# Morph Complexity and Topological Transformations of Membranes

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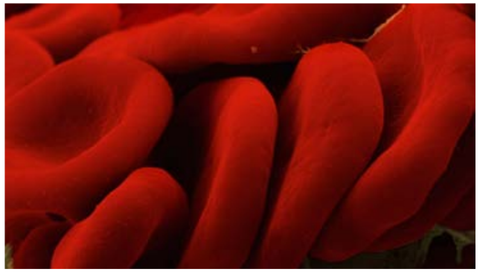
- Shape of Membranes
- Giant Vesicles and Nanovesicles
- Morph Complexity ~ Membrane Necks
- Topological Transformations
- Fission of Membrane Necks

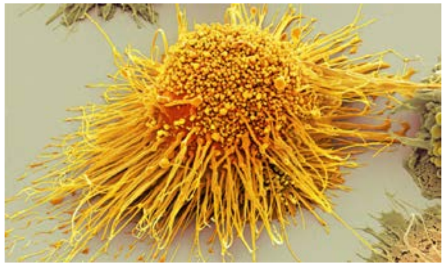
# Shape of Cellular Membranes

- Plasma membranes:

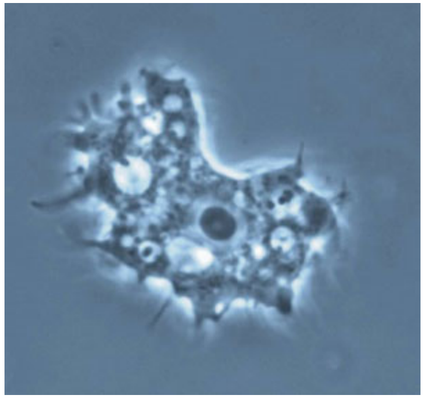
Red blood cells



White blood cell

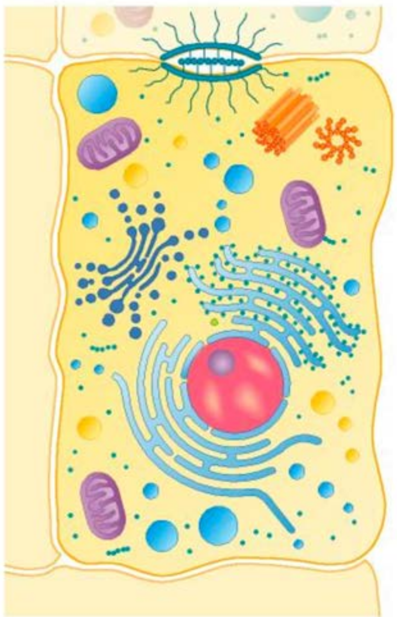


Single Purkinje cell

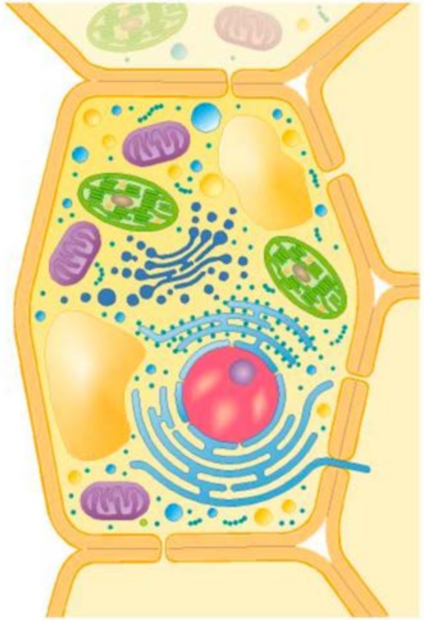


Amoeba

- Intracellular membranes:

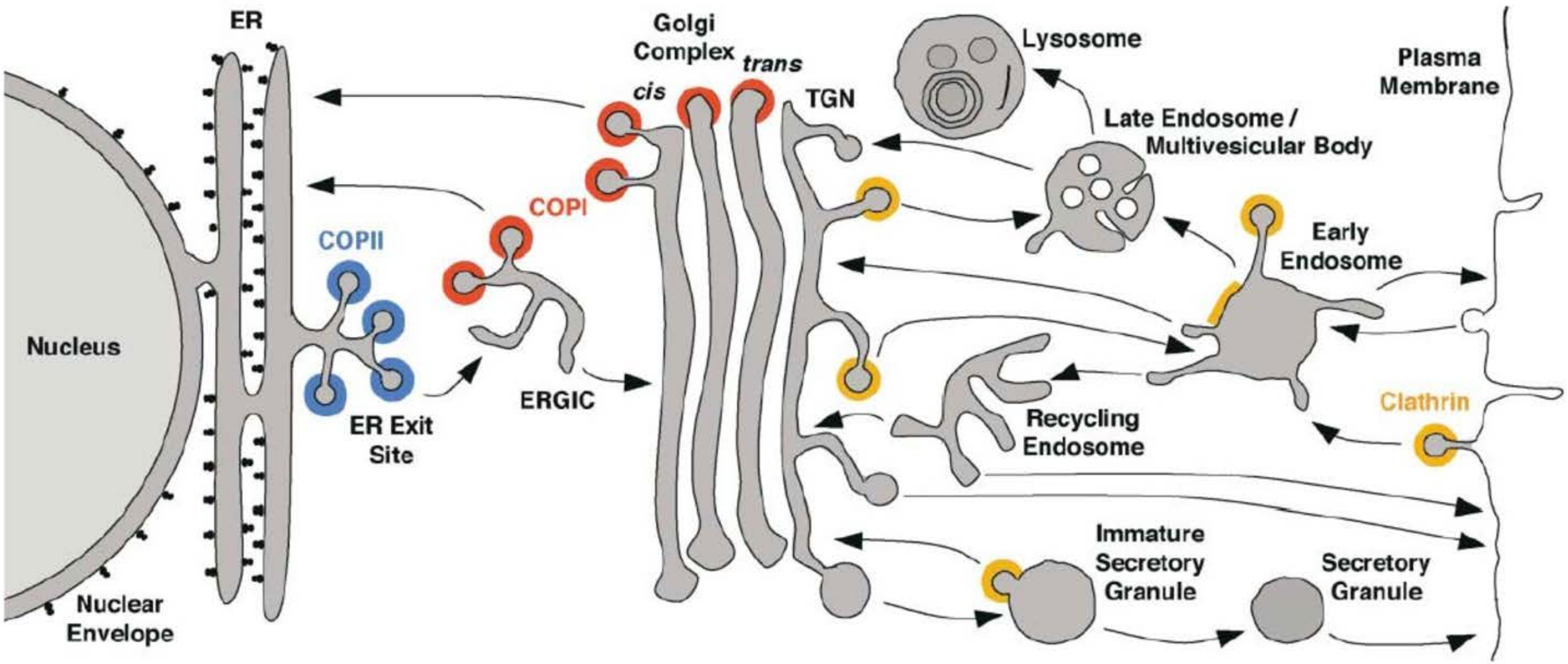


Animal



Plant

# Intracellular Vesicle Trafficking



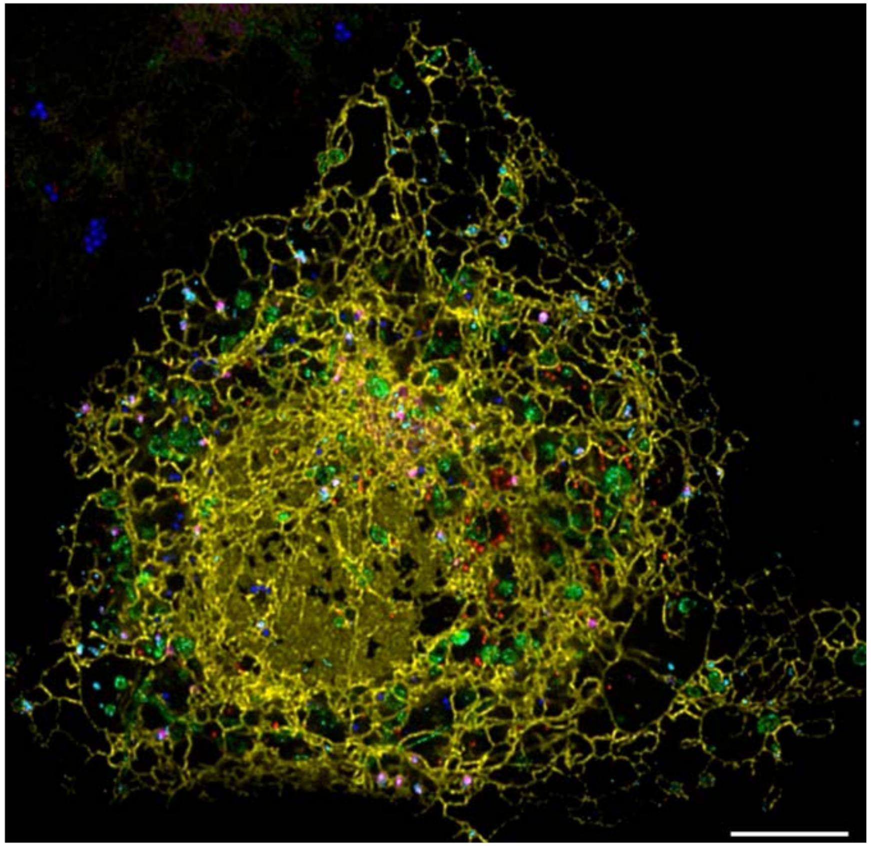

Bonifacino, Glick, *Cell* (2004)

- Colored dots indicate budding of organelle membranes
- Budding and fission via formation of membrane necks

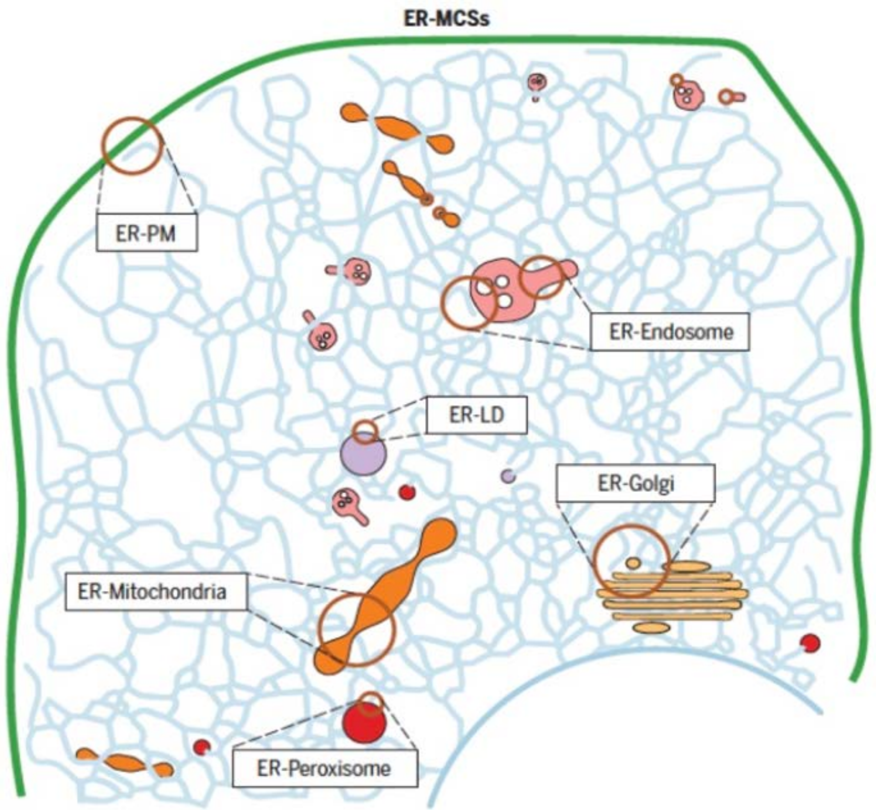
# Endoplasmic Reticulum (ER)

- ER = network of membrane nanotubes with junctions

Valm et al. *Nature* (2017)



reticular network = yellow

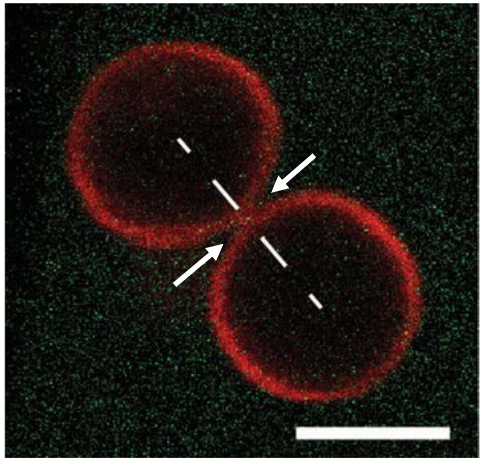



reticular network = light blue

Wu et al, *Science* (2018)

# Synthetic Membrane Compartments

Steinkühler et al, *Nature Comm* (2020)



5 µm

- Giant unilamellar vesicles (GUVs)
- Shape transformations by optical microscopy
- Understanding based on curvature elasticity

- Nanovesicles (SUVs)
- Electron microscopy: limited to a single snapshot for each individual nanovesicle
- Shape transformations by Molecular Dynamics simulations:

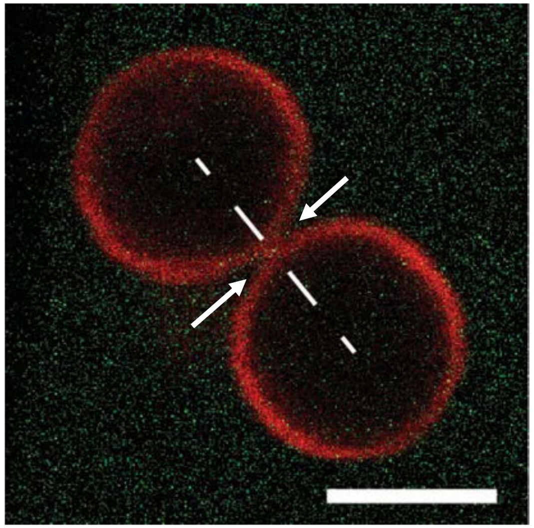
Ghosh, Satarifard et al, *Nano Letters* (2019)

20 nm

- Shape of Membranes
- Giant Vesicles and Nanovesicles
- Morph Complexity ~ Membrane Necks
- Topological Transformations
- Fission of Membrane Necks

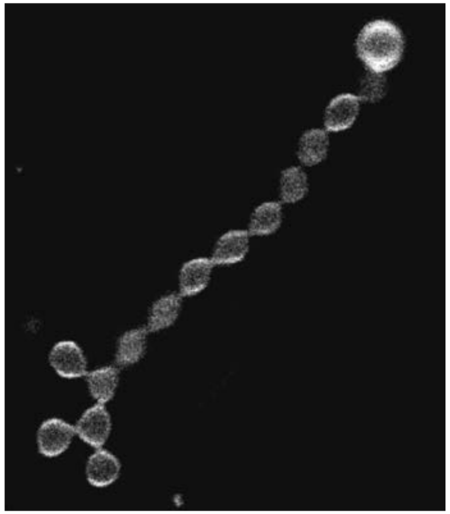
# Morph Complexity of GUVs

- Giant Unilamellar Vesicles (GUVs), size of 5 – 50 μm
- Lipid bilayers, thickness of 4 -5 nm
- Many different shapes with membrane necks:



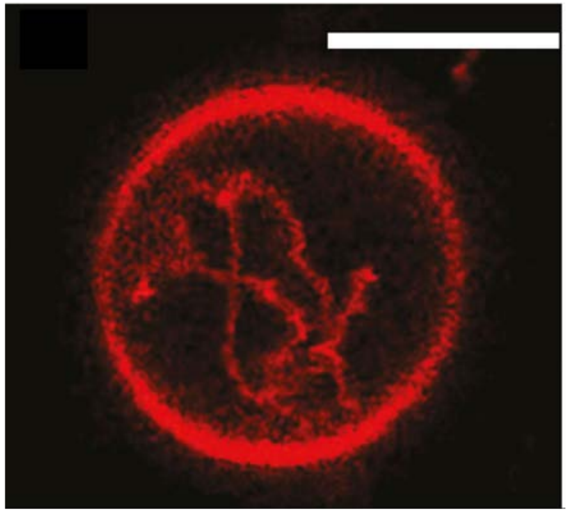
Exposed to His-tagged GFP in exterior solution

Steinkühler et al, *Nature Comm* (2020)



Sucrose inside, glucose outside

Bhatia et al, *Soft Matter* (2020)



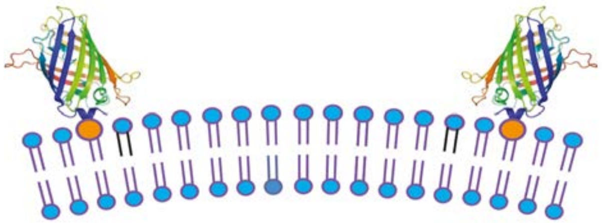
Bilayer contains GM1 with bulky head group

Bhatia et al, *ACS Nano* (2018)

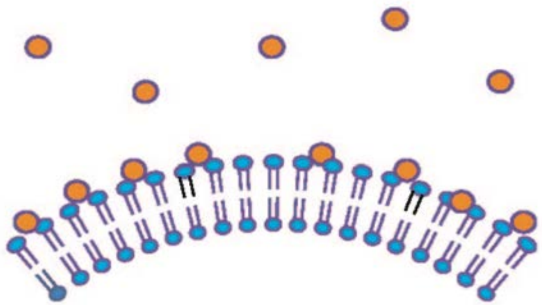
# Key Parameter: Spontaneous Curvature

- Lipid bilayer consists of two leaflets
- Spontaneous or preferred curvature $m$ describes transbilayer asymmetry = asymmetry between two leaflets

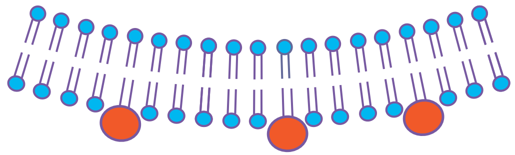
- Different molecular mechanisms for spont curvature:



Binding of GFP to outer leaflet



Adsorption layer of glucose

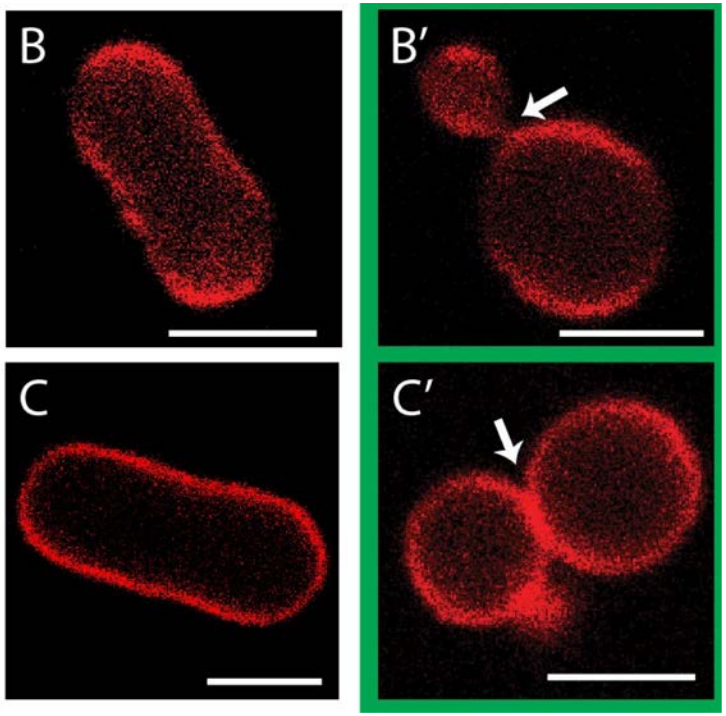


Adsorption of glycolipid GM1

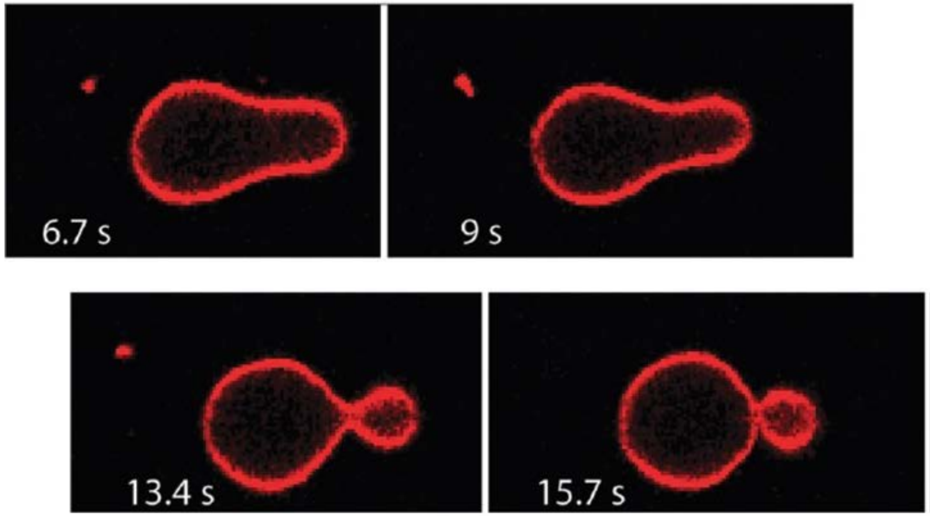
# Membrane Necks or ‘Wormholes’

Neck formation by increase of [GFP]

Steinkühler et al, *Nature Comm* (2020)




Neck formation by osmotic deflation:

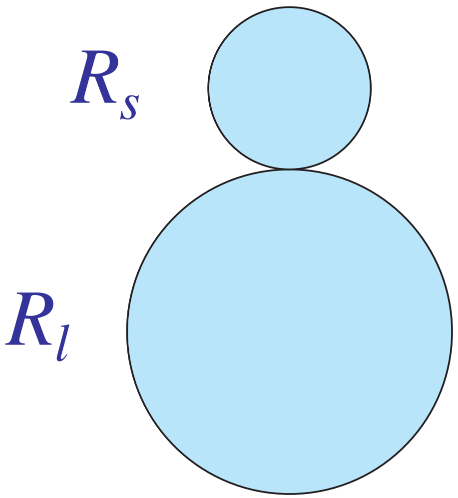



‘Wormhole in 3-dim space’

Bhatia et al, *Soft Matter* (2020)

- Membrane forms (1+1)-sphere connected by ‘wormhole’
- Budding and neck formation ⇔ spontaneous curvature

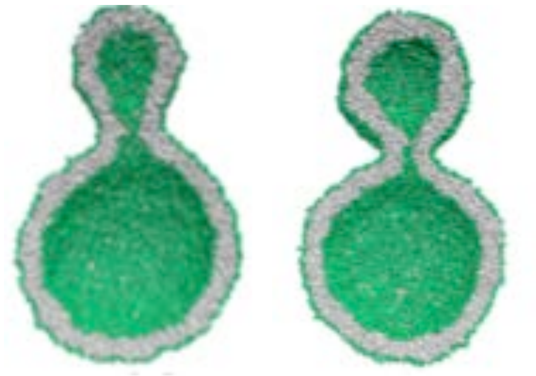
# Closed Neck of (1+1)-Sphere




- (1+1)-sphere for positive spont curv, $m > 0$
- Large and small sphere with radius $R_l$ and $R_s$
- Connected by closed membrane neck
- Neck curvature $M_{\text{ne}} = (1/2)\,(1/R_l + 1/R_s)$
- Closed neck is stable if $\boxed{0 < M_{\text{ne}} \leq m}$
- Local relation between geometry and spont curvature $m$ = material parameter

- Stability criterion obtained from curvature elasticity
- Theory of curvature elasticity ignores molecular details
- What about membrane necks on the molecular scale?

# Closer Look at Membrane Neck



- MD simulations: Hour-glass shape of neck
- Neck has waistline with radius $R_{\text{ne}}$
- Waistline has mean curvature

$$M_{\text{wl}} = (1/2)\,(C_1 + C_2)$$

with principal curvatures $C_1$ and $C_2 = 1/\,R_{\text{ne}}$

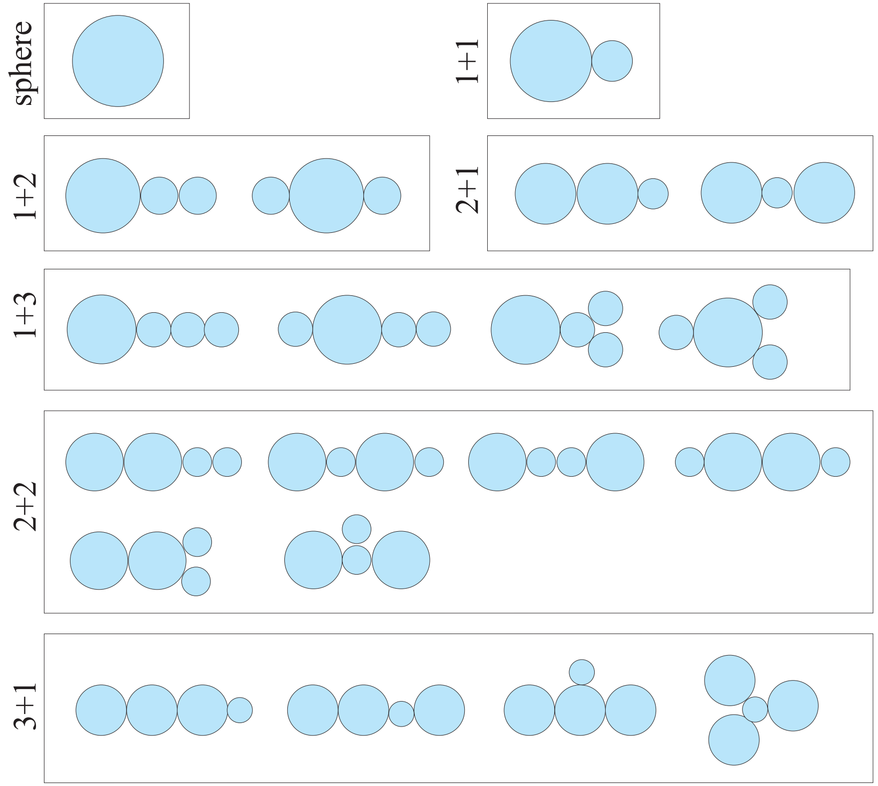
- Neck closure implies vanishing $R_{\text{ne}}$ and divergent $C_2$ !
- Divergence of $C_2$ cancelled by divergence of $C_1$
- Finite limit $M_{\text{wl}} \approx M_{\text{ne}} = (1/2)\,(1/R_l + 1/R_s)$

# Multispheres: Theory

RL, *Advances in Biomembranes and Lipid Selfassembly* Vol. 30 (2019) Ch. 3

- Single membrane forms several spheres, with pairs of neighboring spheres connected by membrane necks:

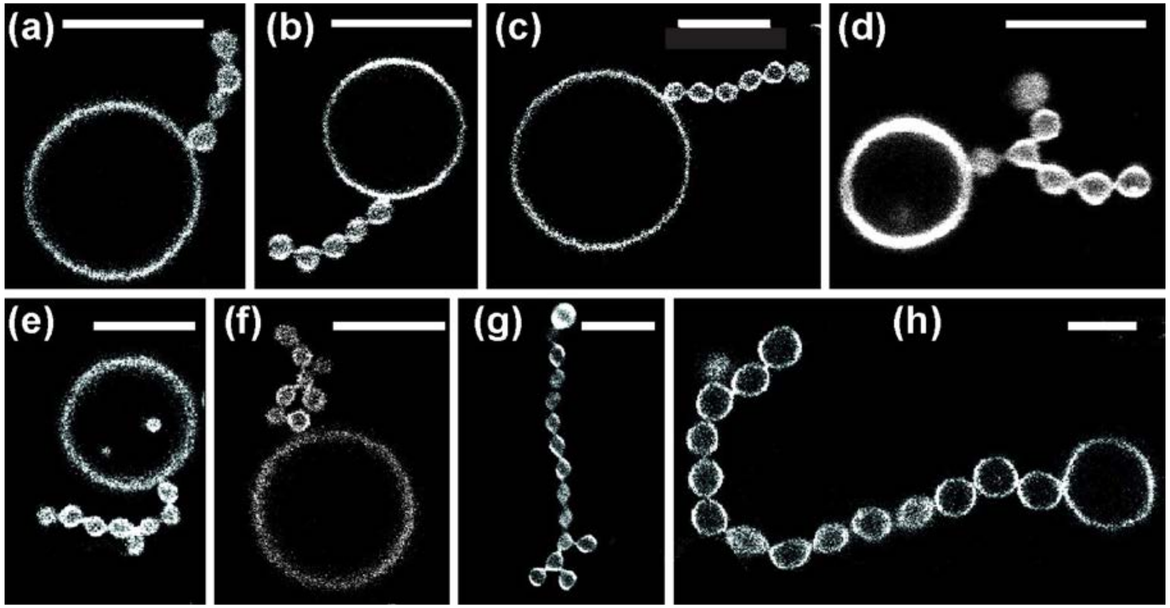
- Only two possible radii
- Large spheres with radius $R_l$
- Small spheres with radius $R_s$
- $(N_l + N_s)$-spheres
- Example: $N_l + N_s \leq 4$
- Overlapping stability regimes



# Multispheres: Experiment

- (1+$N_s$)-spheres, one large, $N_s$ small spheres:

Bhatia et al, *Soft Matter* (2020)



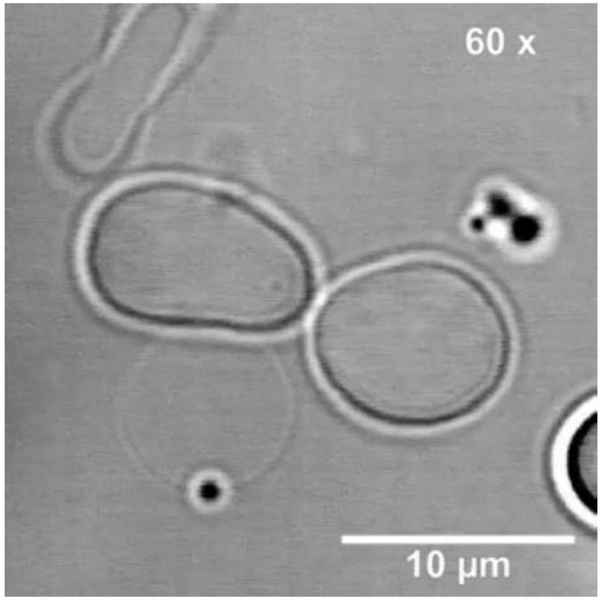

- Only two different radii, $R_l$ and $R_s$
- Each shape formed by single membrane
- $N_s$ membrane necks

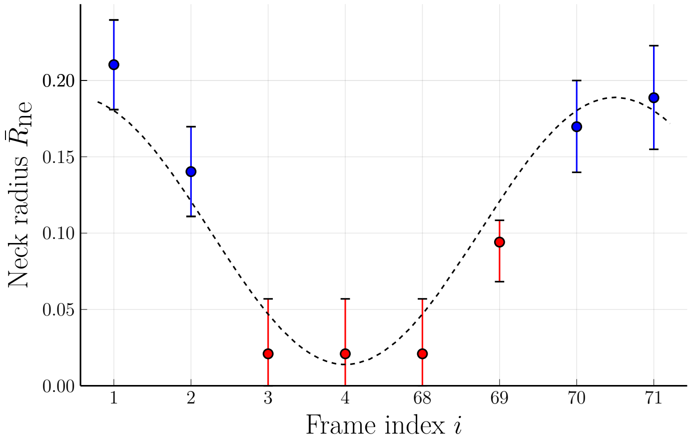
- In general: ($N_l$ + $N_s$)-spheres with $N_l + N_s - 1$ necks
- Surprising mobility: linear ⇔ branched chains
- Degenerate case: $N_*$ equally sized spheres

# Active Neck Oscillations of GUVs

Litschel et al, *Angewandte Chemie* (2018)

Christ, Litschel, Schwille, RL, *Soft Matter* (in press)



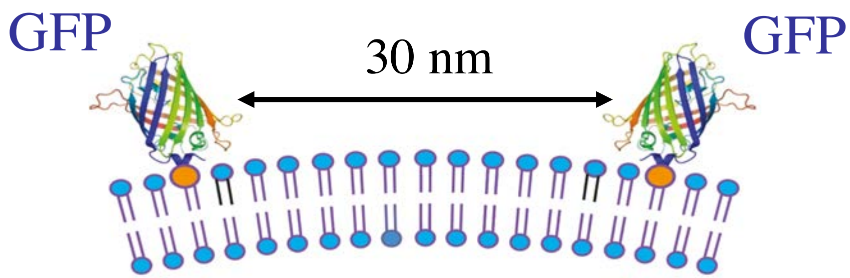




- Shape oscillations generated by Min protein system coupled to ATP
- Dumbbell shape with recurrent closure and reopening of neck
- 26 complete oscillations
- Two branches of dumbbells, symmetric and asymmetric ones
- Oscillations of bound Min proteins
- Oscillations of spont curvature
- Oscillations of neck radius

# Fine Tuning of GUV Morphologies

Steinkühler et al, *Nature Comm.* (2020)

- Binding of GFP to small mole fraction of anchor NTA-lipids:

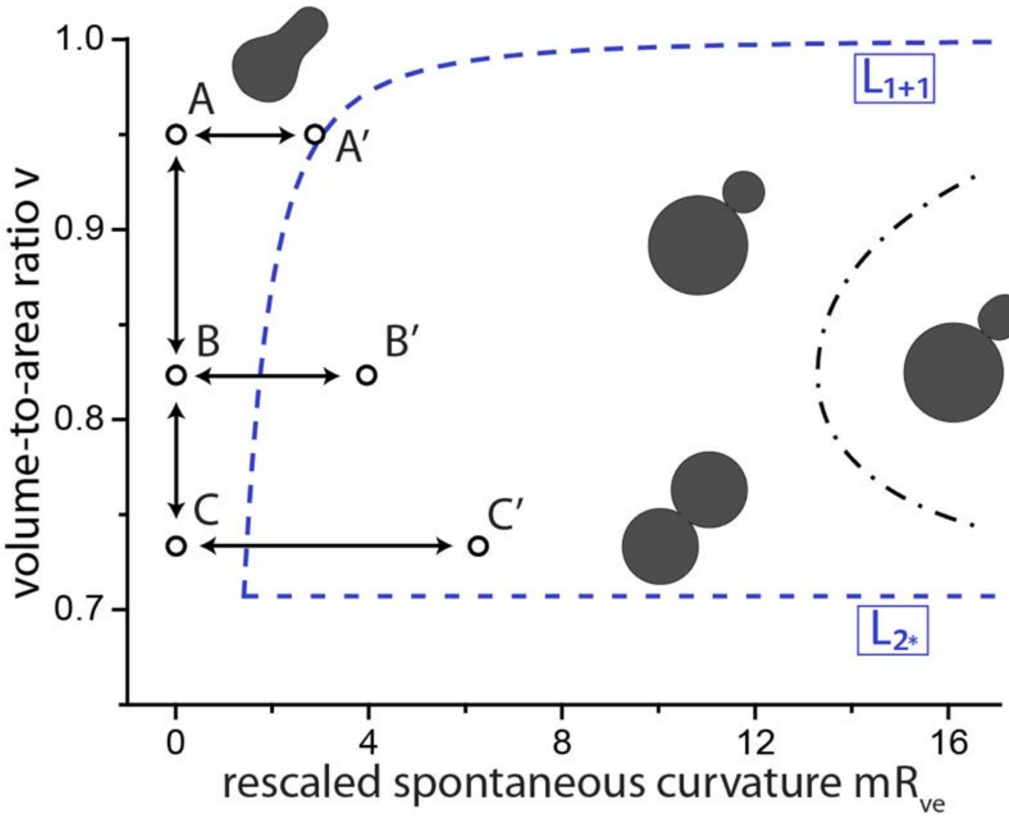



His-tagged GFP
NTA-lipids

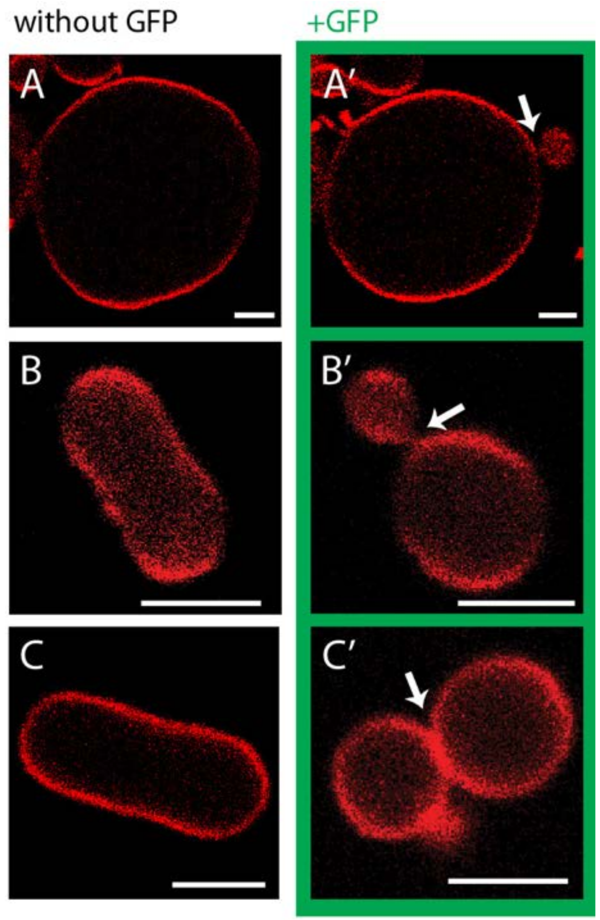
- Dilute regime, no crowding !
- Nanomolar GFP concentration $X$ as control parameter
- Density $\Gamma$ of bound GFP increases linearly with $X$
- Spont curvature $m$ increases linearly with $\Gamma \sim X$

# Controlled Budding of GUVs

- Morphology determined by volume and spont curvature (rescaled):




- Volume via osmotic conditions
  Sp-curvature via GFP concentration



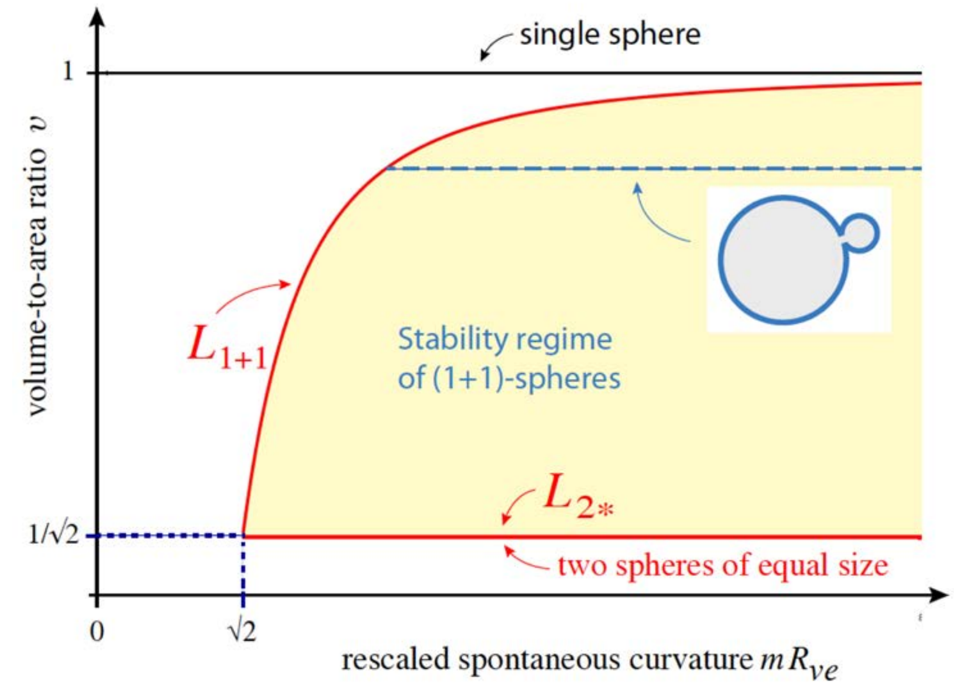

Steinkühler et al, *Nature Comm.* (2020)

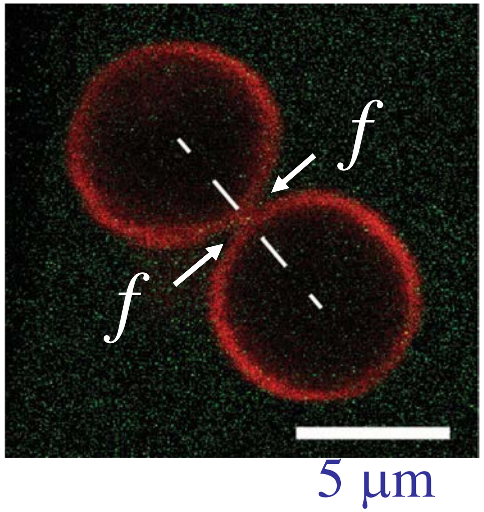
# Constriction Force from Spont Curvature

RL, *Advances in Biomembranes and Lipid Selfassembly* Vol. 30 (2019) Ch. 3

- Sp-curvature $m$ generates constriction force $f$ acting radially on membrane neck:

$$f = 8\pi\,\kappa\,(\,m - M_{ne}\,)$$



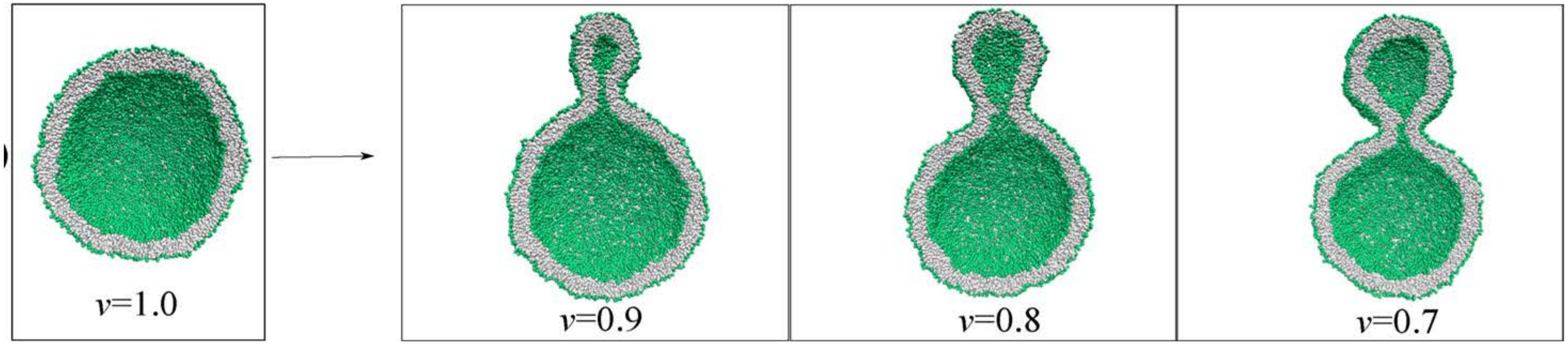




- Increase of $m$ for fixed $v$
- Fixed shape of (1+1)-sphere
- Constriction force $f$ increases

# Controlled Budding of Nanovesicles

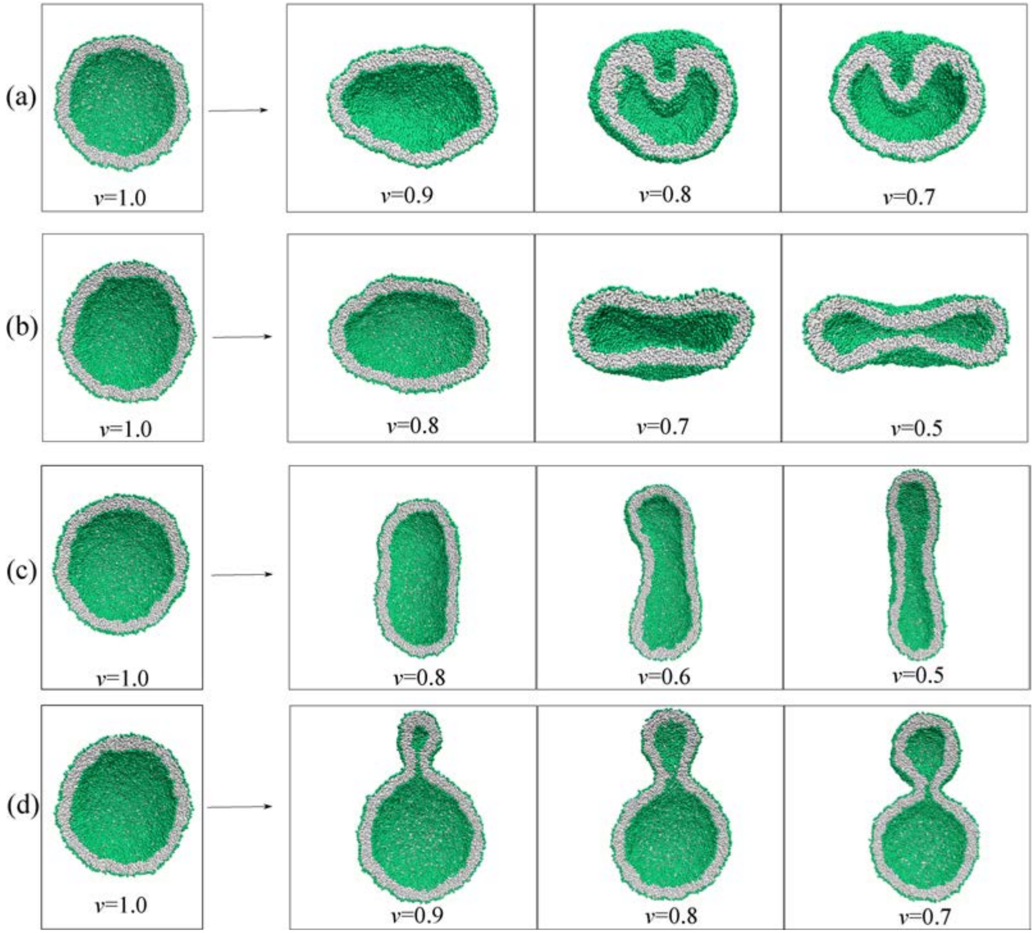
Ghosh, Satarifard et al: *Nano Letters* (2019)

- Spherical nanovesicle with diameter of 36 nm
- Assembly of lipids into inner and outer leaflet
- Controlled number of inner and outer lipids, $N_{il}$ and $N_{ol}$
- Decreasing vesicle volume $v$, corresponding to deflation
- Formation of dumbbell with closed neck for



# Polymorphism of Nanovesicles

Ghosh, Satarifard et al,
*Nano Letters* (2019)




- Four spherical vesicles with diameter 36 nm
- Same volume
- Same total # of lipids
- Different inner and outer lipids, $N_{il}$ and $N_{ol}$
- Reduction of volume: very different shapes

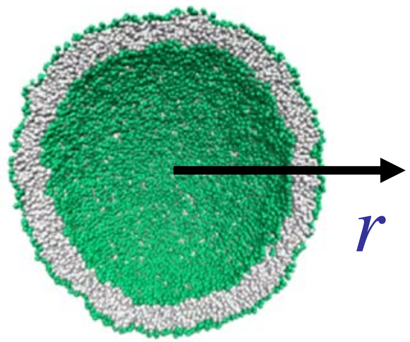
# Fine Tuning of SUV Morphologies

Ghosh, Satarifard et al, *Nano Letters* (2019)

- Two leaflets with different lipid numbers
- Tensionless bilayer:

  One leaflet stretched, the other leaflet compressed

- Spherical vesicle with radial coordinate $r$
- Spont curvature $m$ from stress profile $s(r)$ across bilayer:

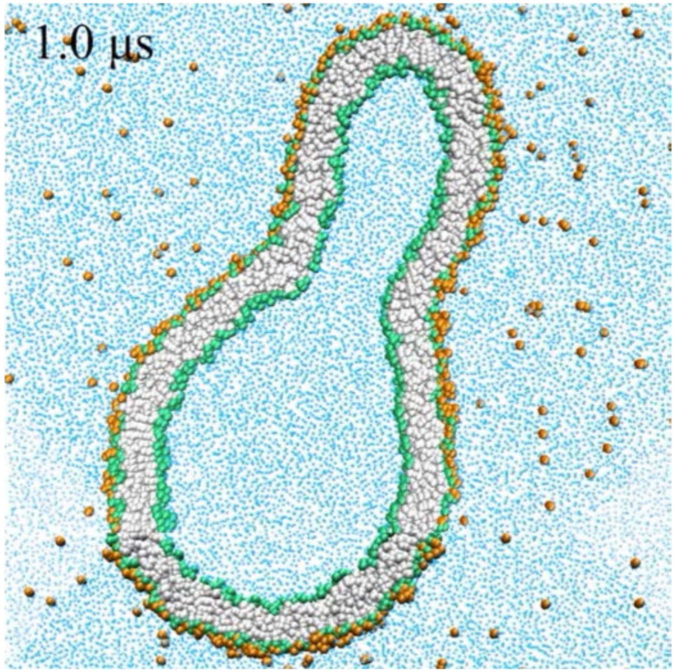



$$2\kappa\left(\frac{1}{R_{\text{mid}}} - m\right) = \int_0^\infty \mathrm{d}r\ s(r)r$$

# Shape Oscillations of Nanovesicles

Ghosh, Satarifard, Grafmüller, RL (submitted)

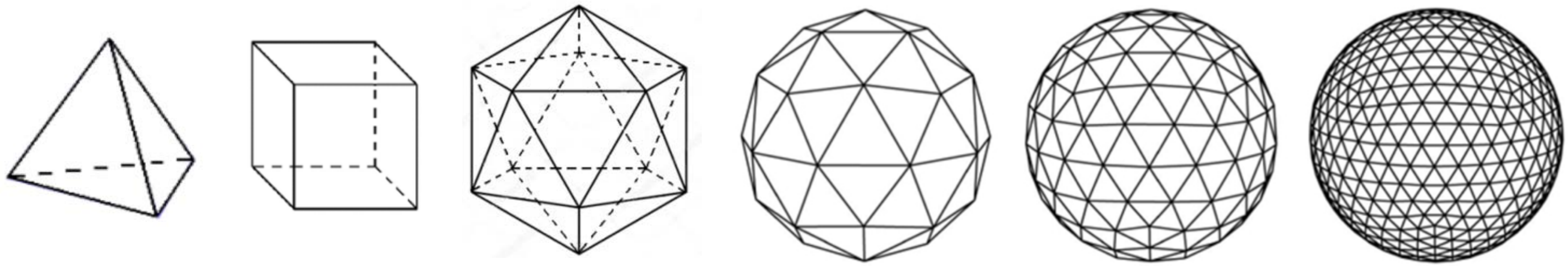
- Nanovesicle exposed to small solutes (orange) that adsorb onto vesicle membrane:

- Shape of Membranes
- Giant Vesicles and Nanovesicles
- Morph Complexity ~ Membrane Necks
- Topological Transformations
- Fission of Membrane Necks

# Topology of Surfaces

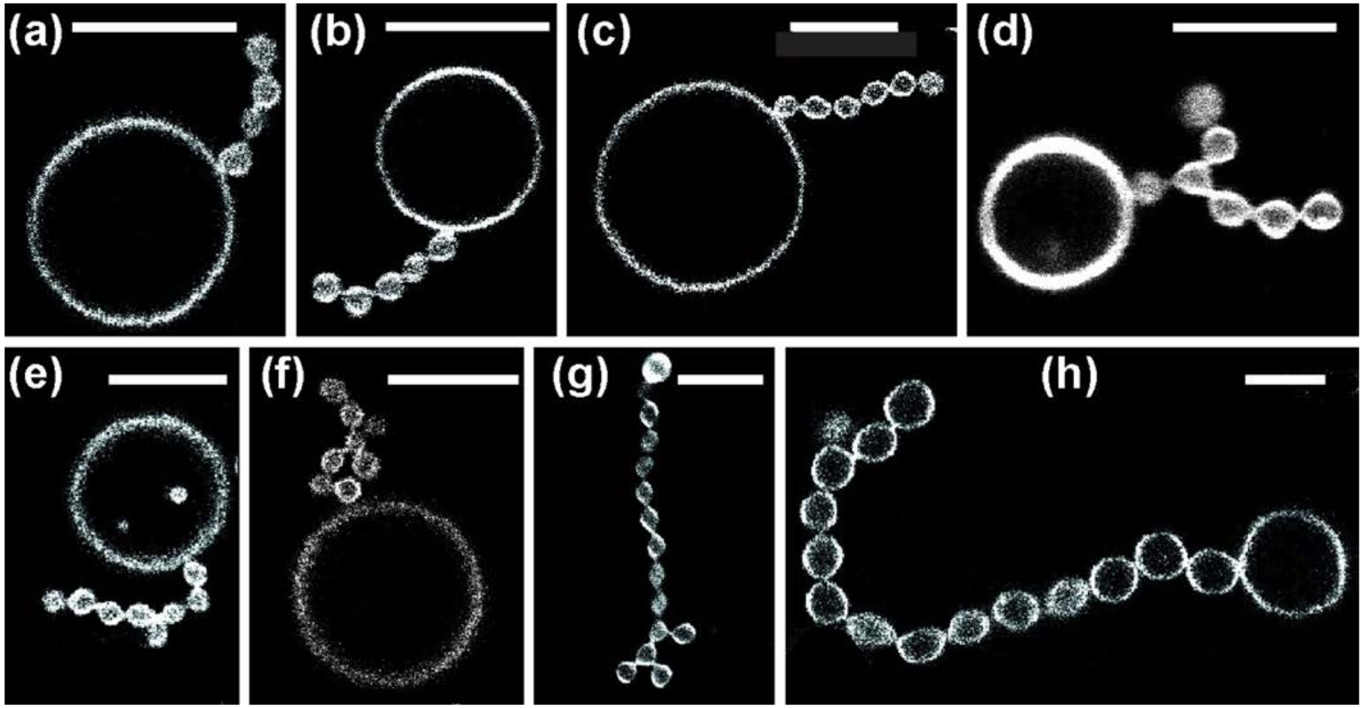
- Closed surface with $F$ faces, $E$ edges, and $V$ vertices




- Euler characteristic $\chi = F - E + V$
- For tetrahedron, cube, ..., and sphere: $\chi = 2$
- Euler characteristic is topological invariant
- Euler characteristic is additive: $\chi = 2 + 2 = 4$ for two spheres

# Topology of Multispheres

- All multispheres have the same topology as a single  sphere !

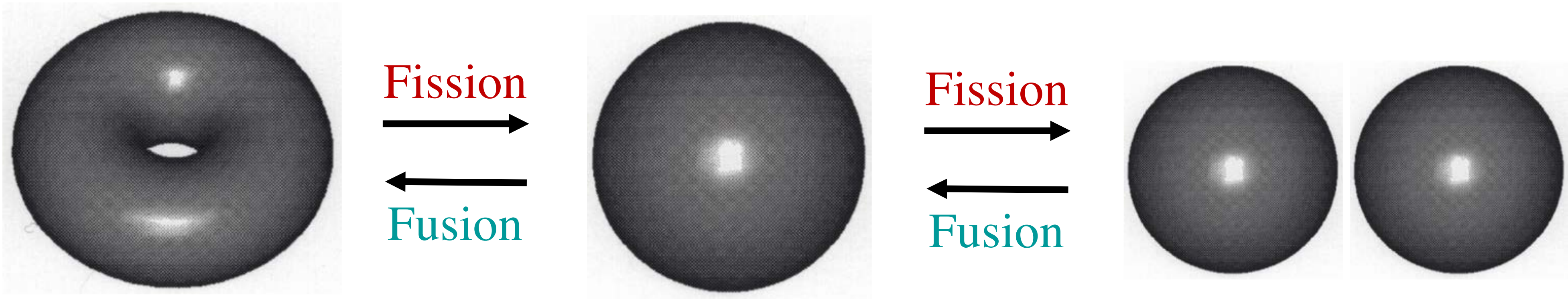



All multispheres have the same Euler characteristic

$$\chi = 2$$

# Topological Transformations

- Topological classification via Euler characteristic $\chi$ :

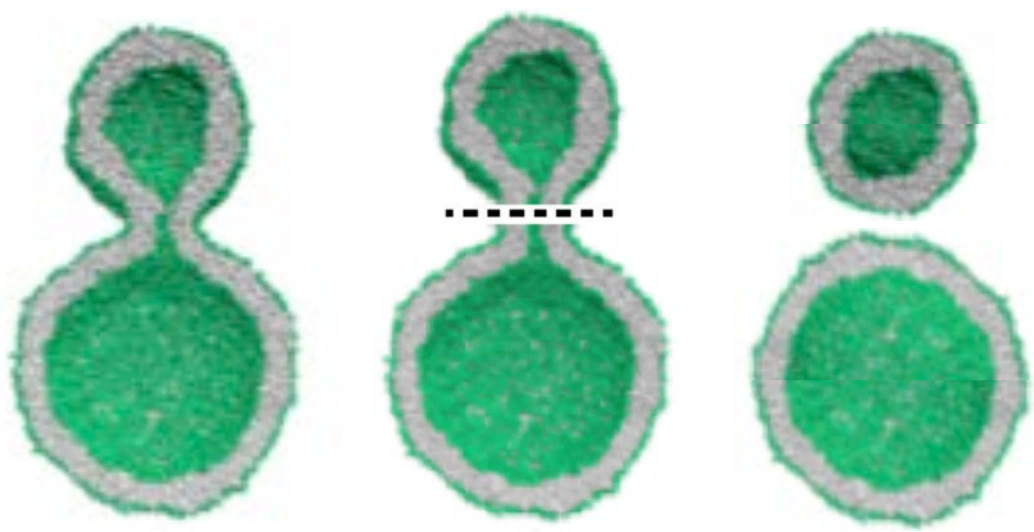



$\chi = 0$ $\chi = 2$ $\chi = 4$

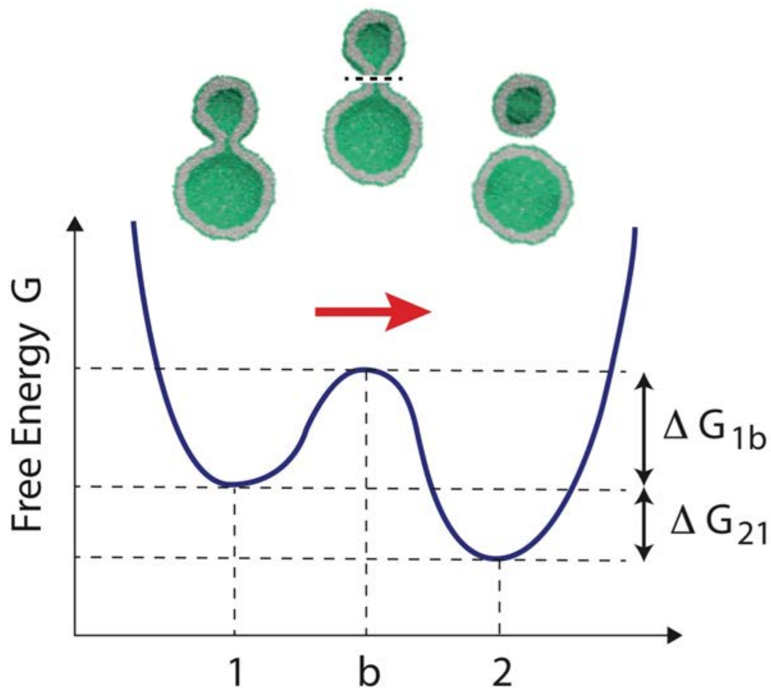
- Topological transformation $\Leftrightarrow$ change $\Delta\chi = \chi_{fin} - \chi_{ini}$
- Fission: Euler characteristic $\Delta\chi > 0$
- Fusion: Euler characteristic $\Delta\chi < 0$

# Fission of Membrane Necks

- Membrane fission implies disrupture/cut of membrane
- Work of fission proportional to length of cut
- Shortest possible cut for dumbbell across membrane neck:



Dumbbell    Broken neck    Two spheres

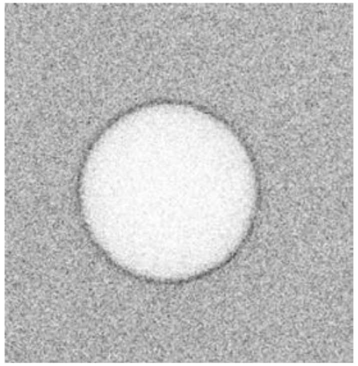


# Neck Fission of GUVs

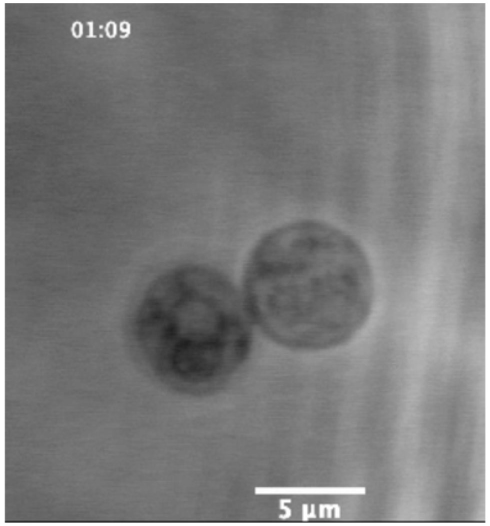
Steinkühler et al:  *Nature Comm*. (2020)

- Osmotic deflation + GFP binding
- Osmotic deflation: Spherical GUV -> dumbbell GUV

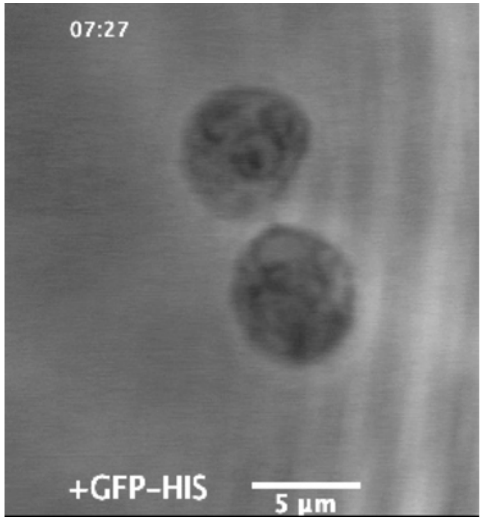
  Increase in GFP  -> Neck cleavage -> Two daughter GUVs



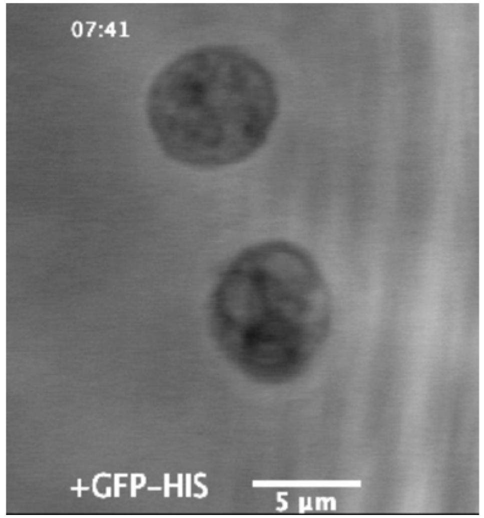
Adsorption of GFP onto GUV membrane




Deflation leads to dumbbell with membrane neck



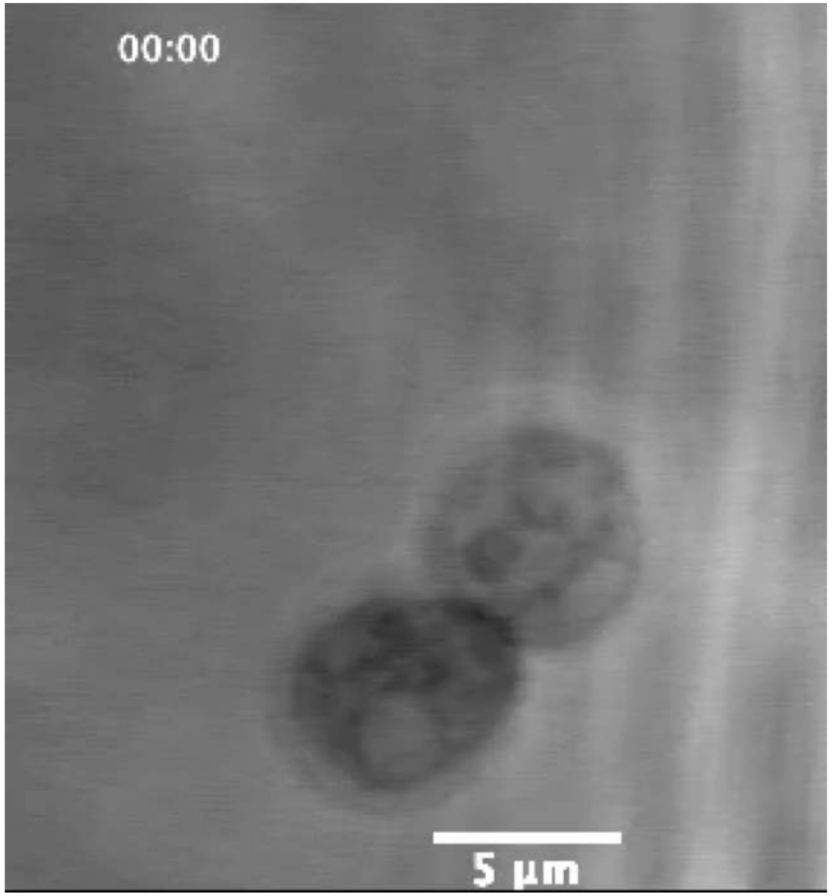

Directly after neck cleavage




Complete division into two smaller GUVs

# Neck Fission of GUVs: Movie

Steinkühler et al: *Nature Comm*. (2020)

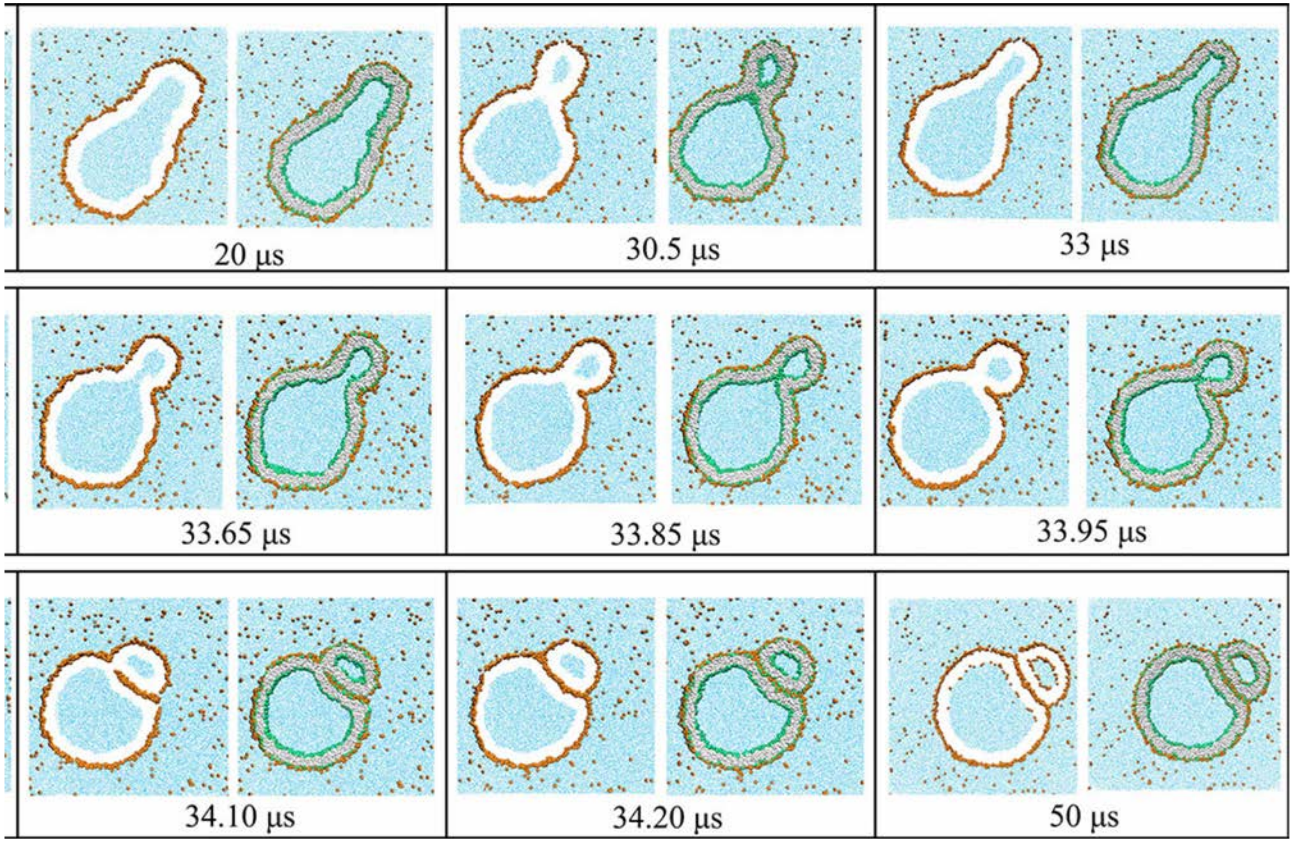


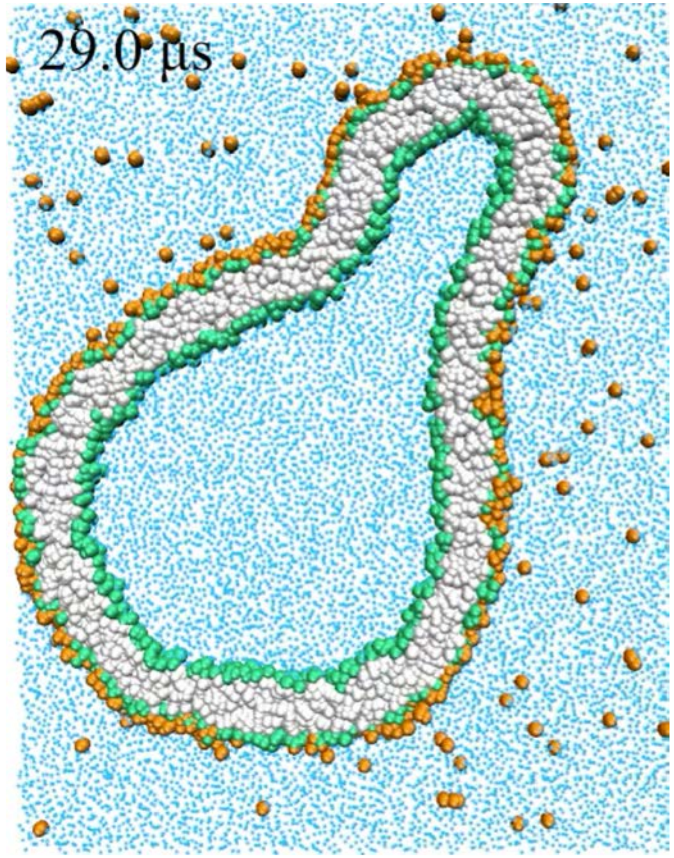

- Two-step process:
- Osmotic deflation:
  Spherical GUV -> dumbbell GUV
- Increase in GFP ->
  Neck cleavage + GUV division

# Neck Fission of Nanovesicles

Ghosh, Satarifard, Grafmüller, RL (submitted)

- Nanovesicle exposed to small solutes (orange) that adsorb onto vesicle membrane:



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  Nano Letters (2019) 19, 7703-7711
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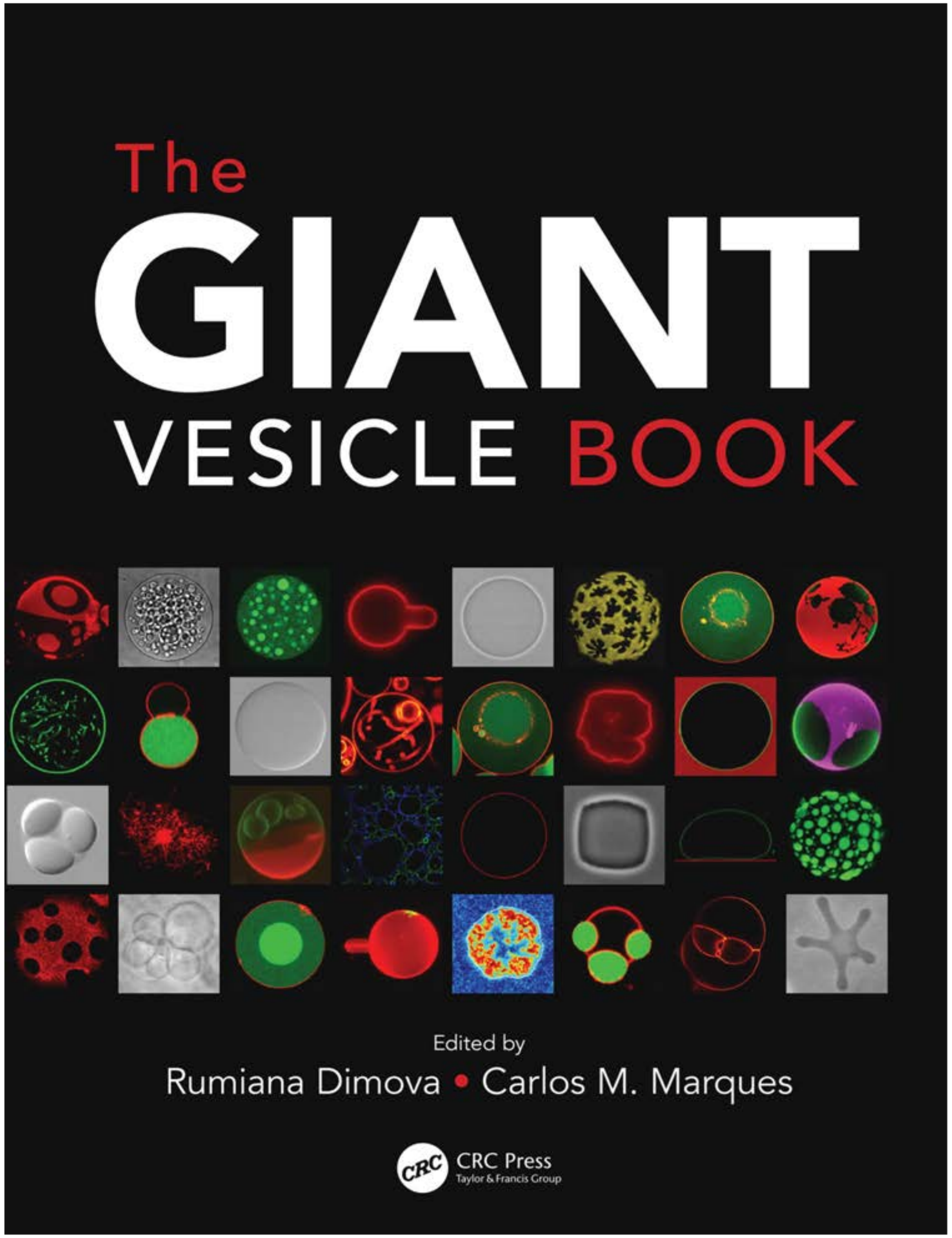
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CRC Press
Taylor & Francis Group

# Coworkers

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Titus Franzmann