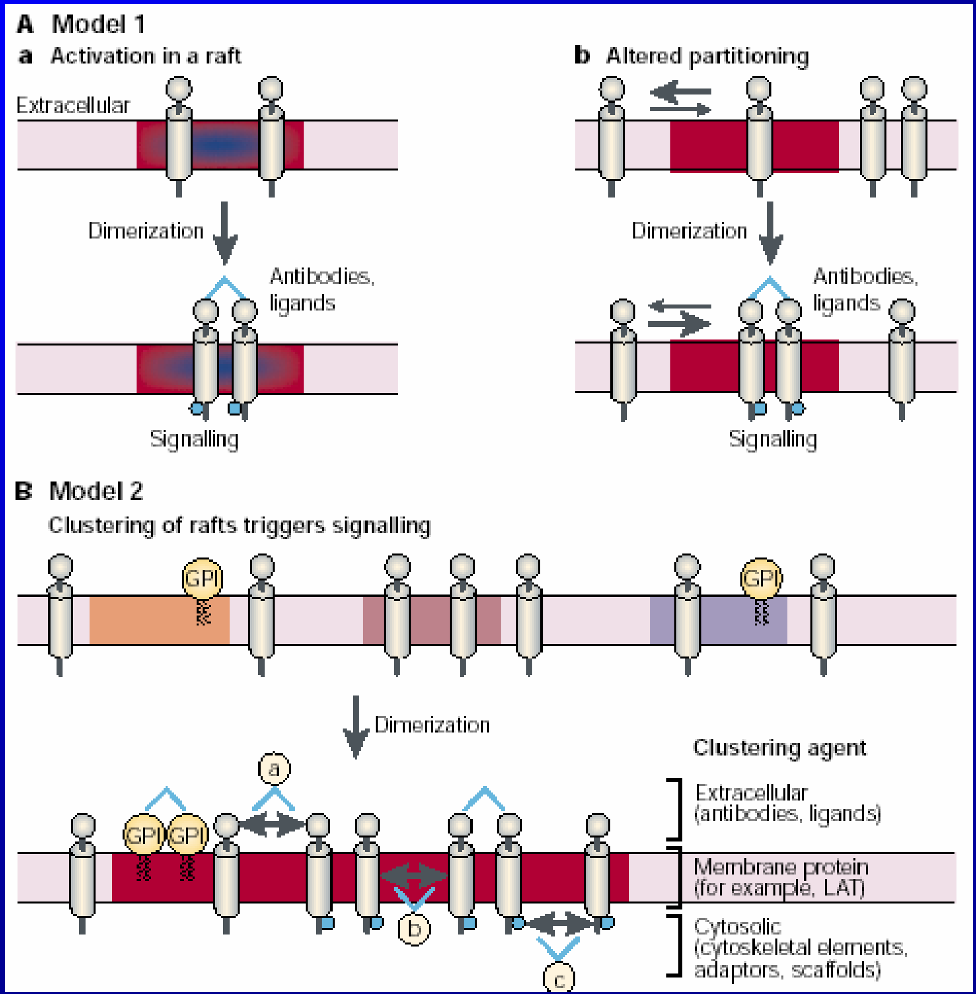
- Signal transduction
- Protein and lipid trafficking and sorting
- Endosome(clathrin)-independent endocytosis:
    - potocytosis and
    - ceveolin-independent endocytosis
- Ca^{2+} homeostasis

Protein and lipid signalling molecules identified in lipid rafts

Protein/lipid
Transmembrane receptors
EGF receptor
Bradykinin B2 receptor
Eph family receptors
TCR
BCR
FcεRI
β1 integrins
Lipid signalling molecules
Sphingomyelin
Ceramide
Phosphoinositides
Diacylglycerol
GPI-linked proteins
CD59
uPAR
EphrinA5
Signalling effectors
$G_{\alpha i1}$, $G_{\alpha i2}$, $G_{\alpha i3}$
Src-family kinases
Ras
PKC α
Shc
Adenylate cyclase
eNOS
PLCγ
PI3K
SHIP
Cbp/PAG

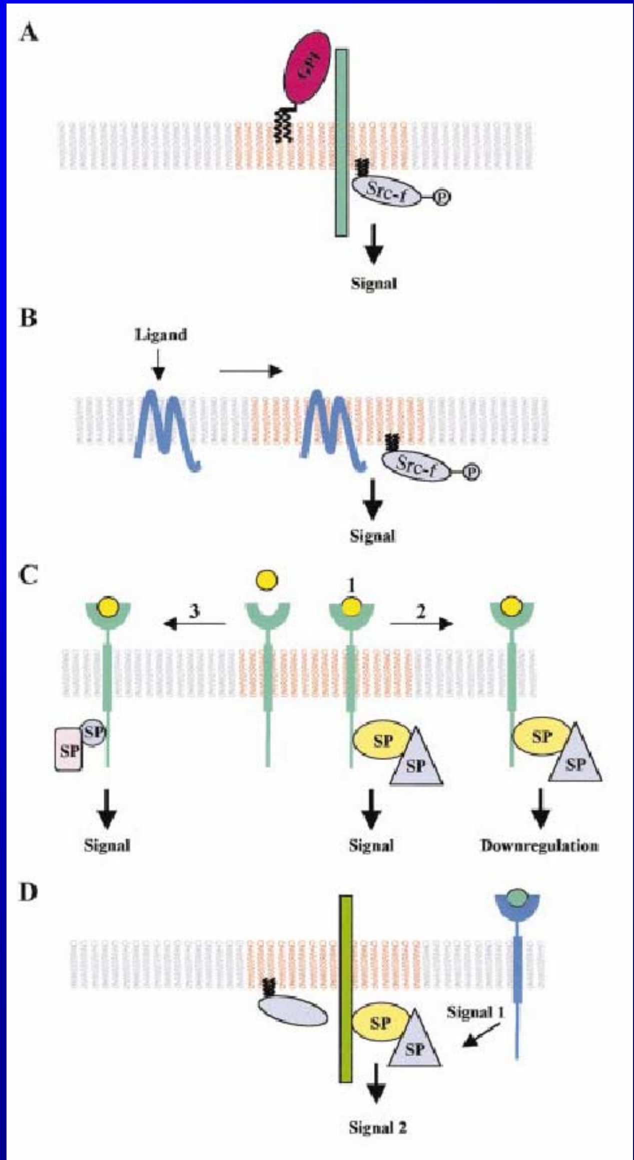
Zajchowski & Robbins (2002) Eur. J. Biochem. 269, 737-752.

Models for signal initiation through rafts



Simons & Toomre (2000) Nat. Rev. Molec. Cell Biol. 1, 31-40.

Proposed roles of lipid rafts in signal transduction



CD59, ephrin

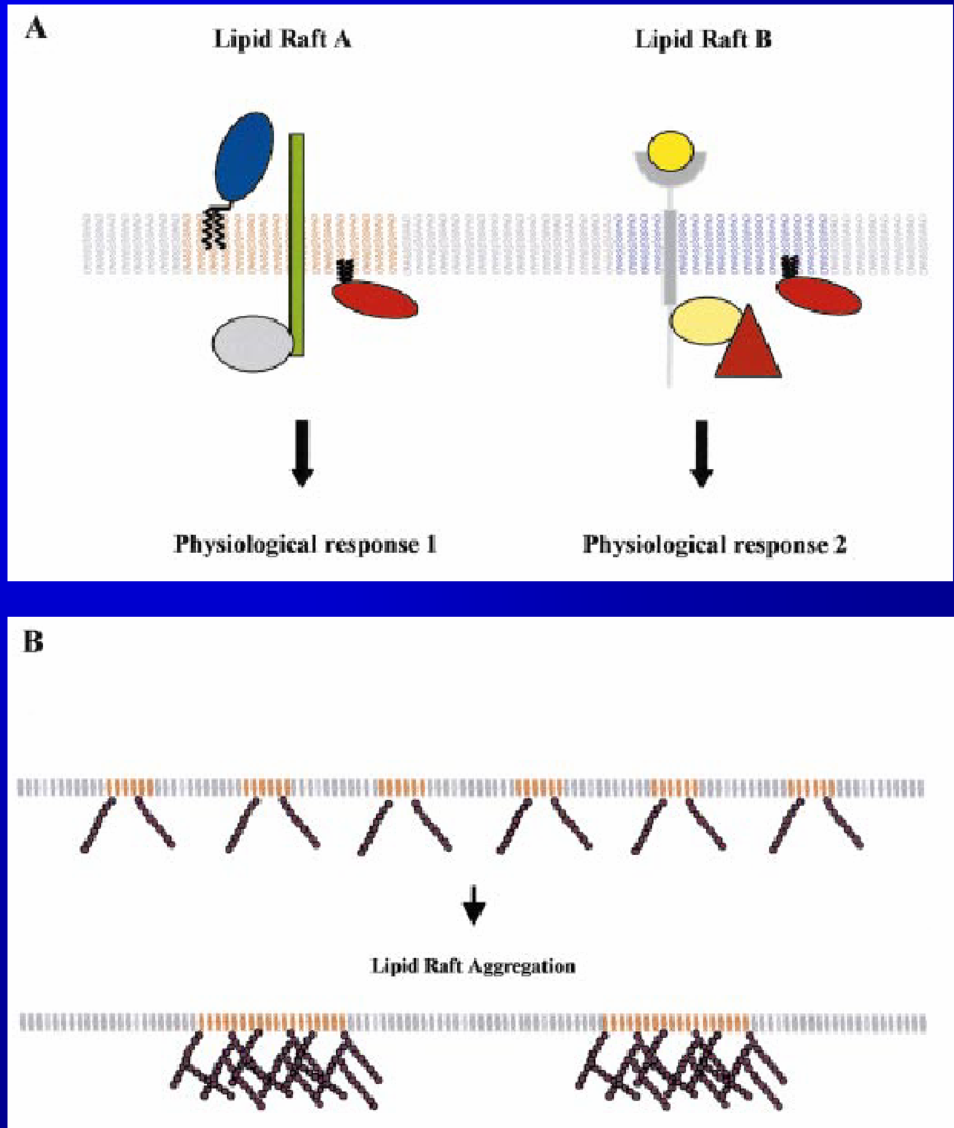
CD20

(1,2) EGFR, PDGFR

(3) IL-2R

Zajchowski & Robbins (2002) Eur. J. Biochem. 269, 737-752.

Signalling specificity by distinct subpopulations of lipid rafts.



Formation of higher-order signalling complexes by clustering of lipid rafts:

- signal amplification
- cross-talk
- spatial regulation

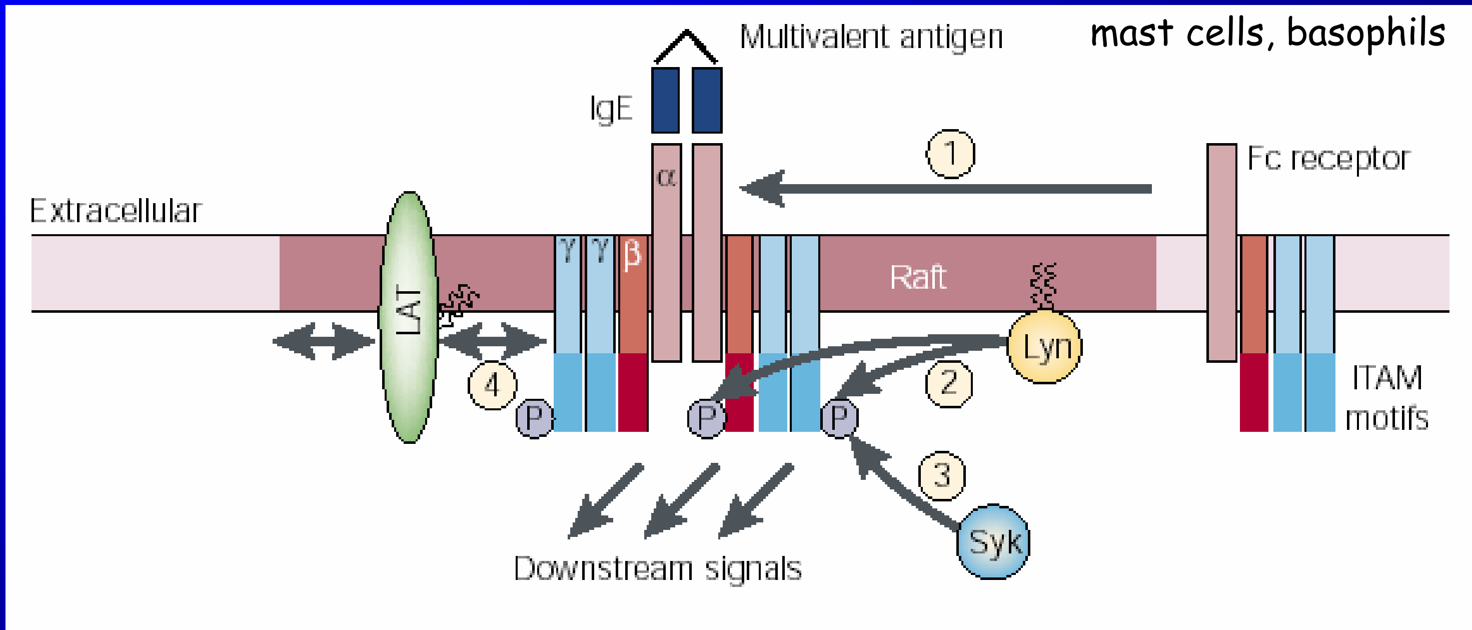
Zajchowski & Robbins (2002) Eur. J. Biochem. 269, 737-752.

Signal transduction processes involving rafts

FcεRI receptor
T-cell receptor
B-cell receptor
EGF receptor
Insulin receptor
EphrinB1 receptor
Neurotrophin
GDNF
Hedgehog
H-Ras
Integrins
eNOS

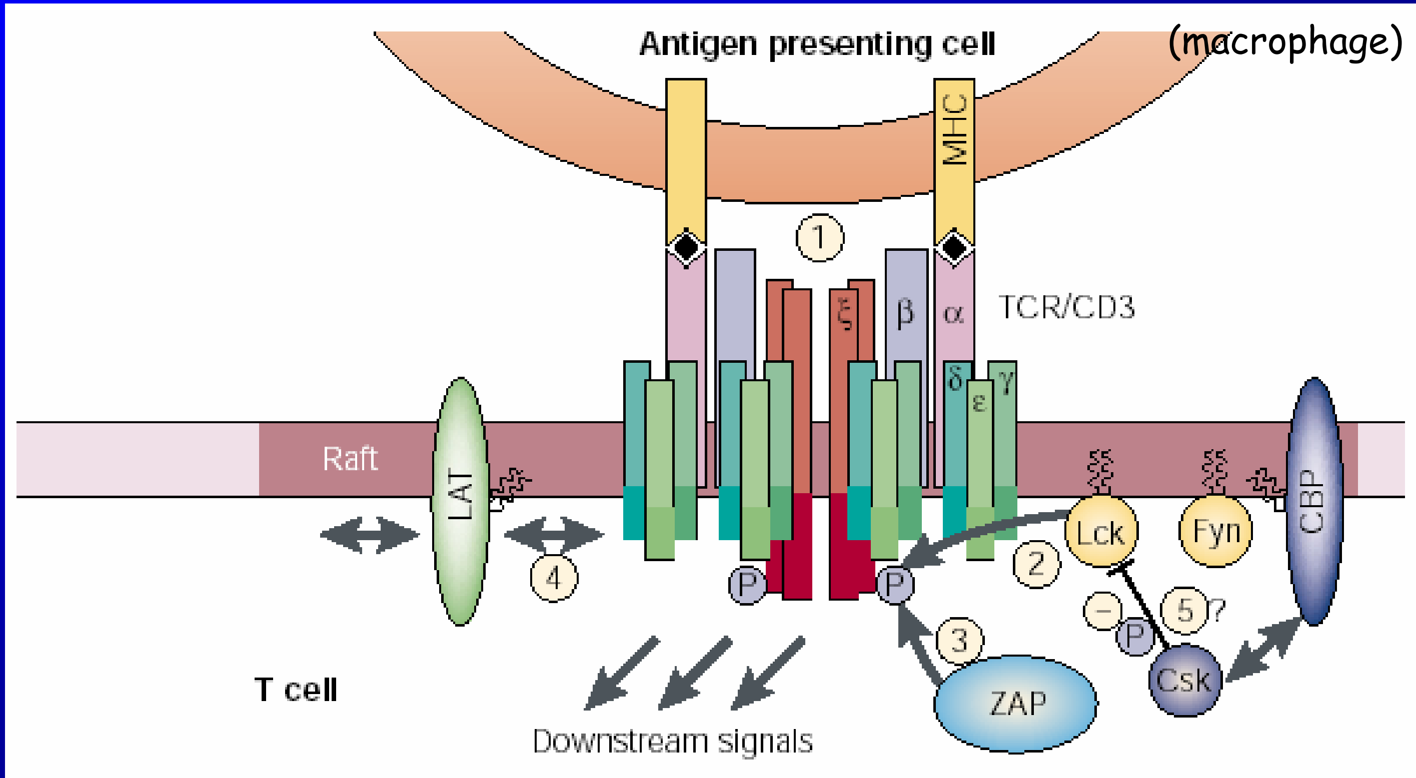
Simons & Toomre (2000) Nat. Rev. Molec. Cell Biol. 1, 31-40.

IgE receptor (FcεRI)-mediated signalling in allergic immune response



Simons & Toomre (2000) Nat. Rev. Molec. Cell Biol. 1, 31-40.

T-cell antigen receptor (TCR)-mediated activation of T lymphocite



Simons & Toomre (2000) Nat. Rev. Molec. Cell Biol. 1, 31-40.

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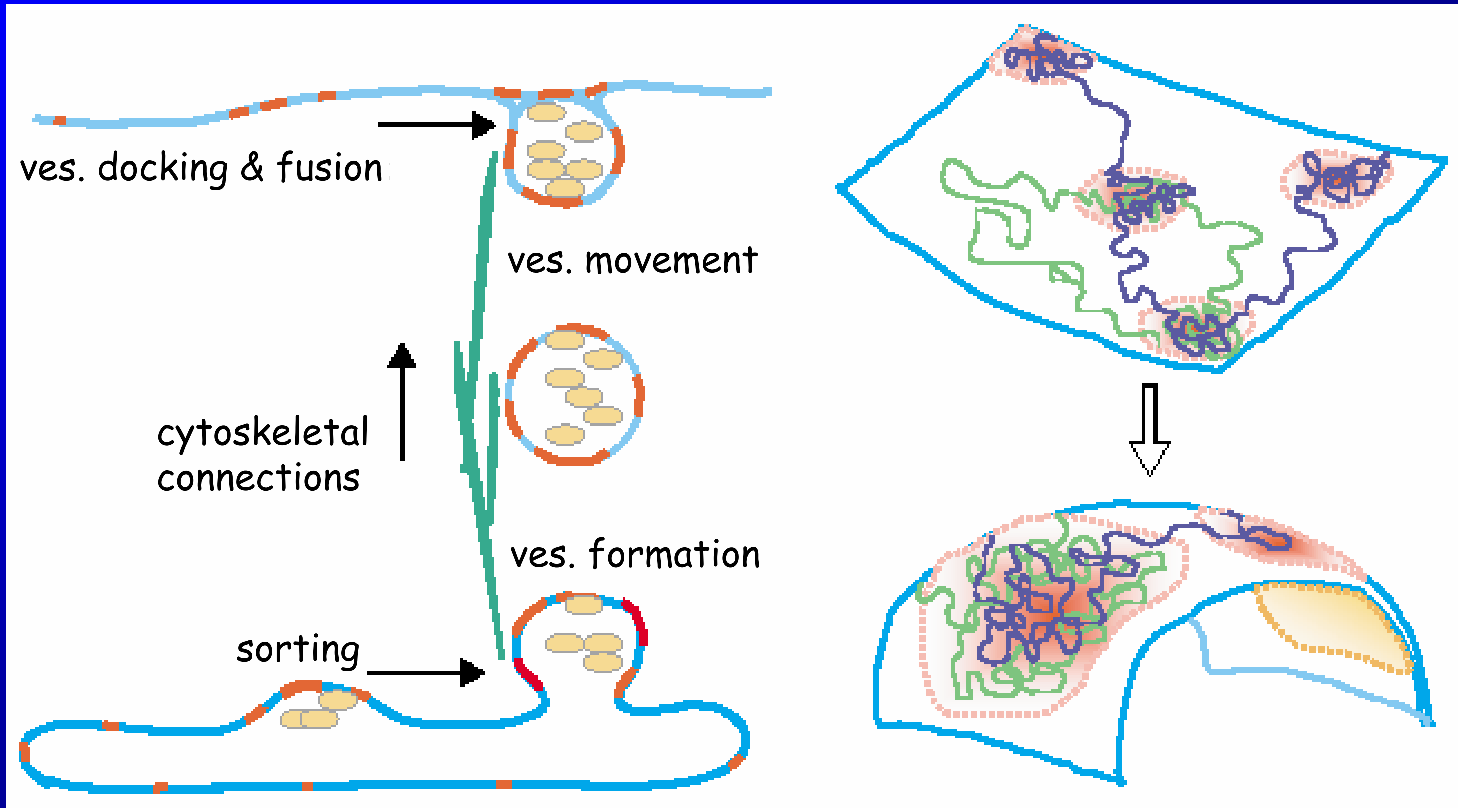
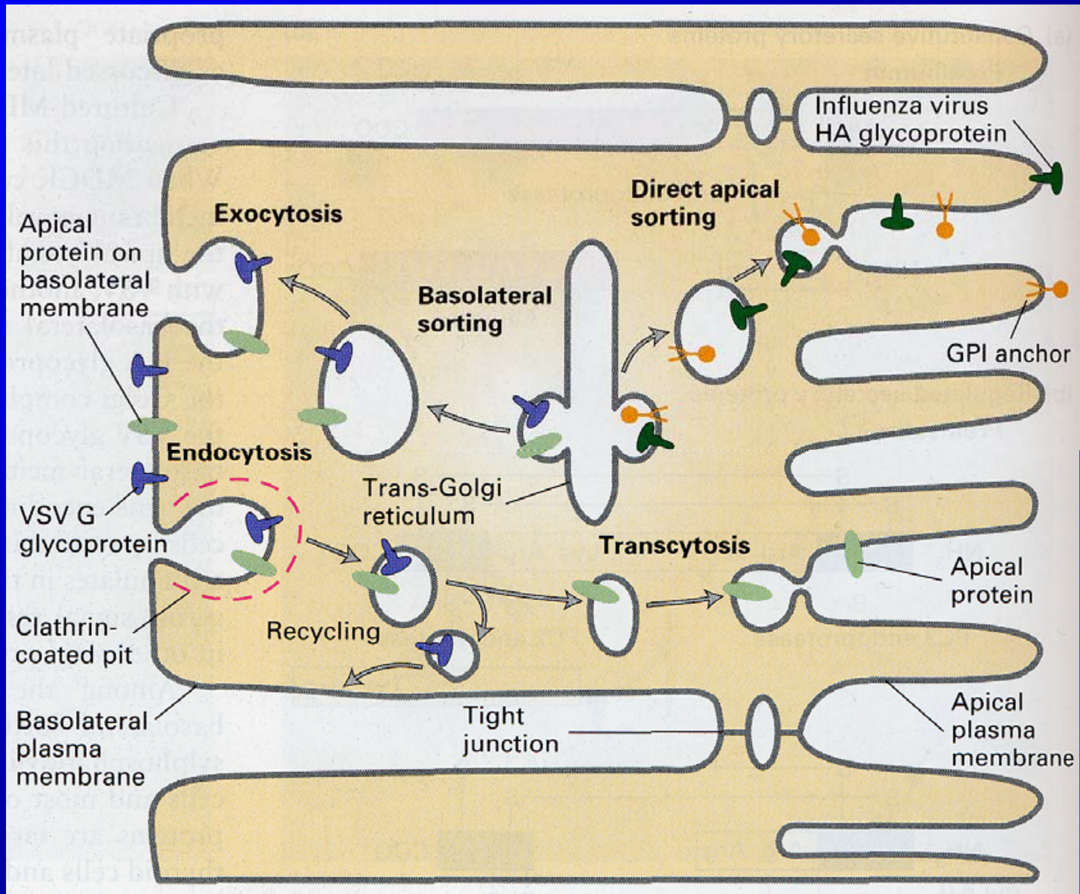
Potential roles of lipid rafts in vesicular transport



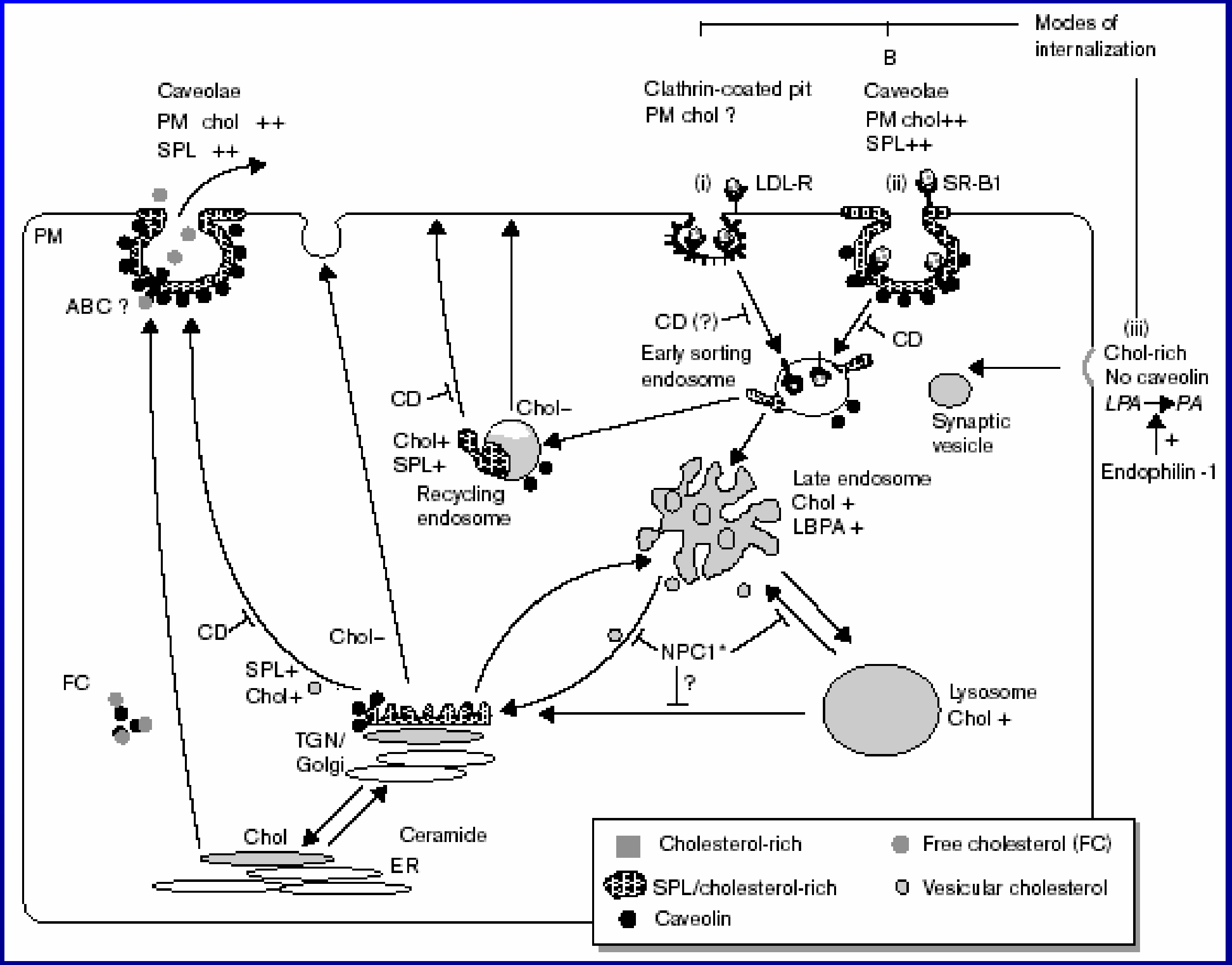
Ikonen (2001) Curr. Opin. Cell Biol. 13, 470-477.

The sorting of proteins in polarized cells
(*e.g.* MDCK epithelial cell)



MCB - Chapter 17

Lipid rafts are involved in cholesterol and sphingolipid traffic



Hoekstra et al. (2001) Curr. Opin. Cell Biol. 12, 496-502.

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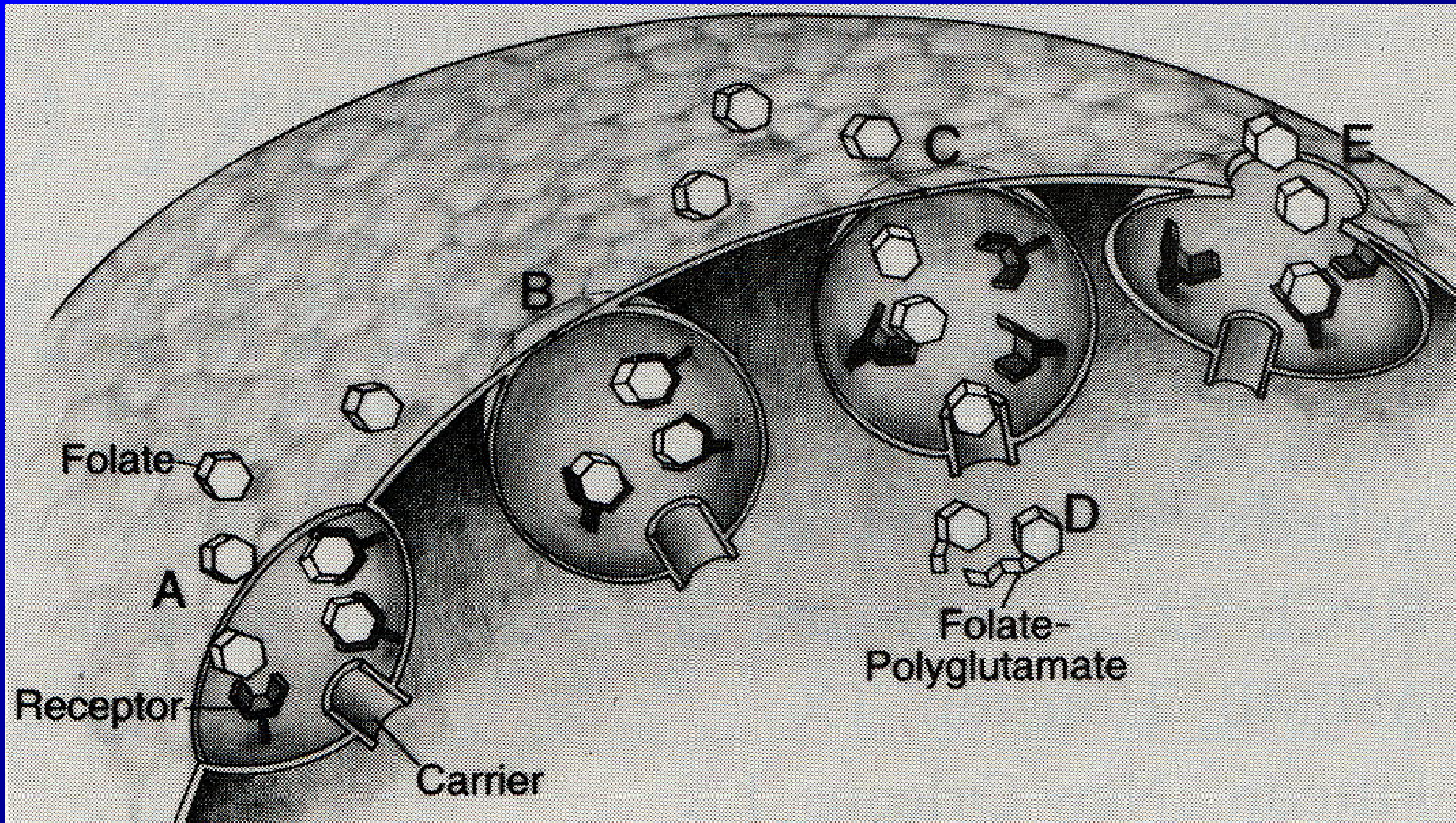
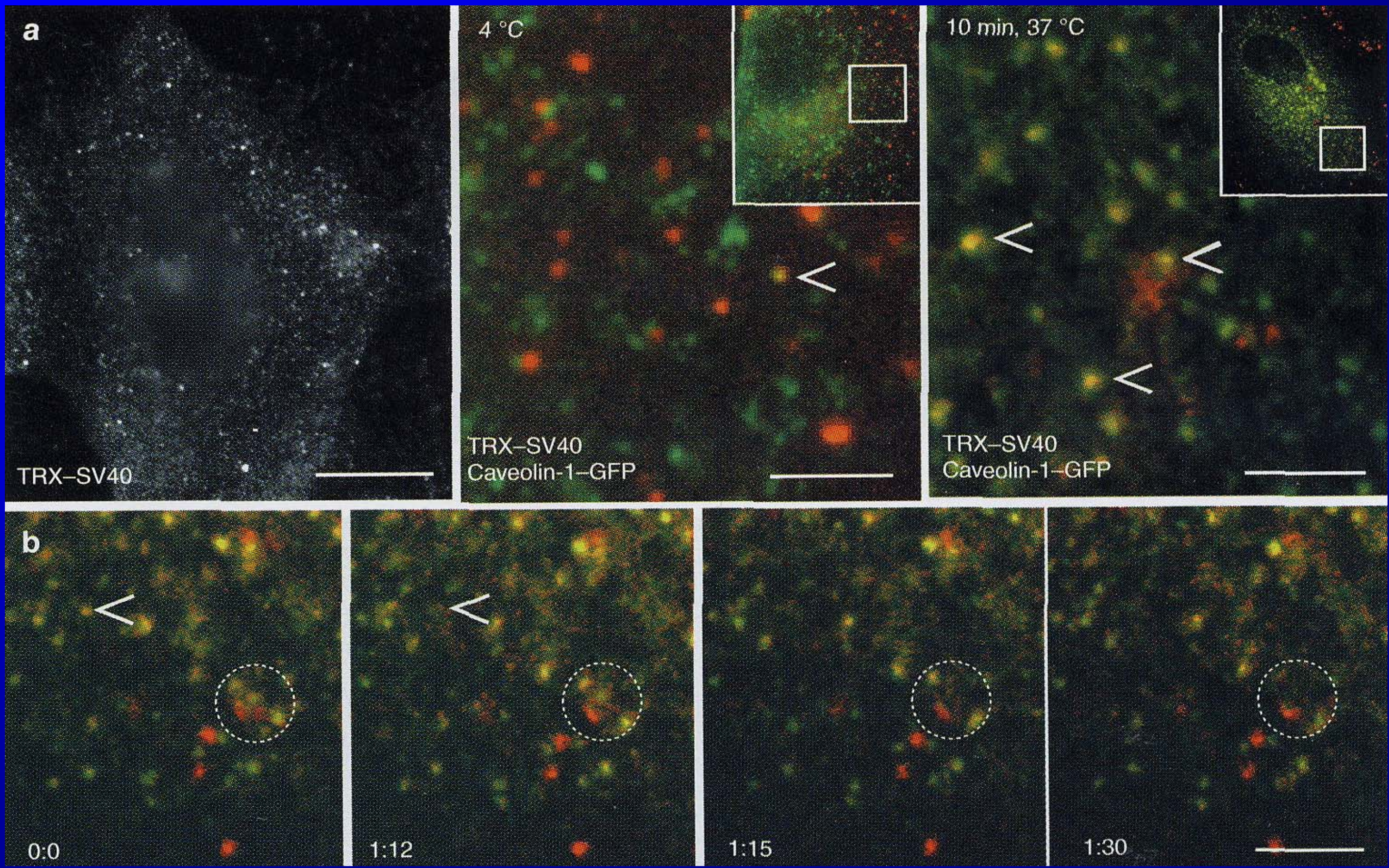
Potocytosis:

sequestration and internalization of molecules and ions by caveolae



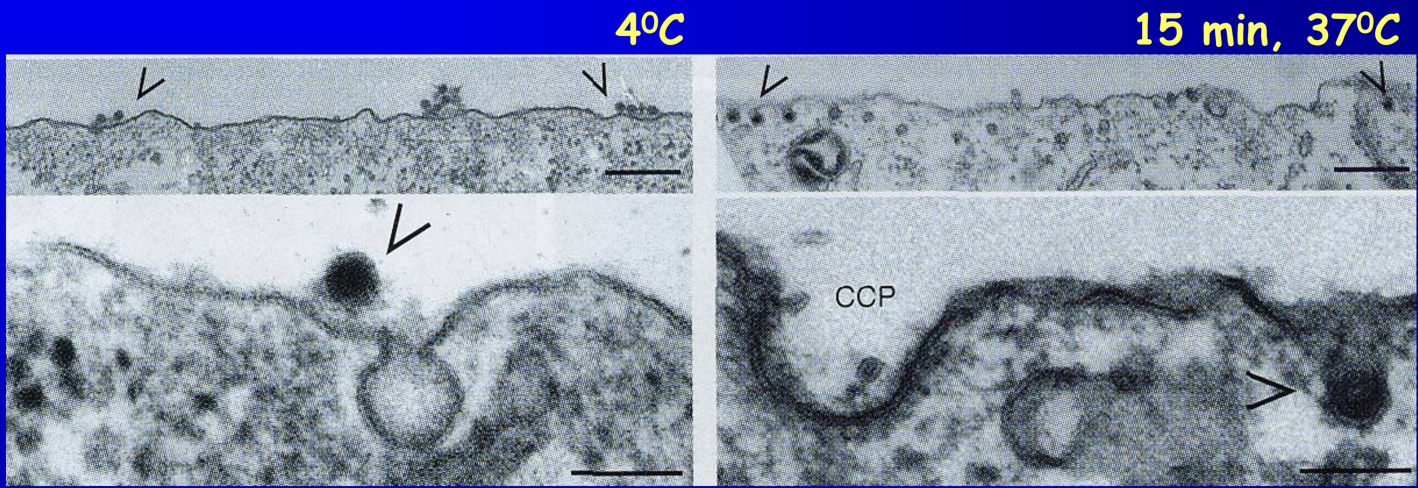
Anderson et al. (1992) Science 255, 410-411.

Caveolar endocytosis of simian virus 40 (SV40) by CV-1 cells



Pelkmans et al. (2001) Nat. Cell Biol. 3, 473-483.

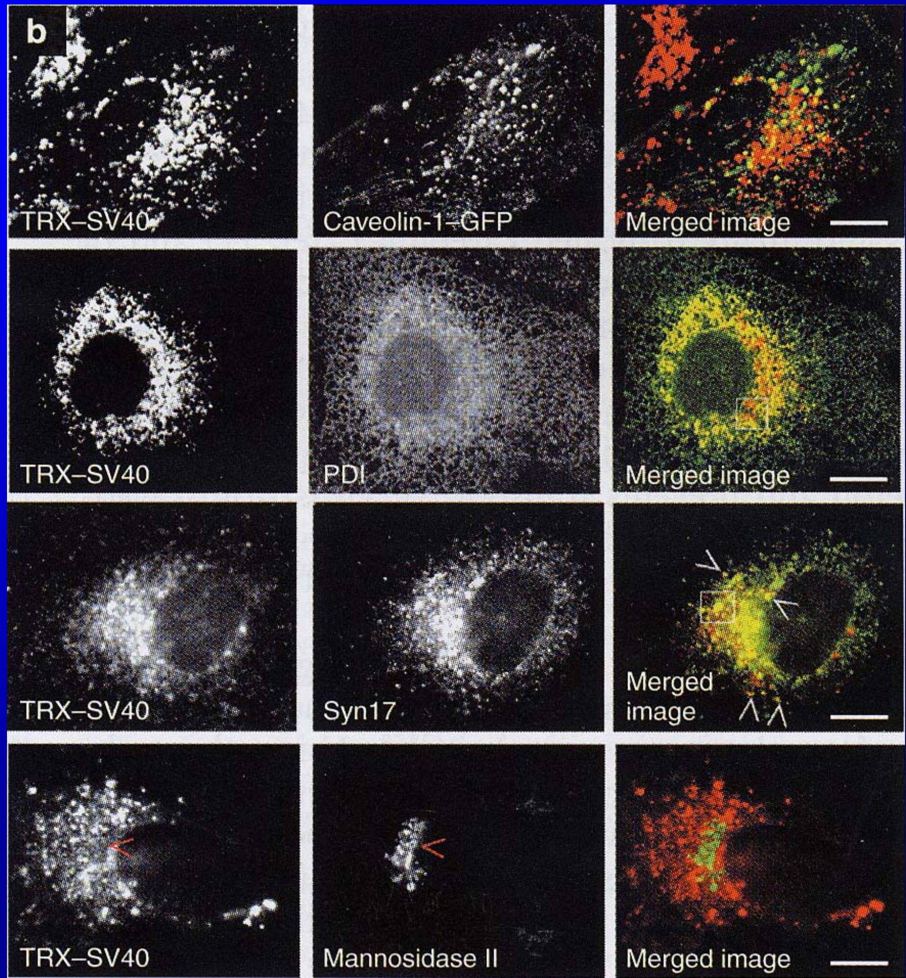
Caveolar endocytosis of SV40 by CV-1 cells



Pelkmans et al. (2001) Nat. Cell Biol. 3, 473-483.

Intracellular localization of SV40 in CV-1 cells

(16h at 37°C after virus binding)



Cav-1 organelles
9 % overlap

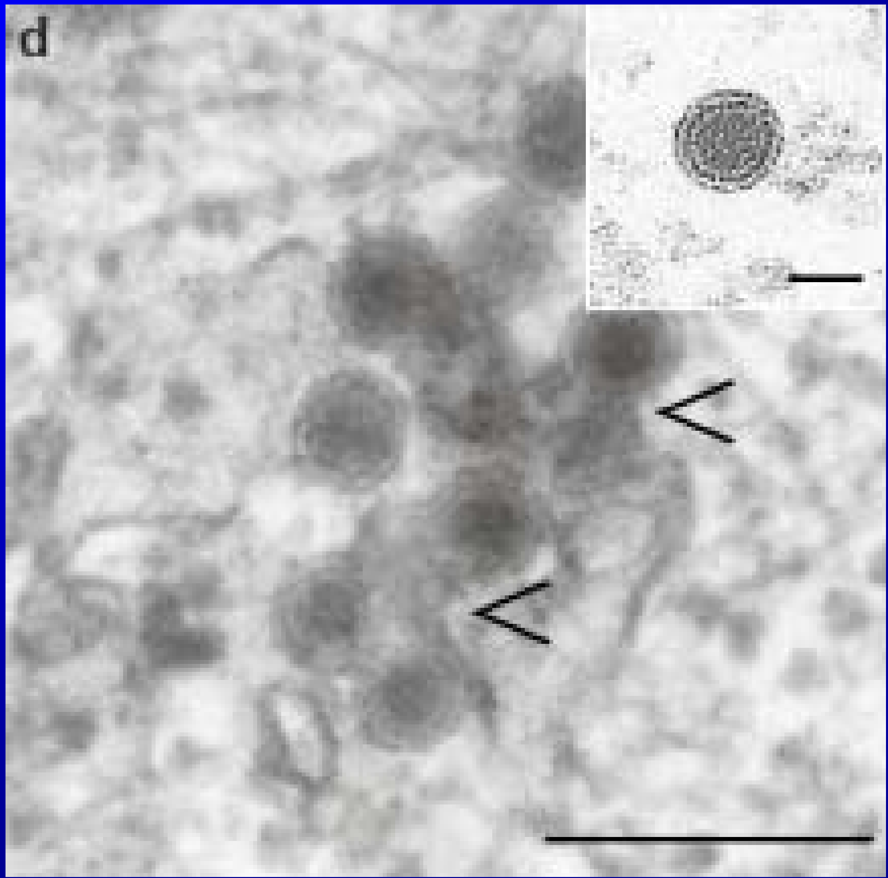
reticular ER
70 % overlap

smooth ER
80 % overlap

Golgi
5 % overlap

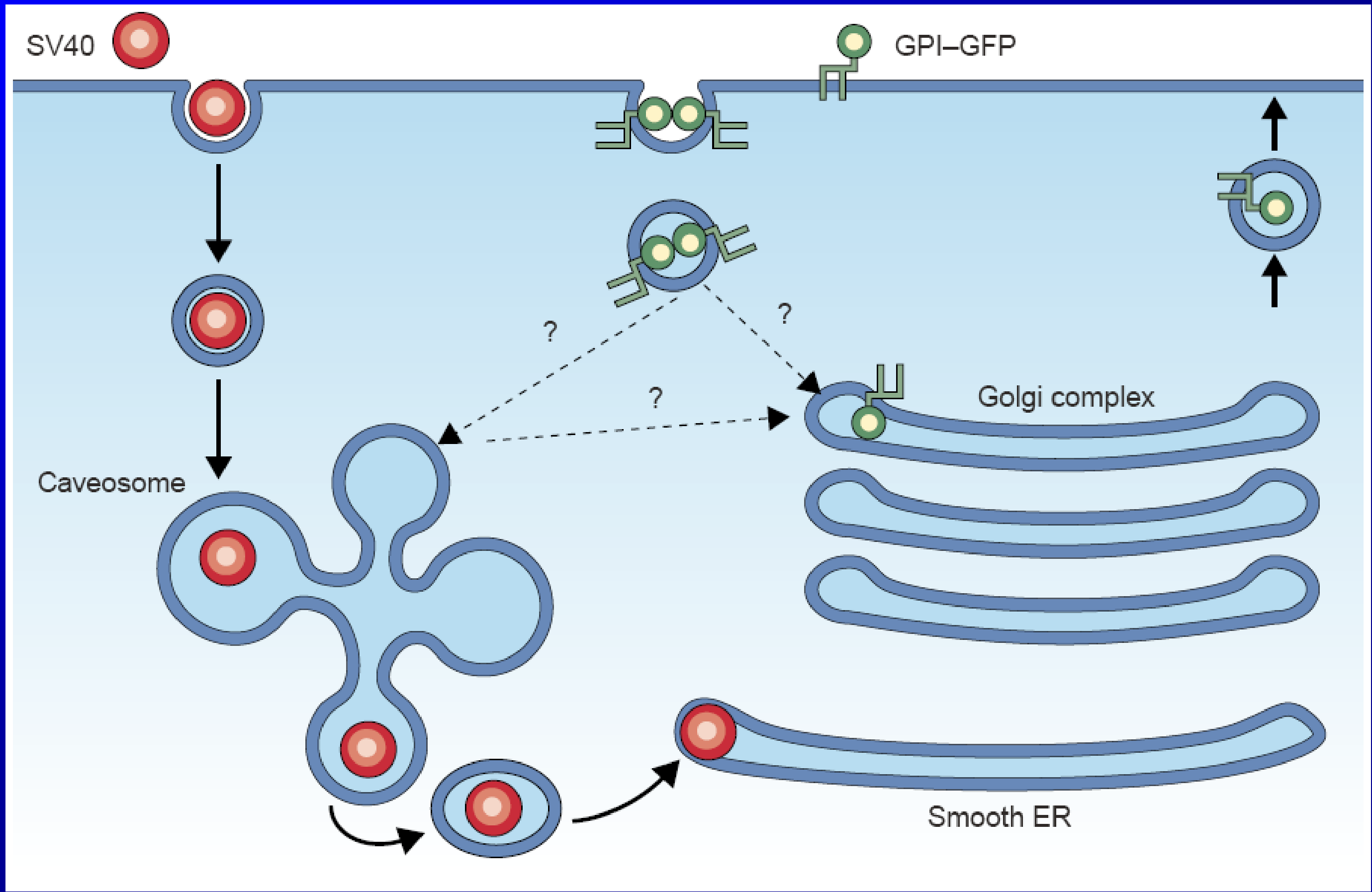
Pelkmans et al. (2001) Nat. Cell Biol. 3, 473-483.

A two-step transport from PM caveolae to ER, through an intermediate organelle - caveosome



Pelkmans et al. (2001) Nat. Cell Biol. 3, 473-483.

Endosome-independent routs for endocytic transport to the ER and Golgi

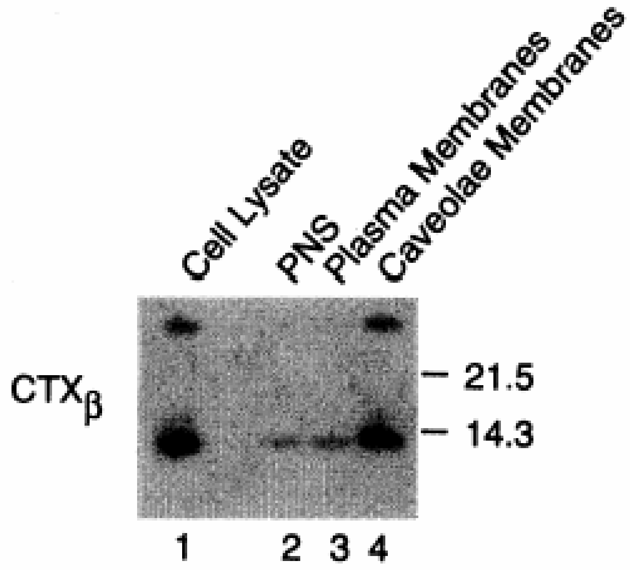


Pfeffer (2001) Nat. Cell Biol. 3, E108-E110.

Caveosome

- does not acidify (neutral pH),
- caveolin-containing compartment,
- lacks coated pit-pathway markers (endosomal, lysosomal, ER or Golgi),
- does not acquire ligands of clathrin-coated vesicle endocytosis.

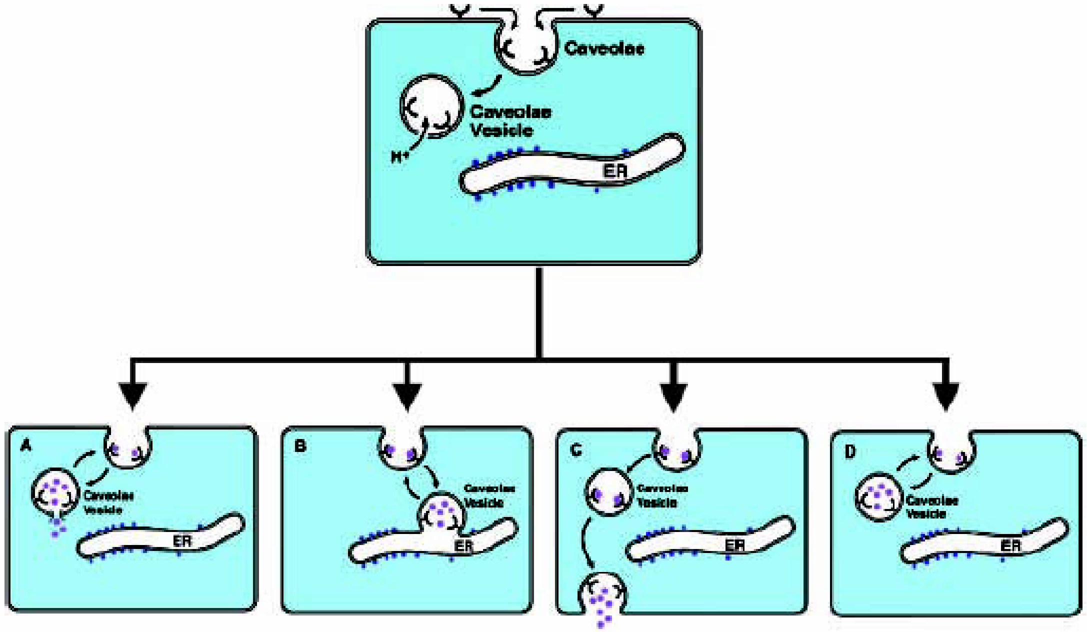
Cholera toxin (CTX) is internalized into hu fibroblasts via caveolae



Detection of bound cholera toxin in purified caveolae. Cells were incubated in the presence of the β subunit of cholera toxin (CTX_β) at 4°C for 1 hr before the cells were fractionated. Immunoblotting (see Fig. 2) was used to detect the β subunit with anti-β subunit IgG.

Smart et al. (1995) Proc. Natl. Acad. Sci. USA 92, 10104-10108.

Four possible fates for molecules internalized by potocytosis



- Folate
- Iron
- Calcium
- Water
- Retinol

- Cholesterol
- SV40 Virus
- AGE
- CD59
- bFGF
- AMF
- GH
- CT

- Albumin
- Transthyretin
- Thrombomodulin
- Plasma Proteins
- Water

- AP
- Bradykinin
- Acetylcholine
- Cholecystokinin
- Endothelin
- gp114

Mineo & Anderson (2001) Histochem. Cell Biol. 116, 109-118.

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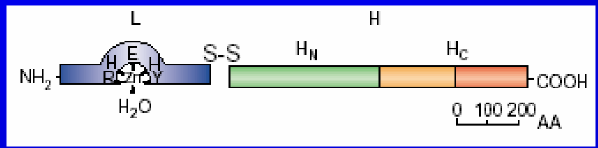
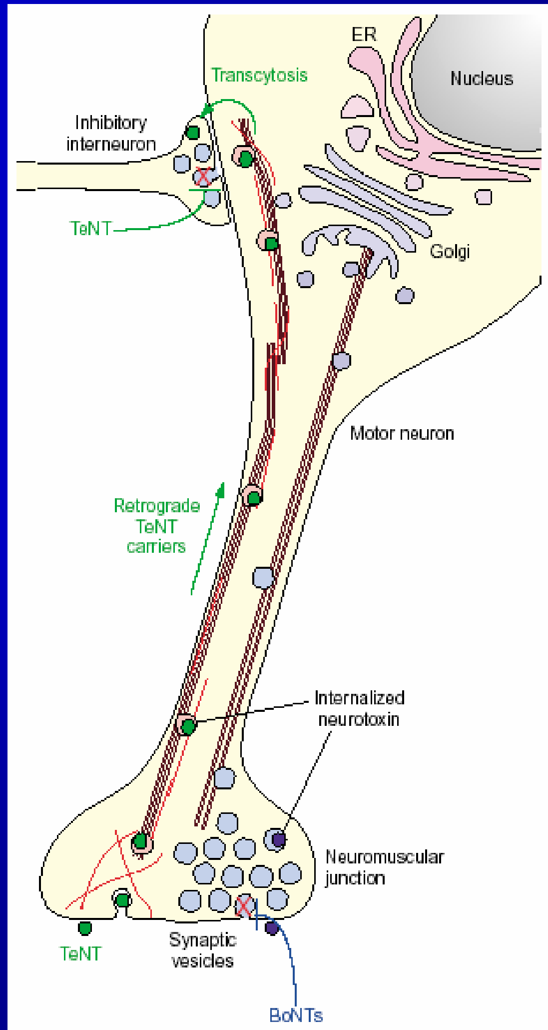
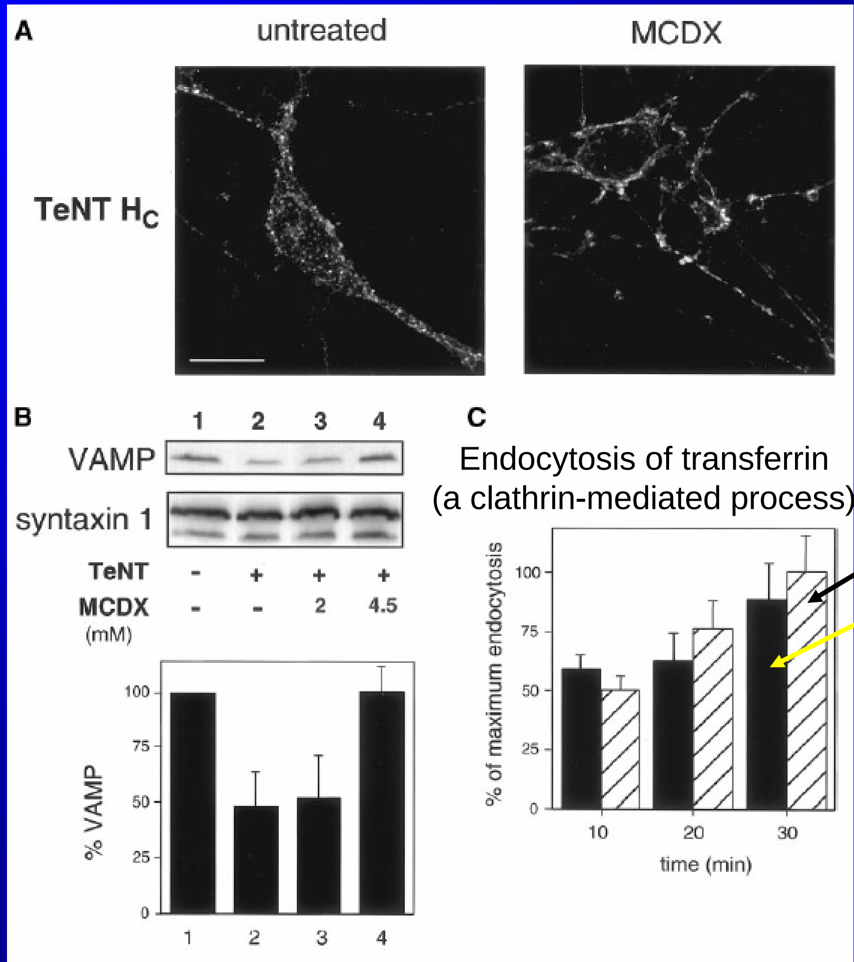
Clathrin-independent receptor-mediated endocytosis of TeNT





Lalli et al. (2003) TRENDS in Microbiol. 11, 431-437.

Cholesterol depletion (raft disruption) blocks the internalization and intracellular activity of TeNT



Spinal cord cells
(neurons lack caveolae)

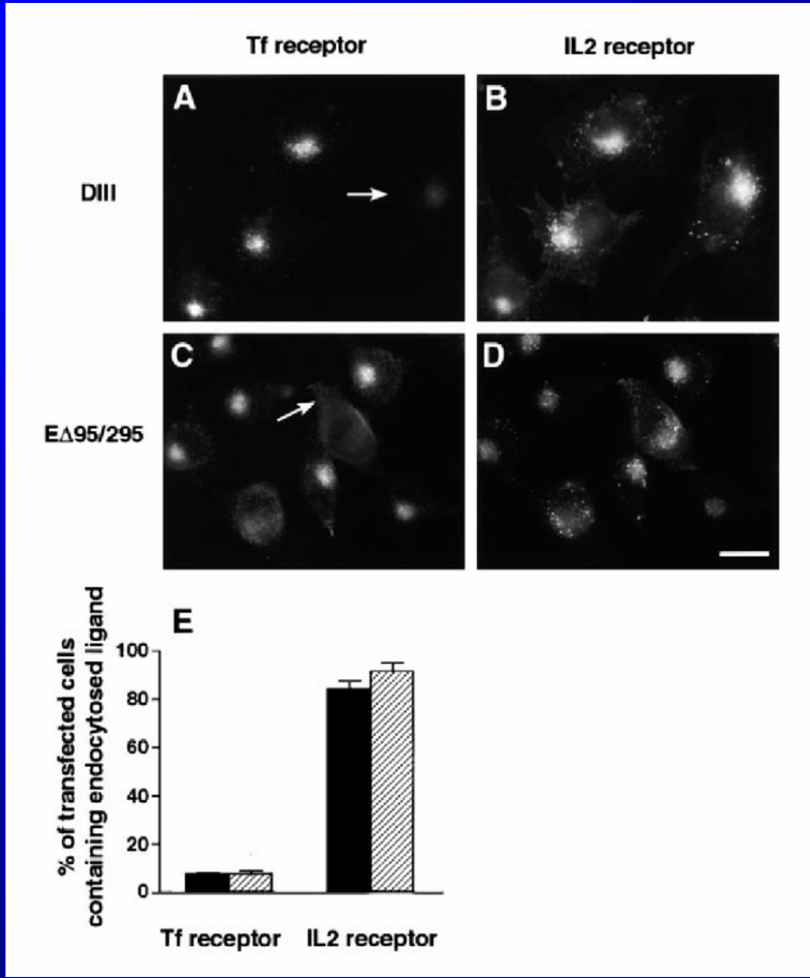
MCDX-treated
MCDX-untreated

TeNT receptor is
a GPI-protein Thy-1

Herreros et al. (2001) Mol. Biol. Cell 12, 2947-2960.

Clathrin-independent receptor-mediated endocytosis of IL2

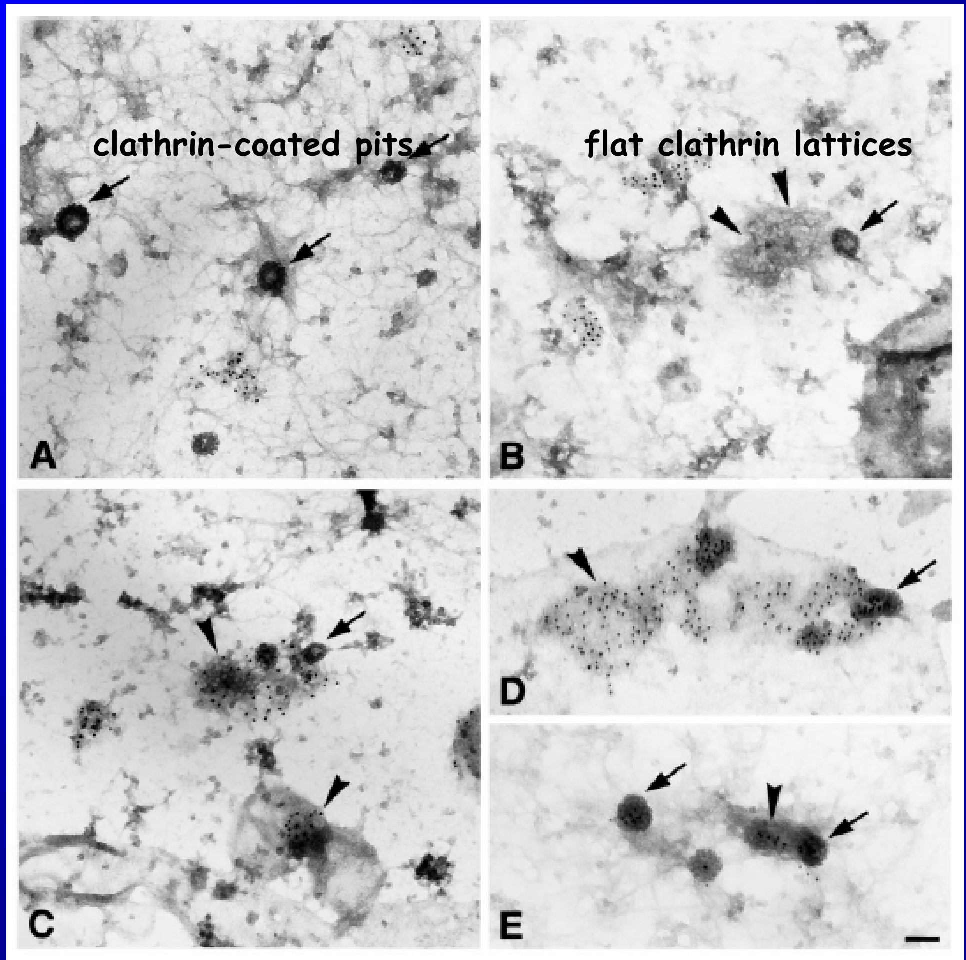
Eps15 mutants - specific inhibitors of clathrin-dependent endocytosis



(mouse fibroblasts)

Lamaze et al. (2001) Mol. Cell 7, 661-671.

Tf receptors are localized in clathrin-coated structures in YT lymphocite PM while IL2 receptors are not



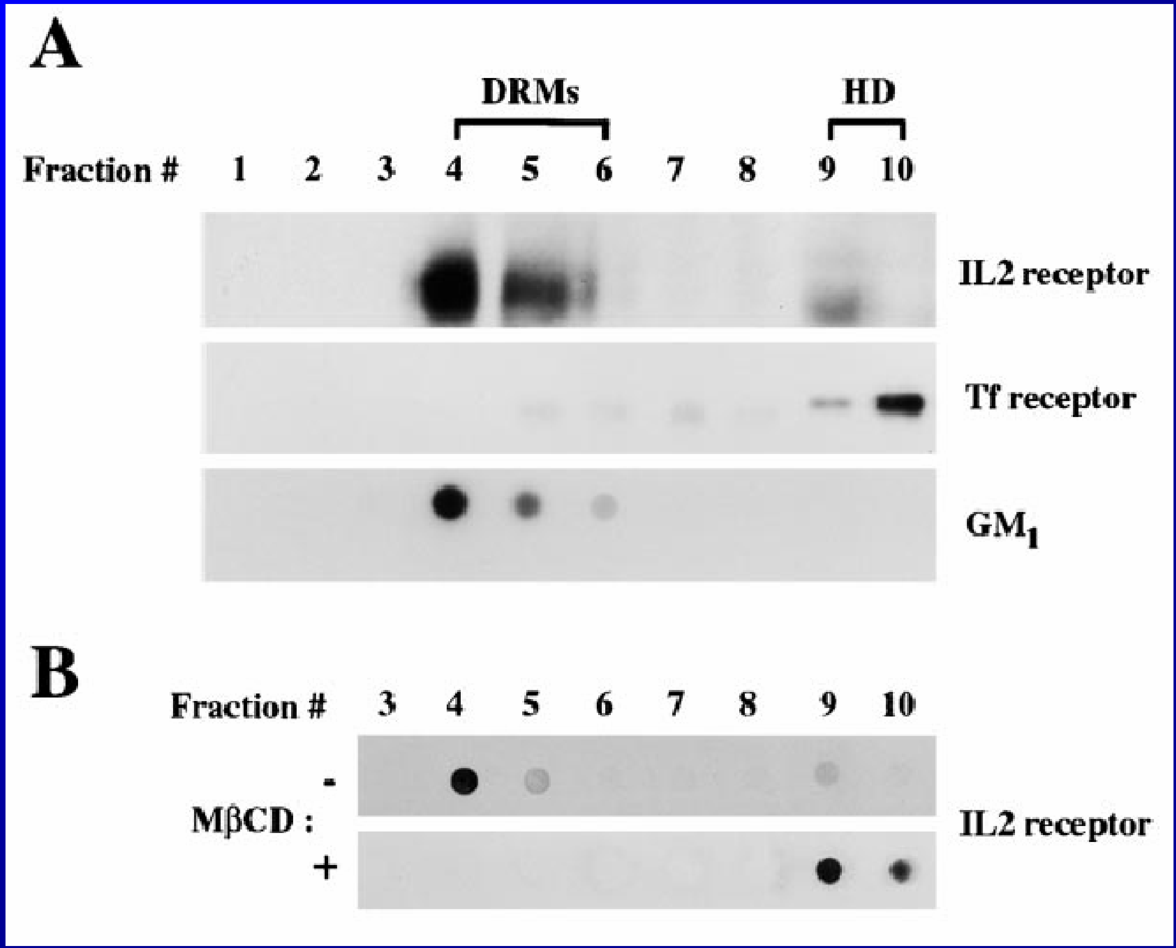
anti-IL2R mAb

anti-TfR mAb

Lamaze et al. (2001) Mol. Cell 7, 661-671.

IL2 receptors are concentrated in lipid rafts

(YT lymphocytes lack caveolae)



Lamaze et al. (2001) Mol. Cell 7, 661-671.

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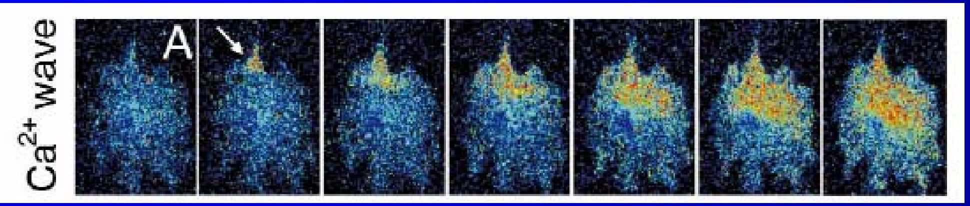
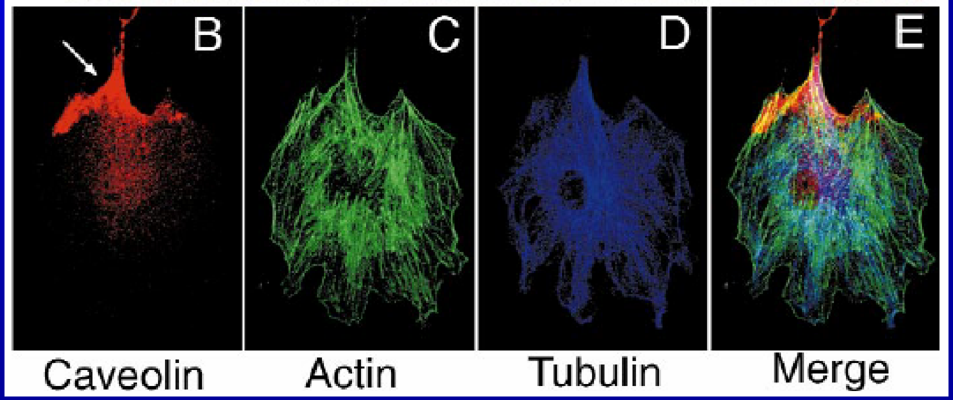
Caveolae are enriched in molecules involved in Ca^{2+} regulation: IP_3R-like protein and Ca^{2+}-ATPase.

ATP stimulation ⇨ IP_3 mobilization

Indo-1 loaded endothelial cell (bovine aortic)

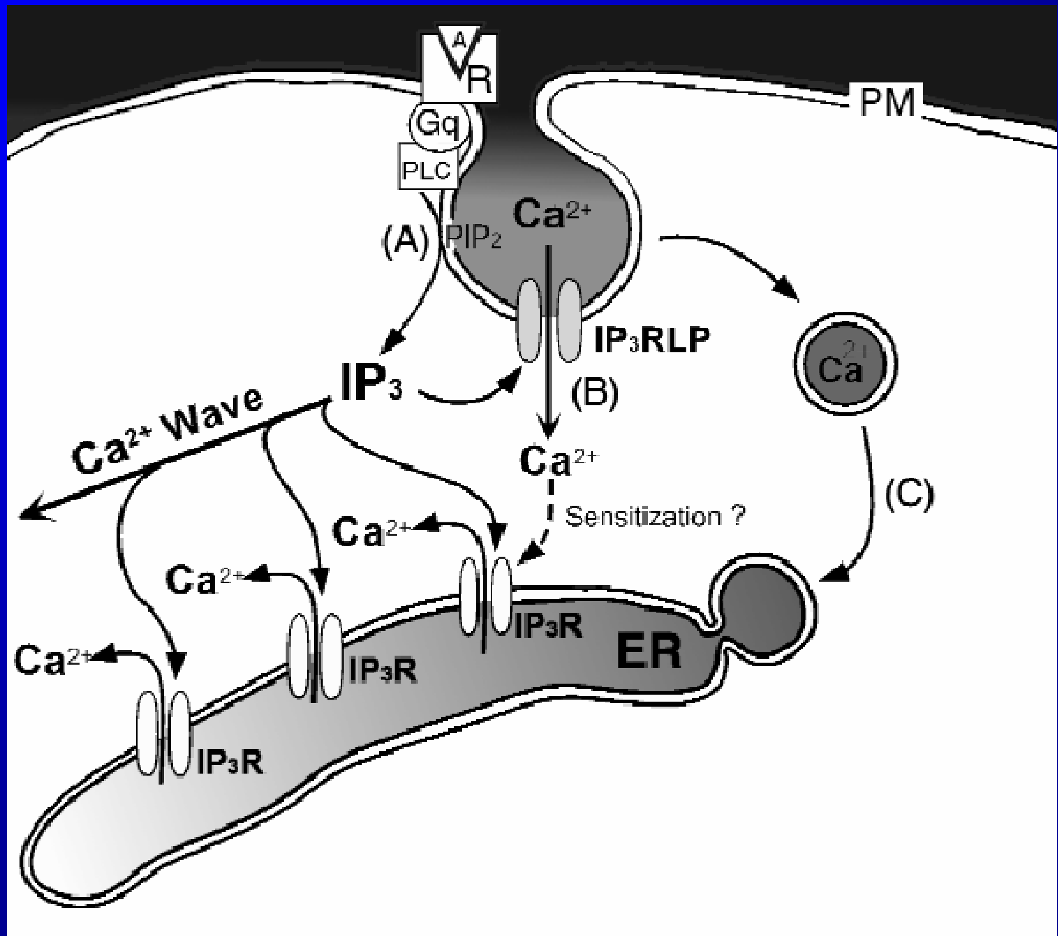


0.34 s intervals



Isshiki & Anderson (1999) Cell Calcium 26, 201-208.

Three ways for how caveolae might regulate Ca^{2+} wave initiation



Isshiki & Anderson (1999) Cell Calcium 26, 201-208.

Key functions for caveolae in Ca^{2+} homeostasis

- regulation of the spatial organization of Ca^{2+} entry sites,

- control of the amount of Ca^{2+} that is delivered at these sites,

- initiation of Ca^{2+} wave formation,

- modulation of Ca^{2+}-dependent signalling cascades in caveolae (*e.g.* eNOS/CaM^{+}/$caveolin^{-}$).

Lipid rafts and human disease

- Muscular distrophy (cav-3 mutation)
- Alzheimer`s disease (generation of β-amyloid)
- Encephalopathies (a conversion of Pr^{C} to Pr^{Sc} in caveolae)
- Cancer (loss of caveolin-1, *i.e.* caveolae)
- Pathogens (cellular entrance point)
- Cardiovascular diseases

Recommended reading:

Ikonen, E. (2001): Roles of lipid rafts in membrane transport. Curr. Opin. Cell Biol. 13, 470-477.

Isshiki, M. and Anderson, R.G.W. (1999): Calcium signal transduction from caveolae. Cell Calcium 26, 201-208.

Lamaze, C. et al. (2001): Interleukin 2 receptors and detergent-resistant membrane domains define a clathrin-independent endocytic pathway. Mol. Cell 7, 661-671.

Mineo, C. and Anderson, R.G.W. (2001): Potocytosis. Histochem. Cell Biol. 116, 109-118.

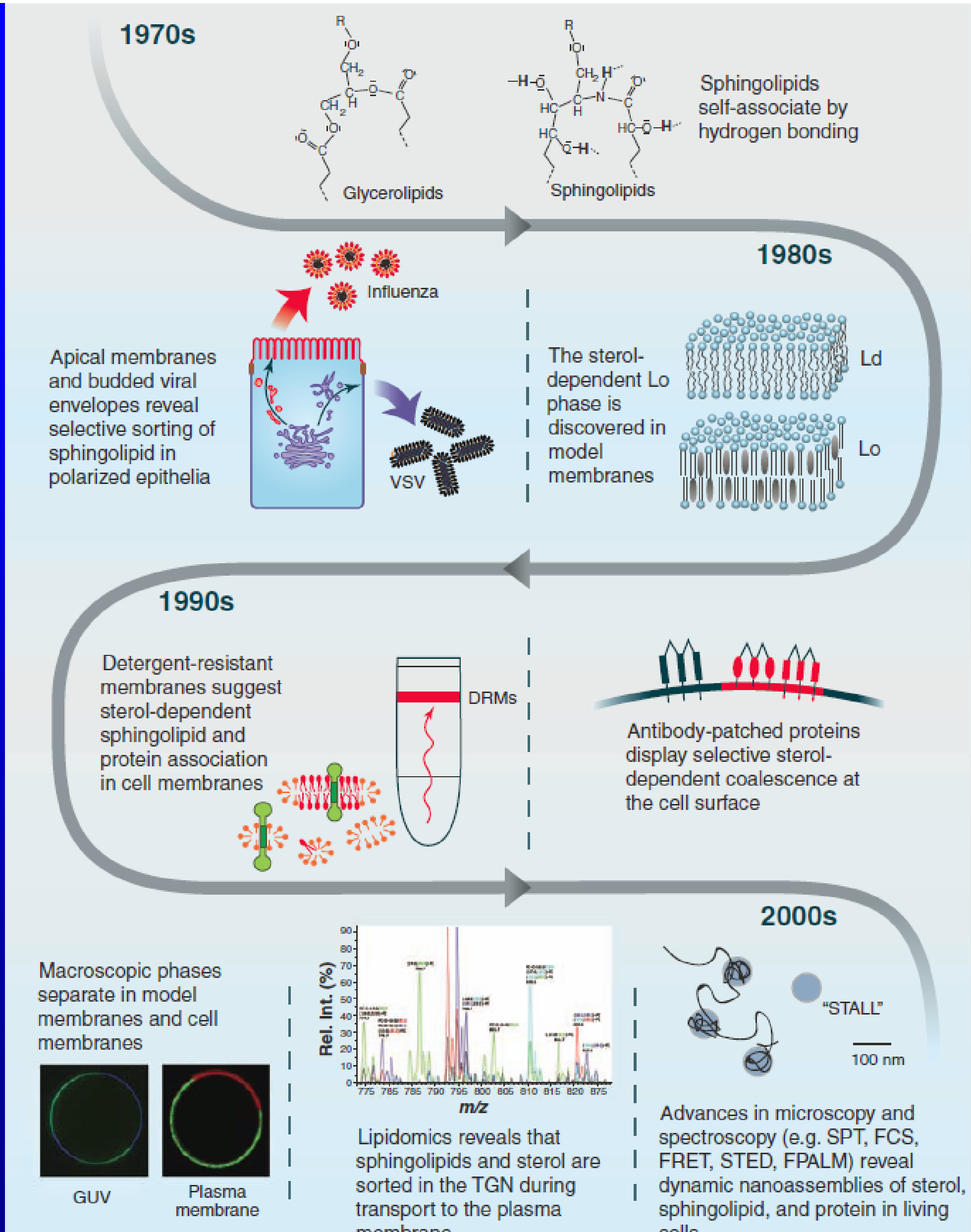
Parton, R.G. (2001): Life without Caveolae. Science 293, 2404-2405.

Pelkmans, L. et al. (2001): Caveolar endocytosis of simian virus 40 reveals a new two-step vesicular-transport pathway to the ER. Nat. Cell Biol. 3, 473-483.

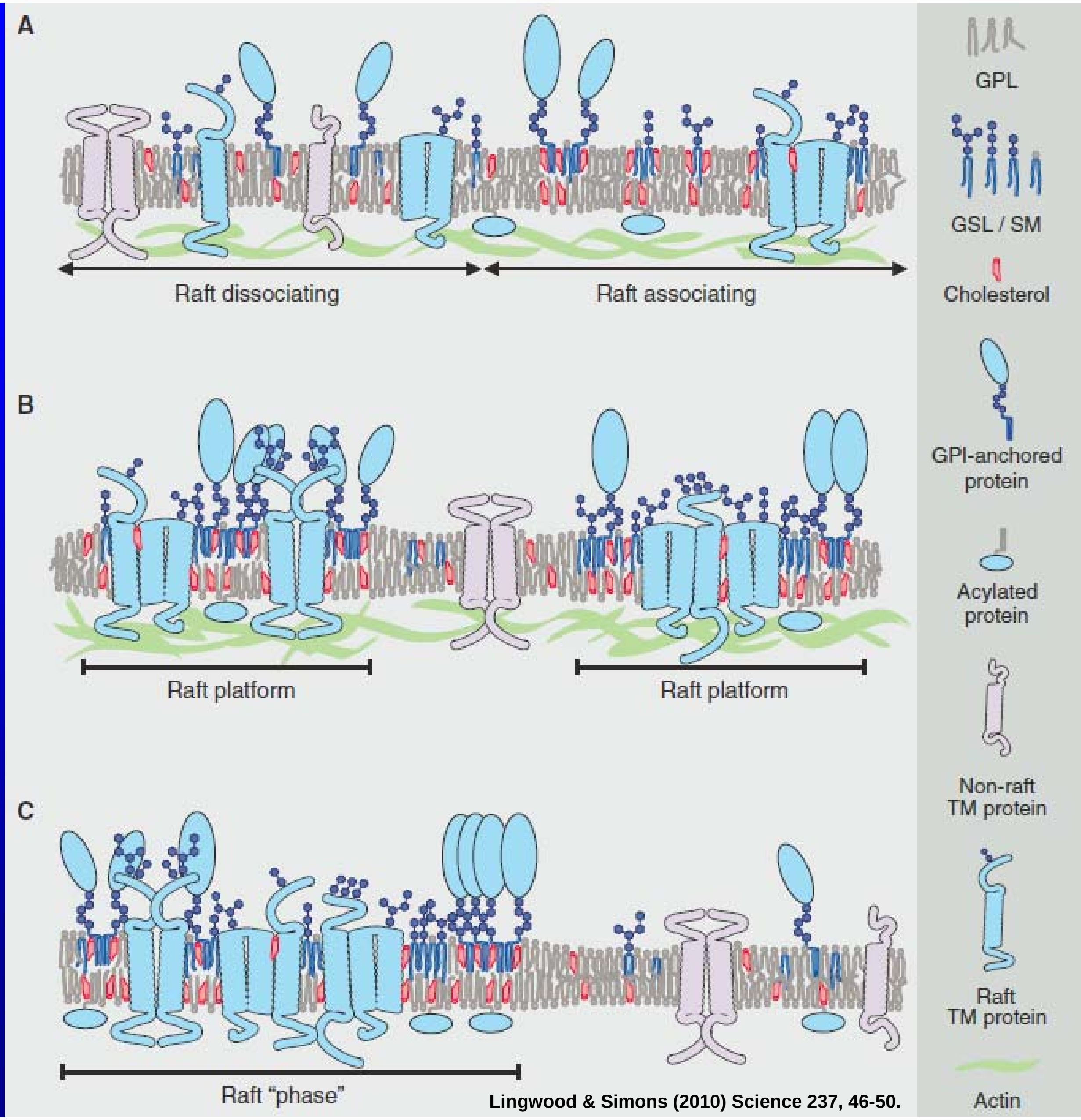
Simons, K. and Toomre, D. (2000): Lipid rafts and signal transduction. Nat. Rev. Mol. Cell Biol. 1, 31-40.

Sprong, H. et al. (2001): How proteins move lipids and lipids move proteins. Nat. Rev. Mol. Cell Biol. 2, 504-513.

Zajchowski, L.D. and Robbins, S.M. (2002): Lipid rafts and little caves. Eur. J. Biochem. 269, 737-752.



Lingwood & Simons (2010) Science 237, 46-50.



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