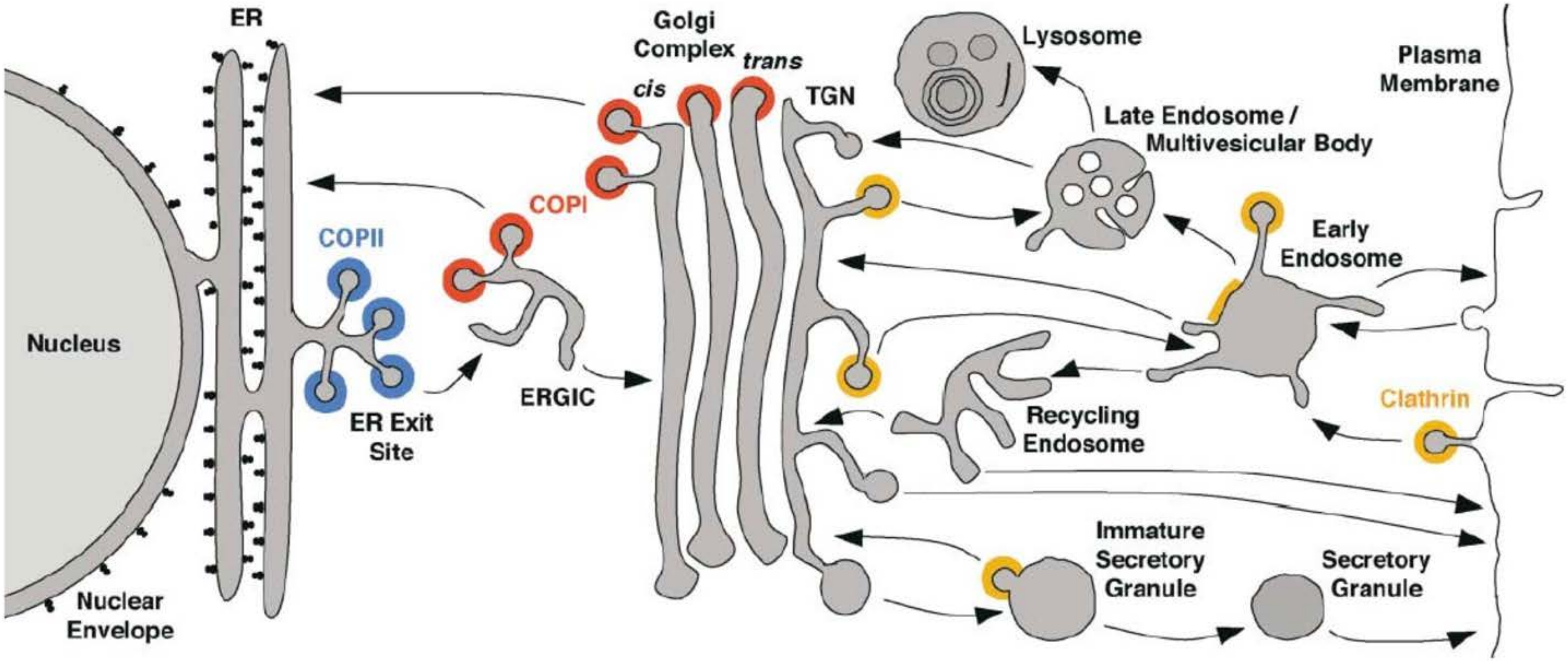
Remodeling Shape and Topology of Fluid Membranes by Curvature and Tension

Reinhard Lipowsky
MPI of Colloids and Interfaces, Potsdam, Germany

- Membrane Compartments
- Remodeling of Membrane Shape
- Membrane Necks and Multispheres
- Remodeling of Membrane Topology
- Fission and fusion processes

Intracellular Vesicle Trafficking



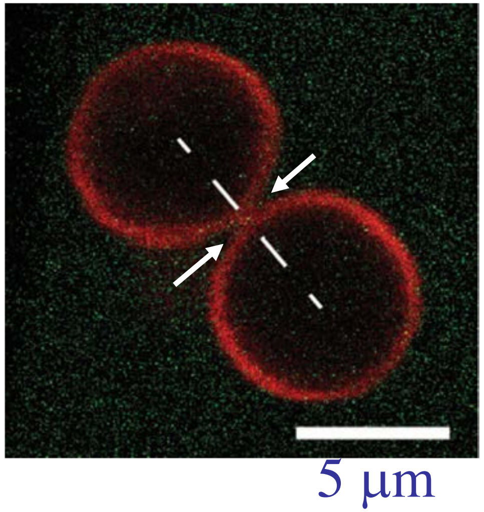
Bonifacino,Glick, *Cell* (2004)

- Colored dots: vesicle formation by budding and fission
- Arrowheads: vesicle uptake by adhesion and fusion

Synthetic Membrane Compartments

Steinkühler et al, *Nature Comm* (2020)

- Giant vesicles or GUVs
- Remodeling observed by optical microscopy



- Nanovesicles or SUVs
- Electron microscopy: limited to a single snapshot for each individual nanovesicle
- Remodeling studied via Molecular Dynamics simulations

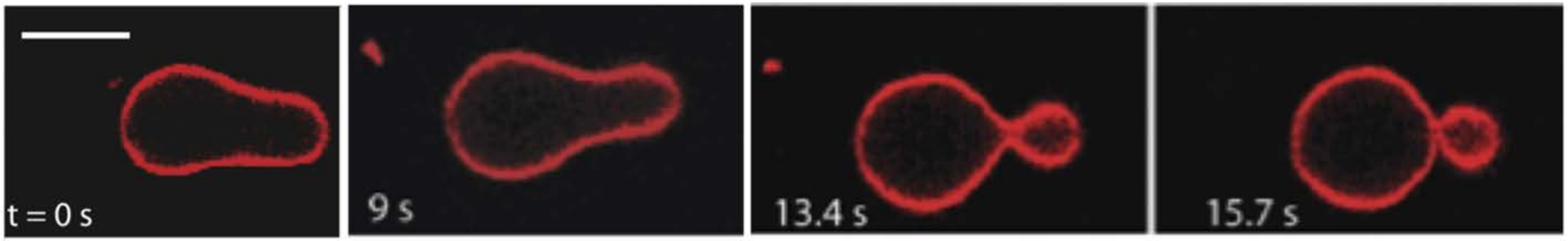
Ghosh, Satarifard et al, *Nano Letters* (2019)

- Remodeling of membrane shape
    - Examples for GUVs and nanovesicles
    - Membrane elasticity
    - Membrane necks and multispheres
- Remodeling of membrane topology
    - Relation to fission and fusion processes
    - Fission of membrane necks

Budding of Giant Vesicles

- Pear-like vesicle transformed into two-sphere vesicle
- Snapshots from time lapse over 16 s:

Bhatia et al, *Soft Matter* (2020)



Scale bar: 5 µm

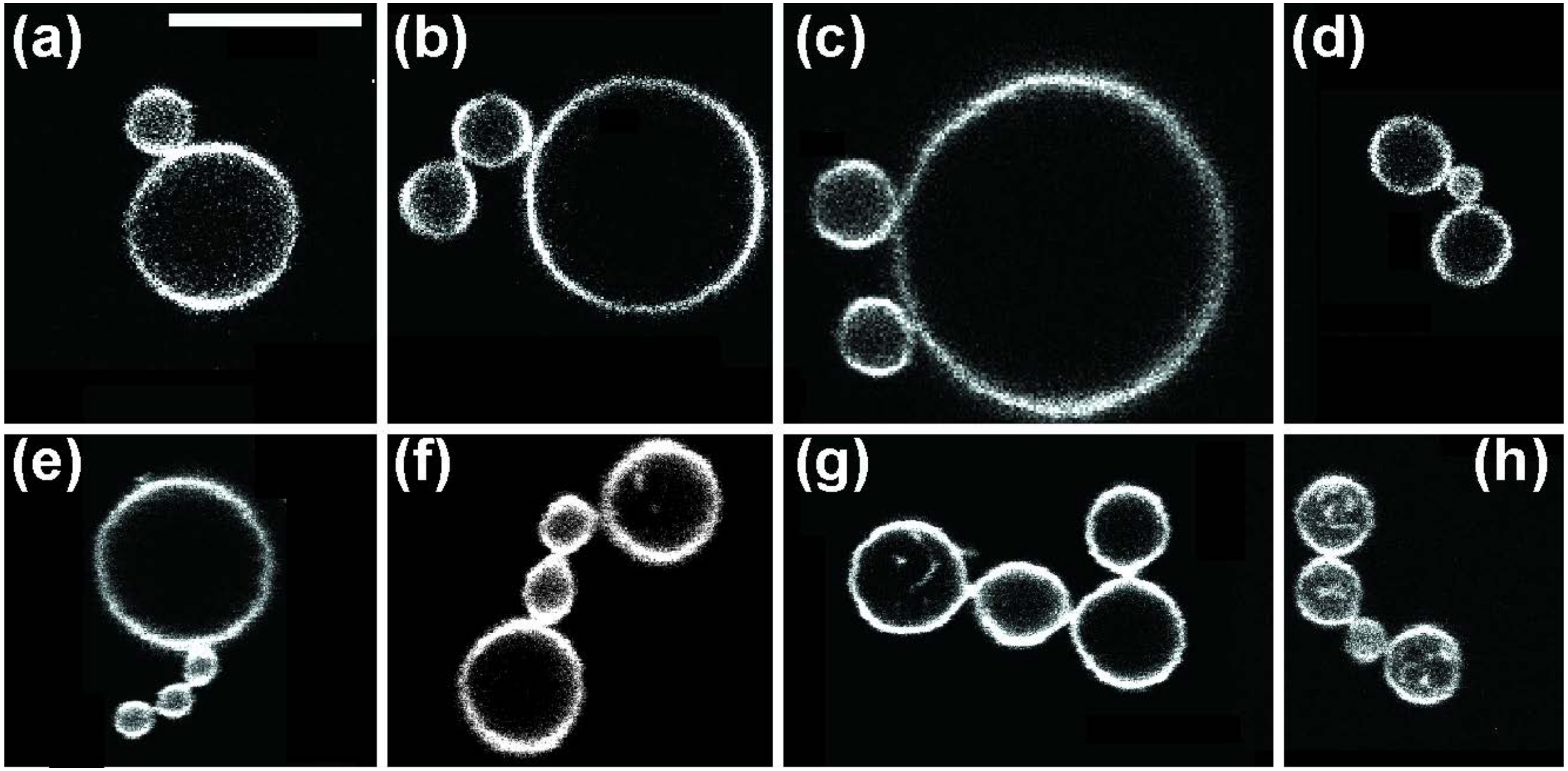
'Fluid worm hole in three dimensions'

- Membrane exposed to asymmetric sucrose/glucose solutions
- Membrane forms two spheres connected by a single neck
- Same membrane system leads to proliferation of necks !

Multispheres with Many Necks

Scale bar: 10 μm

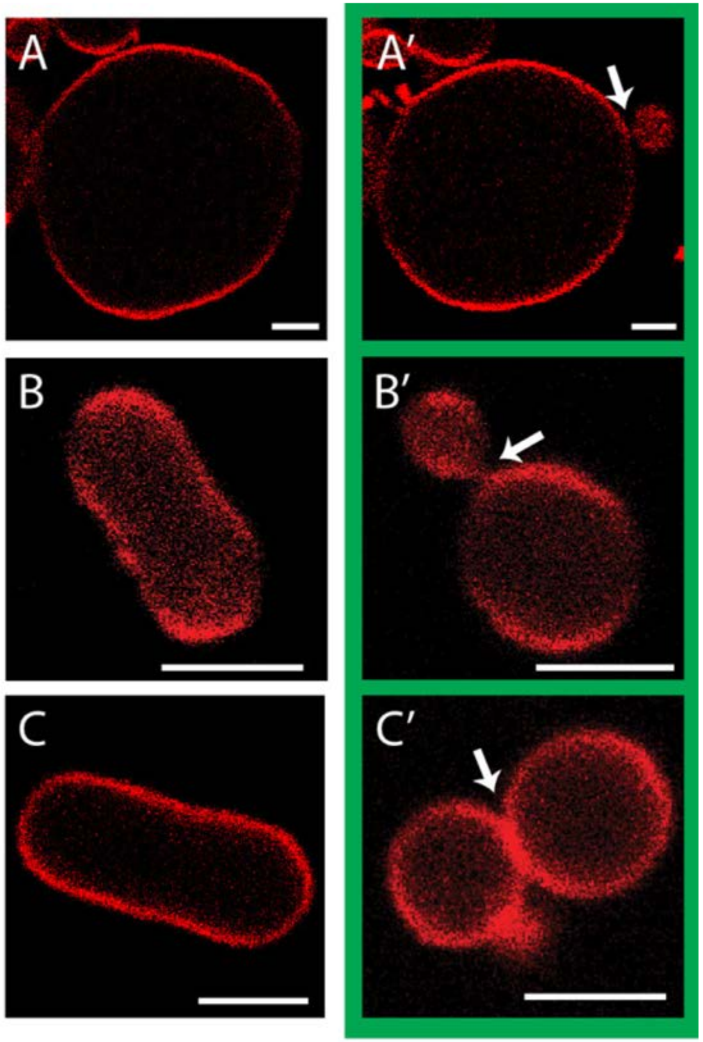
Bhatia et al, *Soft Matter* (2020)



- One membrane forms several spheres connected by necks
- Each shape involves only two different sphere radii

Controlled Budding of GUVs

Steinkühler et al, *Nature Comm* (2020)



No GFP + GFP

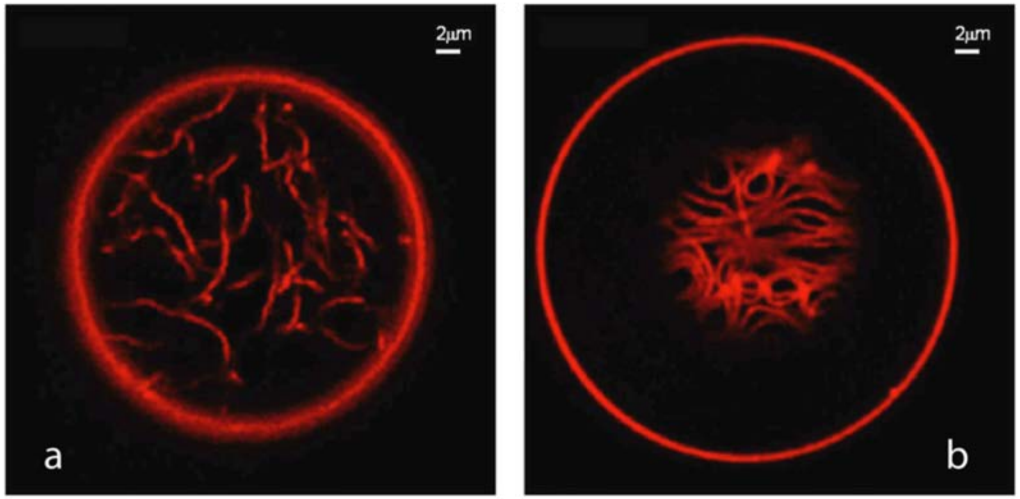
- Vesicles exposed to His-tagged GFP in the exterior solution
- GFP binds to anchor lipids in the vesicle membrane
- Membrane curvature fine-tuned by nanomolar concentration of GFP
- Low densities of membrane-bound GFP generate strongly curved membranes

GUVs and Aqueous Phase Seperation

Li et al, *PNAS* (2011) Liu et al, *ACS Nano* (2016)

- Aqueous phase separation within giant vesicles
- Example: PEG and dextran in water
- Formation of many stable nanotubes, no pulling forces
- Wetting properties determine patterns of nanotubes:

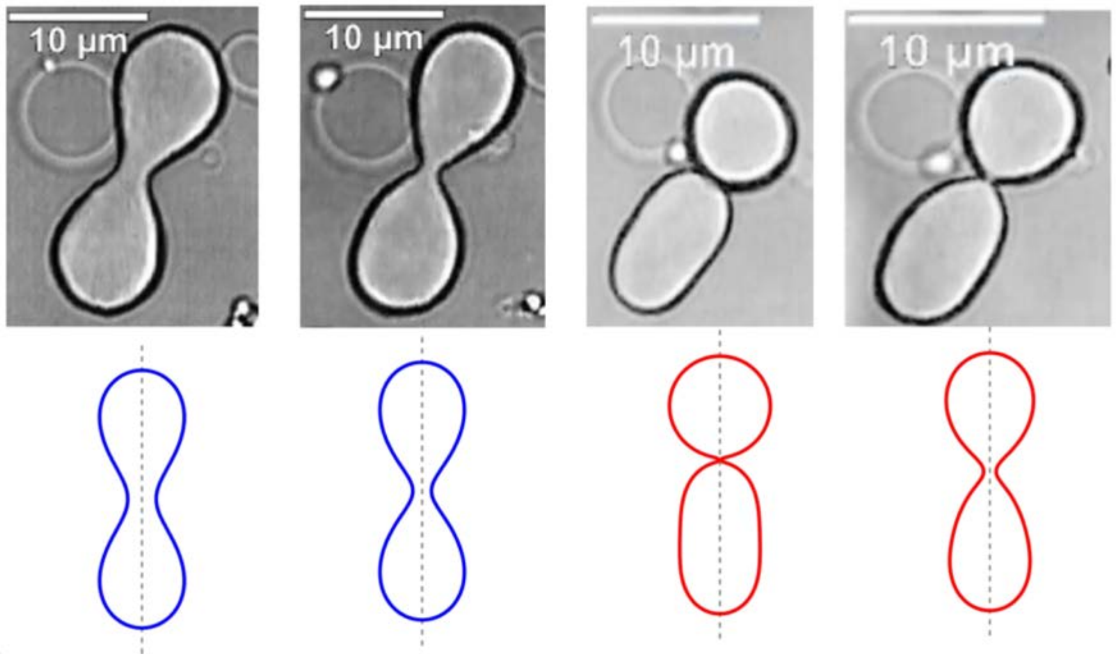
Complete wetting: tubes stay in one liquid phase



Partial wetting: tubes bind to liquid-liquid interface

Active Shape Oscillations of GUVs

- Min proteins D and E in interior solution
- MinD/E binds to membrane and unbinds via ATP hydrolysis



Litschel et al, *Angewandte Chemie* (2018)

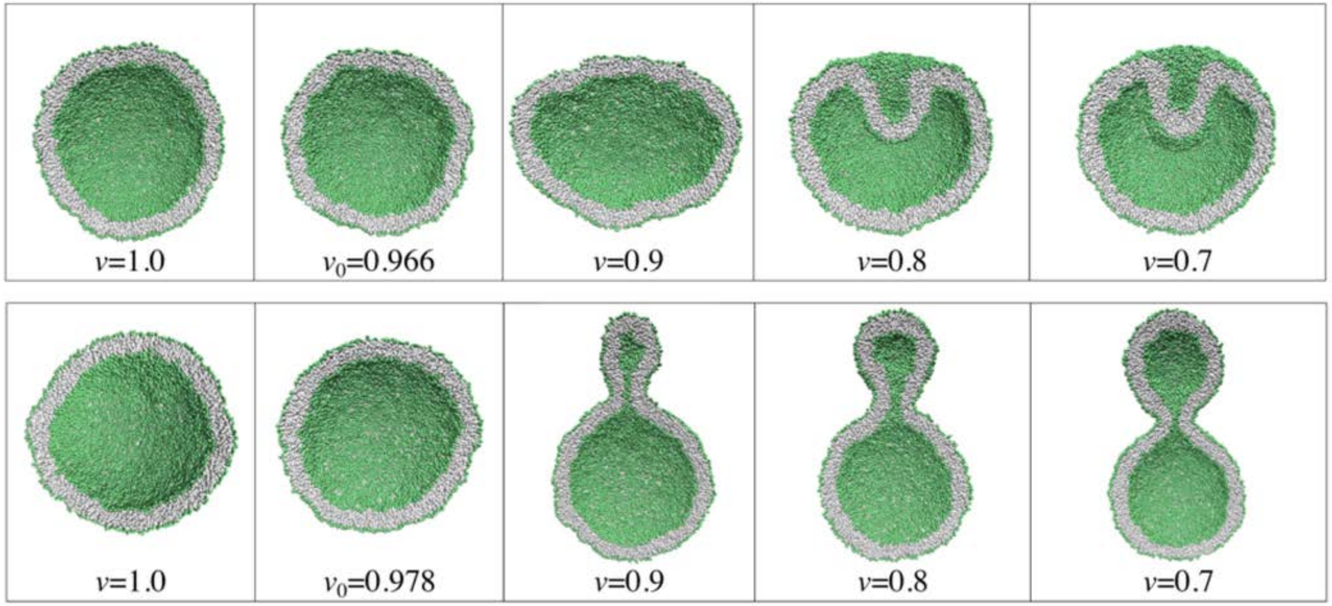
Christ et al, *Soft Matter* (2021)

- Cyclic closure and opening of membrane neck
- Time-dependent spontaneous curvature

Remodeling of Nanovesicle Shape

- Spherical nanovesicles with diameter of 36 nm
- Shape transformations by volume reduction

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



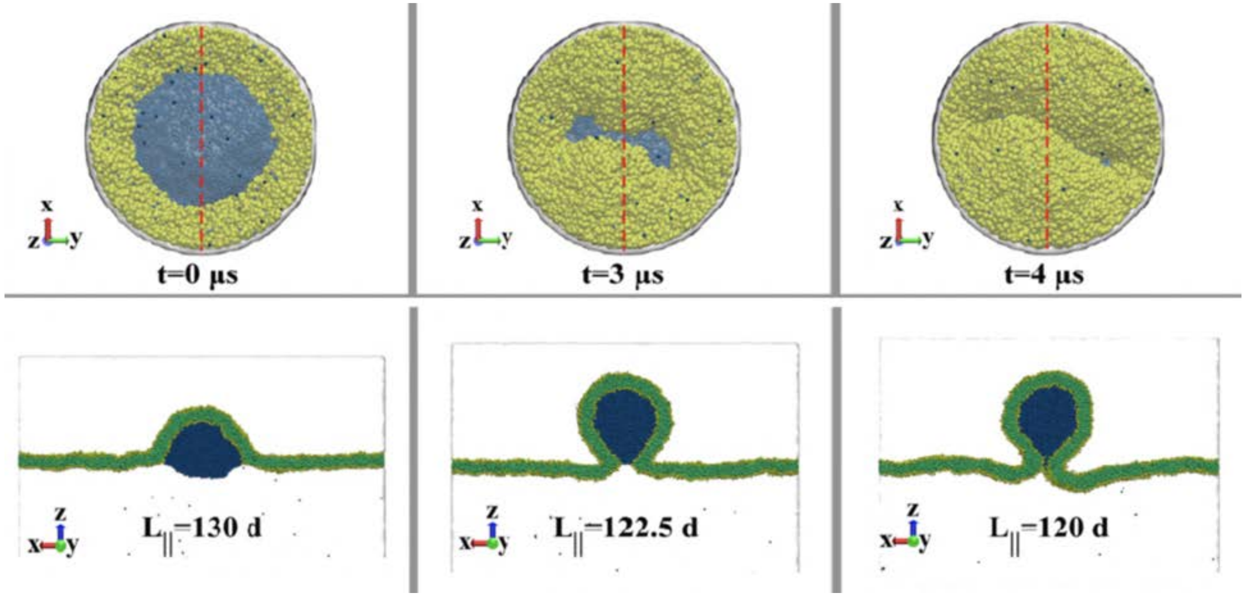
- Inner leaflet compressed
- Outer leaflet compressed

- Controlled by mechanical tensions in the two leaflets

Non-axisymmetric Membrane Necks

Satarifard et al, ACS Nano (2018)

- Engulfment of nanodroplet (blue) by lipid bilayer
- Neck shape controlled by mechanical bilayer tension:



Bottom view

Side view

- Formation of tight-lipped neck reveals negative line tension of contact line

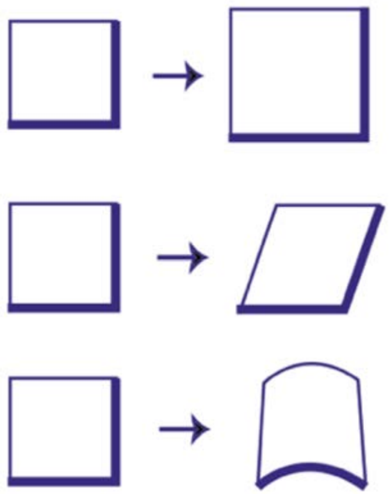
Elasticity of Fluid Membranes

- Biomembrane as thin elastic sheet
- Elastic Deformations

Stretching

Shearing

Bending



- Fluid Membranes

Membrane tension

Shear -> Flow

Curvature elasticity

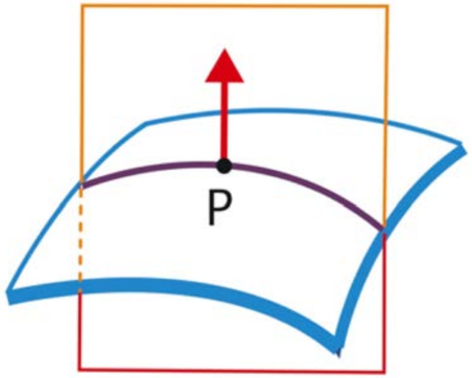
Theory of Membrane Elasticity

- Elastic stretching: Area A and tension Σ

  Mechanical tension $\Sigma = K_A\,(A - A_0)/A_0$

  area compressibility modulus K_A , optimal area A_0

  Stretch energy $E_{\mathrm{st}} = \frac{1}{2} K_A\,(A - A_0)^2/A_0$



- Elastic bending: mean curvature M

  Bending energy $E_{\mathrm{be}} = \int dA\; 2\,\kappa\,(M - m)^2$

  bending rigidity κ , spontaneous curvature m

- Total elastic energy: Stretch energy + bending energy

$$E_{\mathrm{el}} = E_{\mathrm{st}} + E_{\mathrm{be}} = \frac{1}{2} K_A\,(A - A_0)^2/A_0 + E_{\mathrm{be}}$$

Composite Nature of Membrane Tension

- Mechanical tension Σ plus spontaneous tension $\sigma = 2\,\kappa\, m^2$
- Spontaneous tension leads to spontaneous tubulation
- Total membrane tension $\Sigma_{tot} = \Sigma + \sigma$

Lipowsky, *Faraday Disc* (2013)

- Spontaneous tension $\sigma = 2\,\kappa\, m^2$ is material parameter
- Tension σ measured by micropipette aspiration of tubulated GUVs

Bhatia et al, *ACS Nano* (2018)

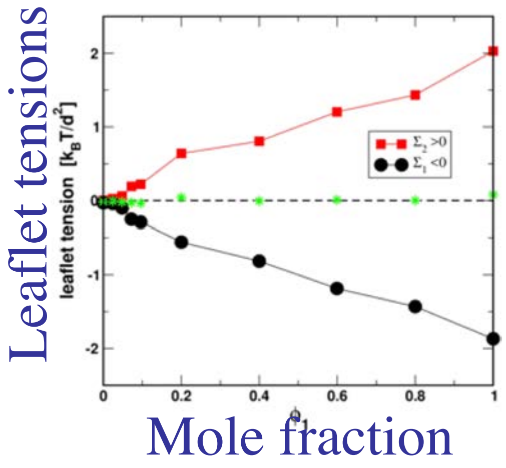
- Mechanical tension Σ depends on vesicle size and shape
- Difficult to measure, exact computation for multispheres

Bilayer and Leaflet Tensions

- Bilayer membrane consists of two leaflets, $l1$ and $l2$
- Mechanical bilayer tension $\Sigma = \Sigma_{l1} + \Sigma_{l2}$
- Tensionless bilayers: $\Sigma = 0$ implies $\Sigma_{l2} = - \Sigma_{l1}$

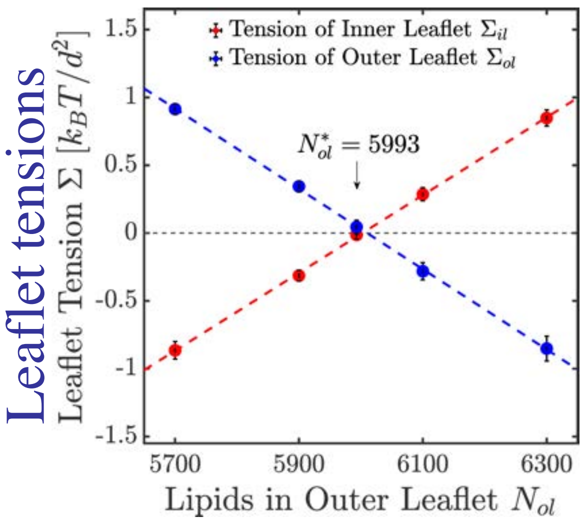
  => One leaflet stretched and one leaflet compressed
- Unique reference state with $\Sigma_{l2} = \Sigma_{l1} = 0$

Leaflet Tensions without Flip-Flops



Sreekumari, RL, *JCP*(2018)

- Planar bilayer, two lipids A and B
- Lower leaflet contains only lipid B
- Leaflet tensions versus mole fraction of lipid A in upper leaflet



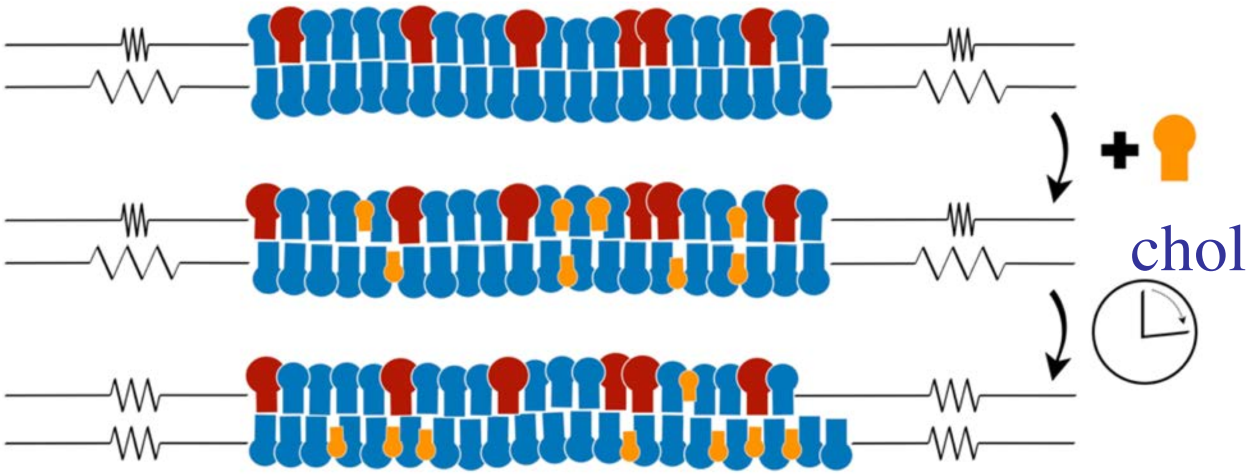
Ghosh, Satarifard et al, *Nano Letters* (2019)

- Spherical nanovesicle, one lipid A
- Fixed total number of lipids $N = N_{\mathrm{ol}} + N_{\mathrm{il}}$
- Leaflet tensions versus lipid number N_{ol} of outler leaflet

Leaflet Tensions with Flip-Flops

Miettinen, RL, *Nano Letters* (2019)

- Tensionless planar bilayer with two lipids (blue and red)
- Addition of cholesterol (orange) to both leaflets:



- Cholesterol undergoes flip-flops between leaflet
- Leaflet tensions decay to zero

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  - Examples for GUVs and nanovesicles
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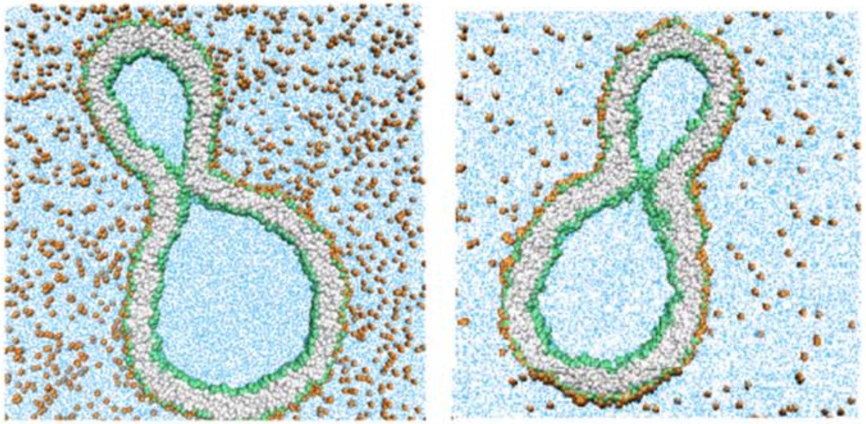
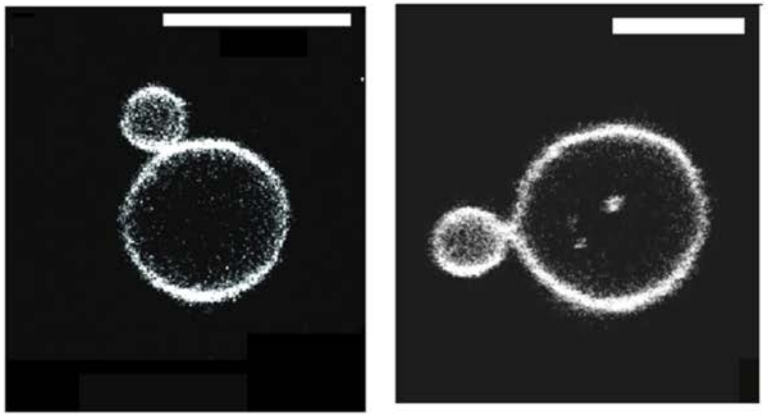
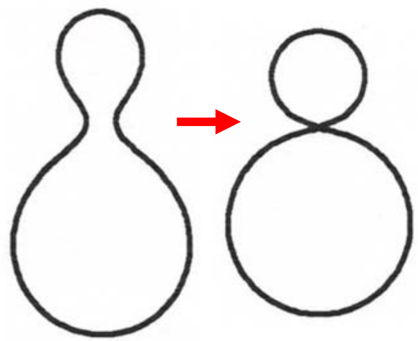
Separation of Length Scales

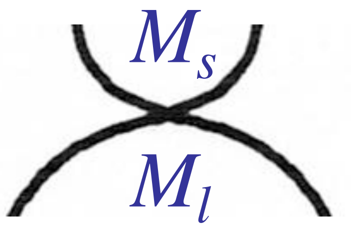



Nanoscale: Hourglass-shaped neck Micron scale: Pointlike neck

- Bilayer thickness $\ll$ vesicle size $R_{ve} = \sqrt{A/(4\pi)}$
- Simple relations for local properties of necks
- Neck closure: principal curvatures of waistline diverge but mean curvature M_{wl} remains finite

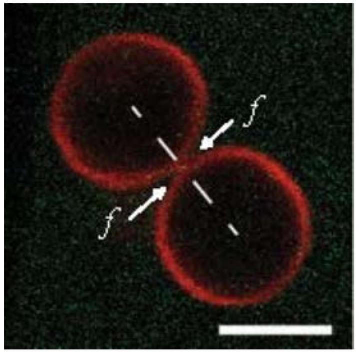


Local Properties of Membrane Necks



- Limiting value: $M_{\text{wl}} \approx \frac{1}{2}(M_l + M_s) \equiv M_{\text{ne}}$
- Defines neck geometry via neck curvature M_{ne}
- Stability of closed neck depends on spontaneous curvature m :

  neck is stably closed for $M_{\text{ne}} \leq m$



- Constriction force at neck, $f = 8\pi\,\kappa\,(\,m - M_{\text{ne}}\,)$
- Total membrane tension $\Sigma_{\text{tot}} = 4\,\kappa\,m\,M_{\text{ne}}$
- Mechanical membrane tension $\Sigma = 2\,\kappa\,m\,(2M_{\text{ne}} - m\,)$
- Relations between local neck geometry and elastic parameters

Multispheres: Geometry

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

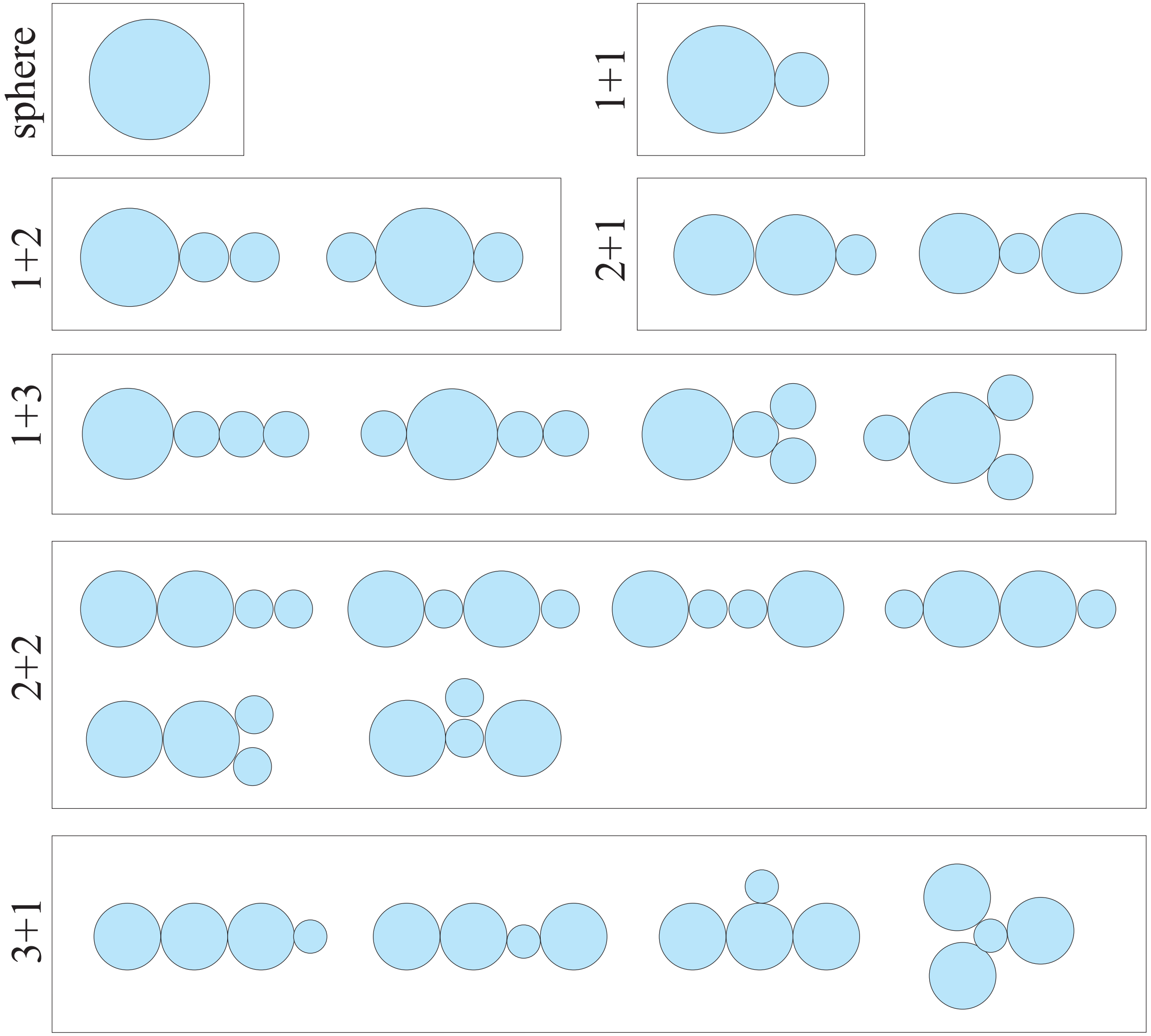
- Multispheres with large and small spheres
- Rescaled large sphere radius r_l and small sphere radius r_s
- Multispheres consisting of N_l large and N_s small spheres
- $(N_l + N_s)$-geometry determined by two simple equations:

$$N_l \, r_l^{\,2} + N_s \, r_s^{\,2} = 1$$

$$N_l \, r_l^{\,3} + N_s \, r_s^{\,3} = v$$

- Two nonlinear equations for two unknowns r_l and r_s
- Simple equations generate morphological complexity

Multispheres up to $N_l + N_s \leq 4$



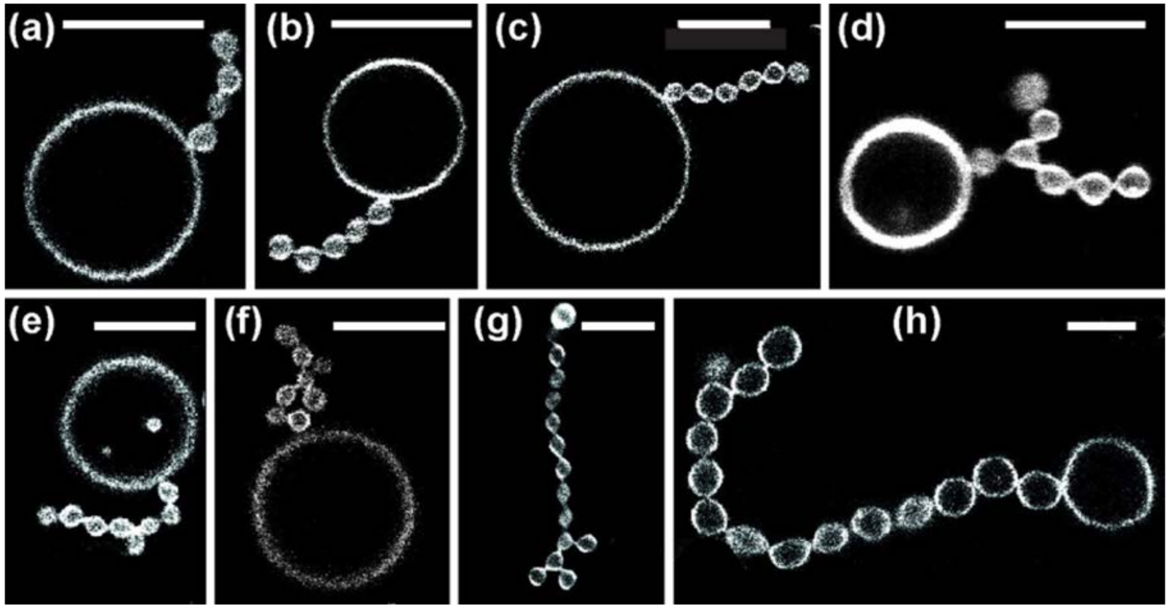
$(1+N_s)$-Multispherical Vesicles

Bhatia et al, Soft Matter (2020)

- $(1+N_s)$-spheres with one large sphere and a chain of N_s small spheres:

(1+4) (1+5) (1+6) (1+7)

linear chains



branched chain

branched chains

(1+8) (1+8) (1+14) (1+15)

(N_*)-Multispherical Vesicles

Bhatia et al, Soft Matter (2020)

- Multispheres consisting of N_* equally sized spheres:

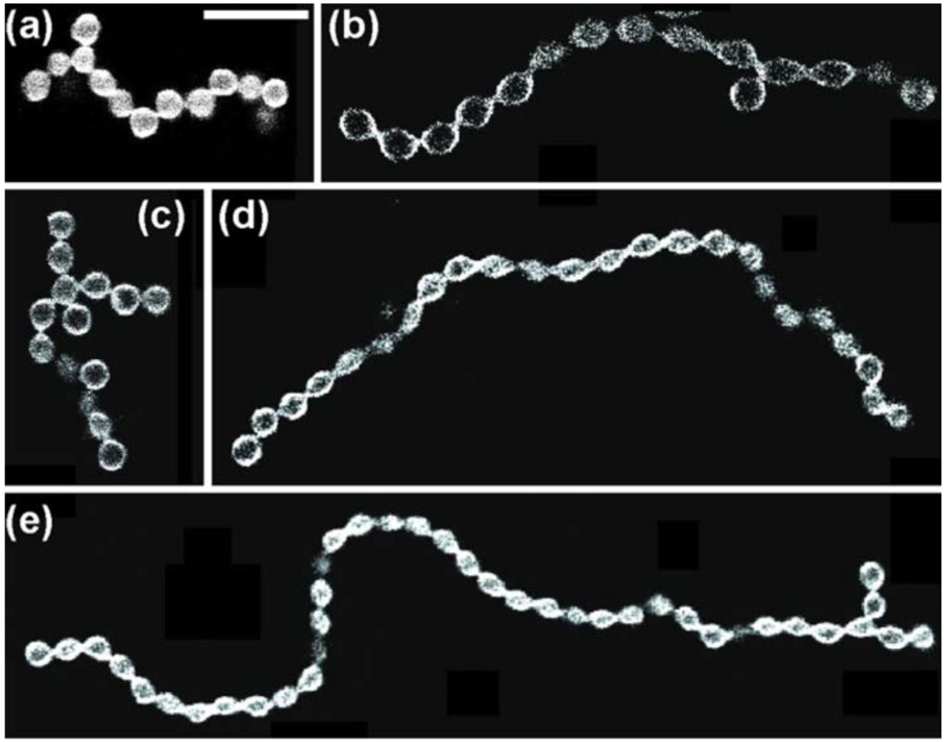
$N_* = 14$ branched

$N_* = 15$ branched

$N_* = 15$ branched

$N_* = 24$ linear

$N_* = 39$ branched



- Surprising mobility: linear ⇔ branched chains

- Remodeling of membrane shape
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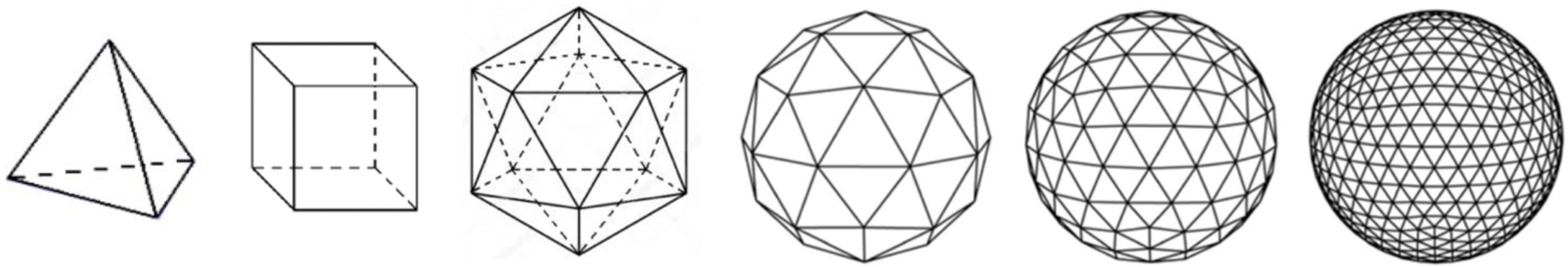
Topology of Surfaces

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

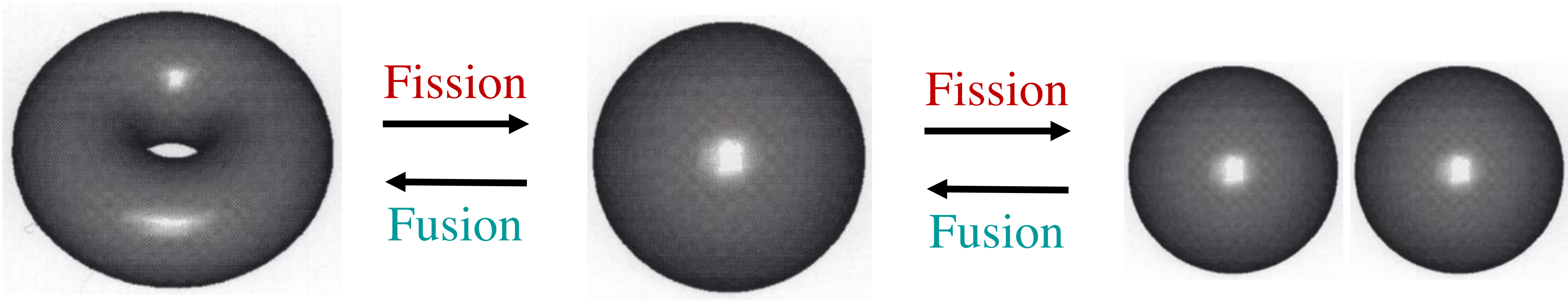


tetrahedron cube icosahedron sphere

- 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

Remodeling of Membrane Topology

• Topological classification via Euler characteristic χ :



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

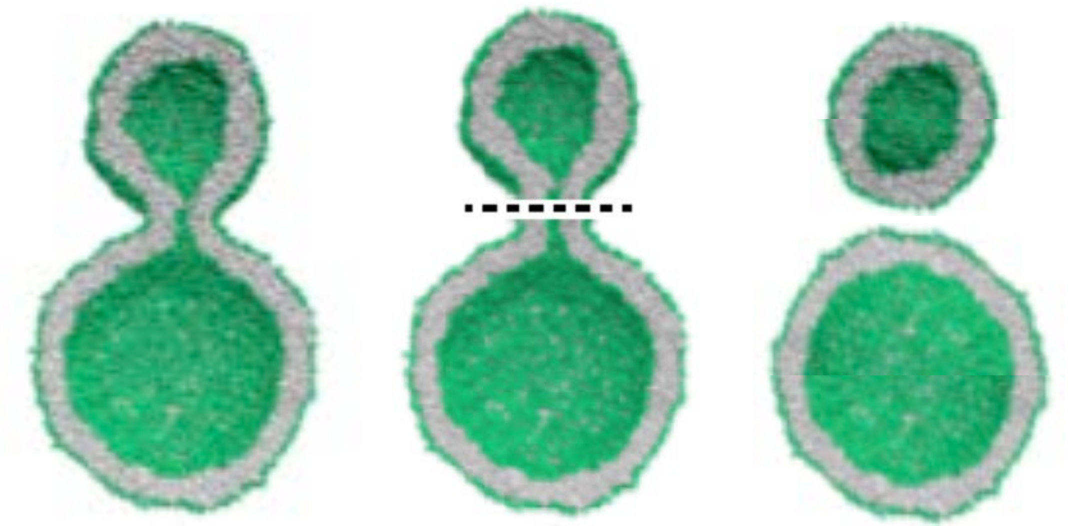
• Topological transformation ⇔ change $\Delta\chi = \chi_{fin} - \chi_{ini}$

• Fission: Euler characteristic $\Delta\chi > 0$

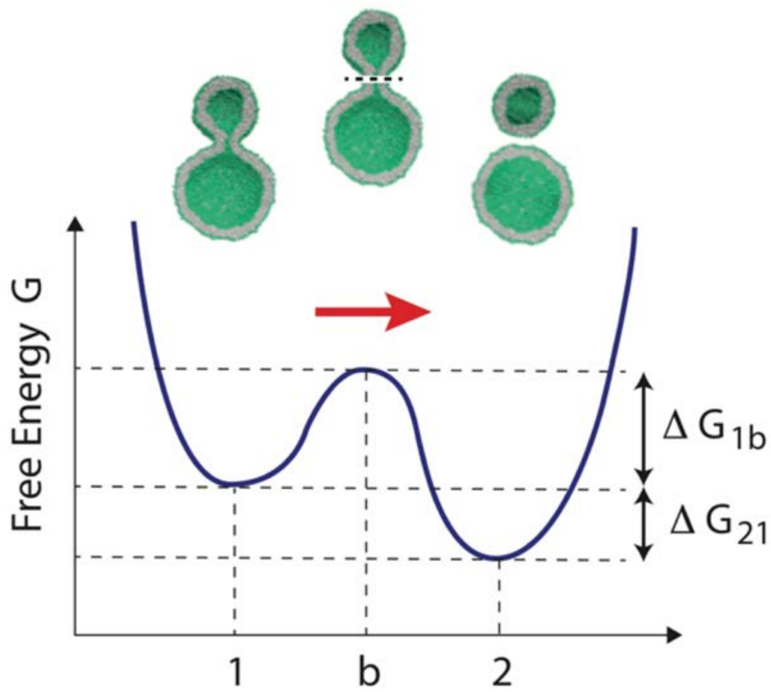
• Fusion: Euler characteristic $\Delta\chi < 0$

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

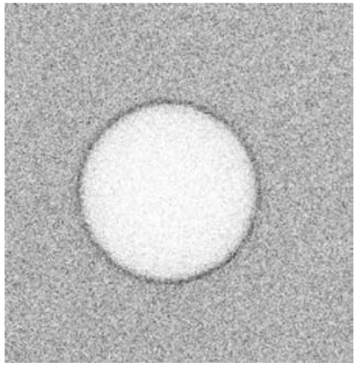


Neck Fission of GUVs

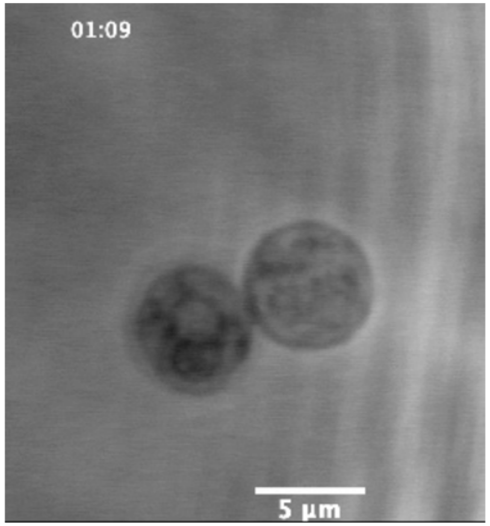
Steinkühler et al: *Nature Comm*. (2020)

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

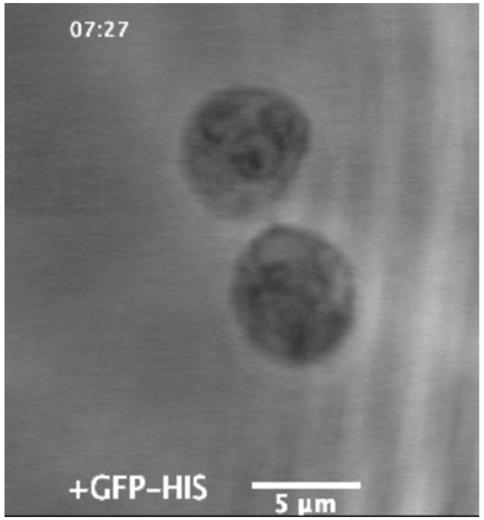
  Increase in GFP -> Neck cleavage -> Two daughter GUVs



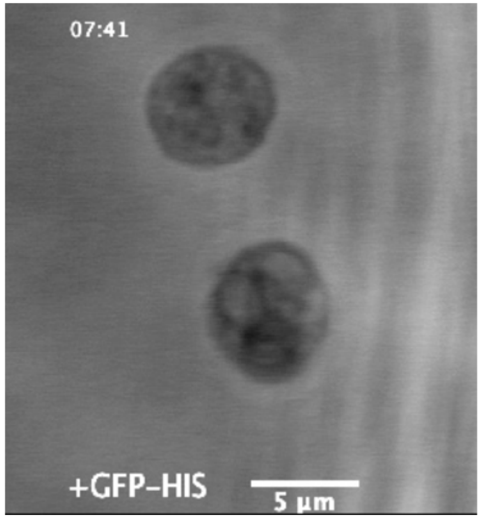
Adsorption of GFP onto GUV membrane



Deflation leads to dumbbell with membrane neck



Directly after neck cleavage



Complete division into two smaller GUVs

Constriction Force from GFP

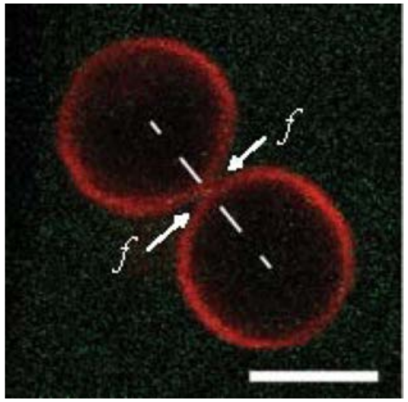
Steinkühler et al:
Nature Comm. (2020)

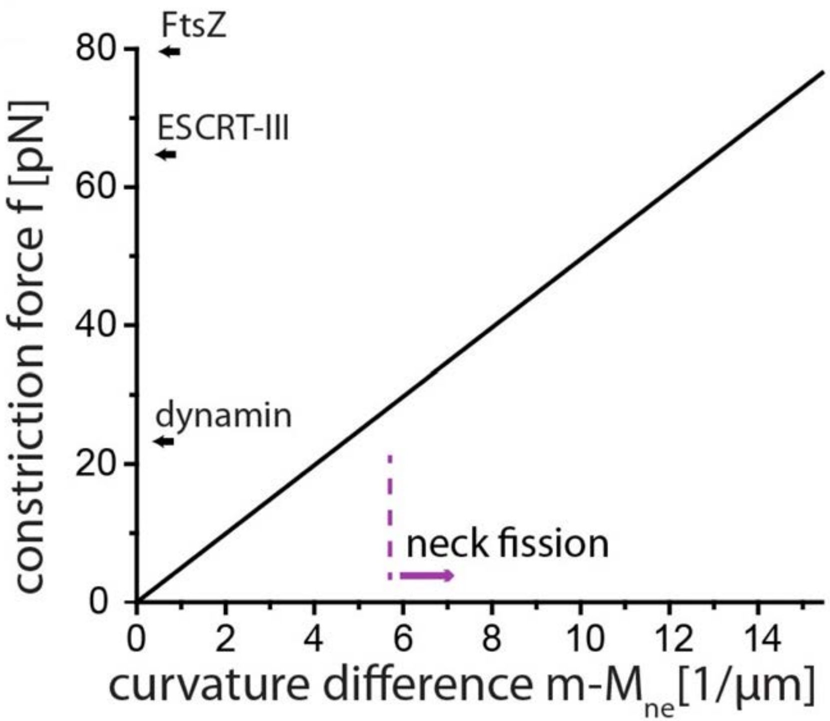
- Small GFP concentrations X in the solution generate large spontaneous curvature

$$m = \frac{1.86}{\mu\text{m}} \frac{X}{\text{nM}} \quad \text{for } 0 < X < 24 \text{ nM}$$

- Constriction force

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

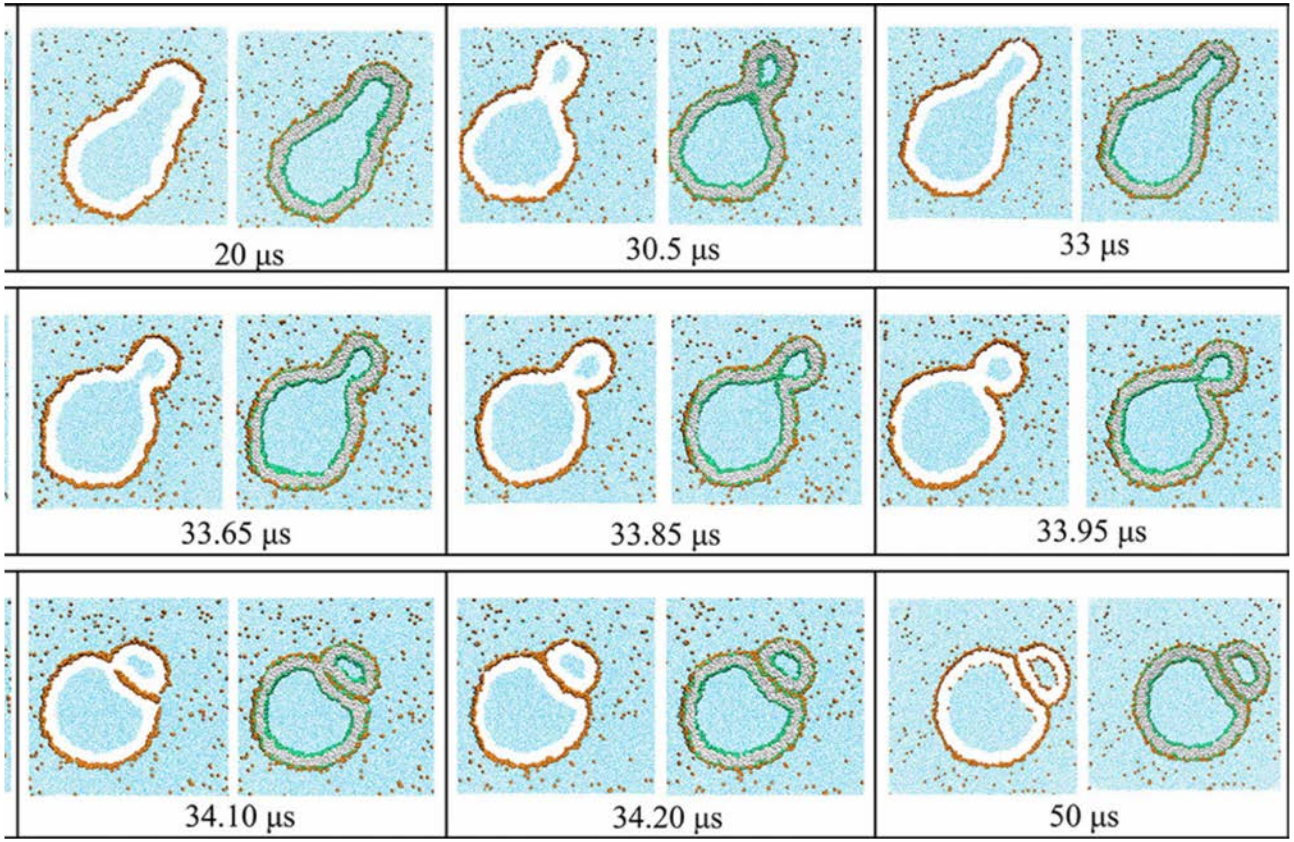


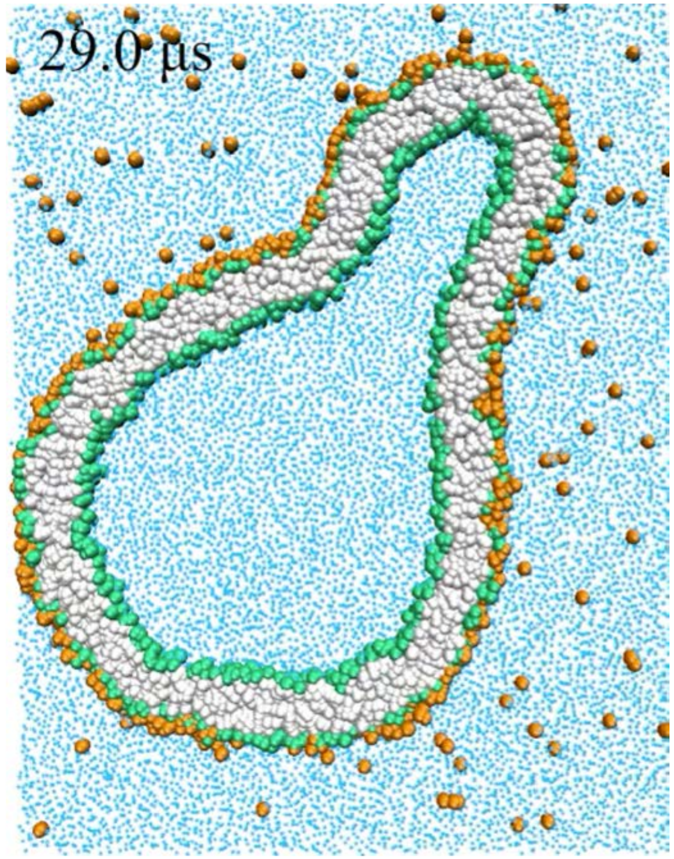


Neck Fission of Nanovesicles

Ghosh et al, ACS Nano (2021)

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

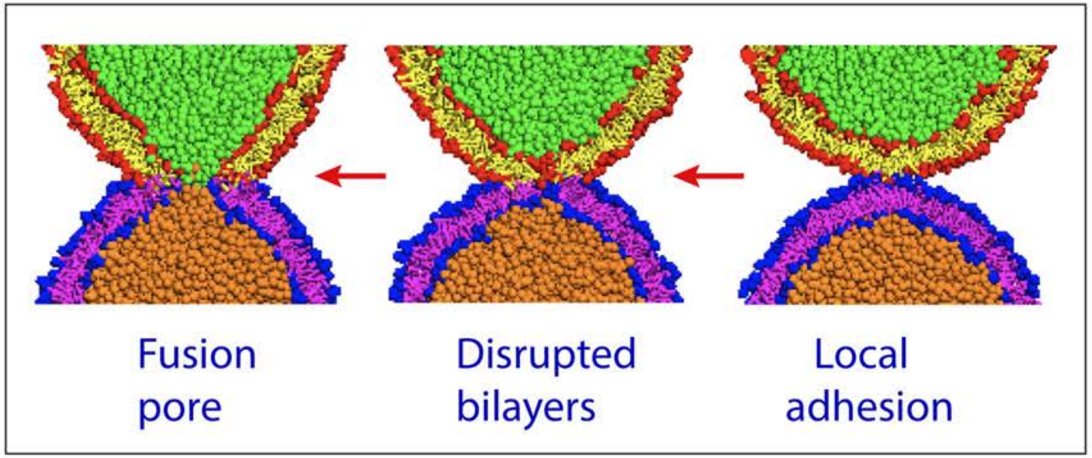


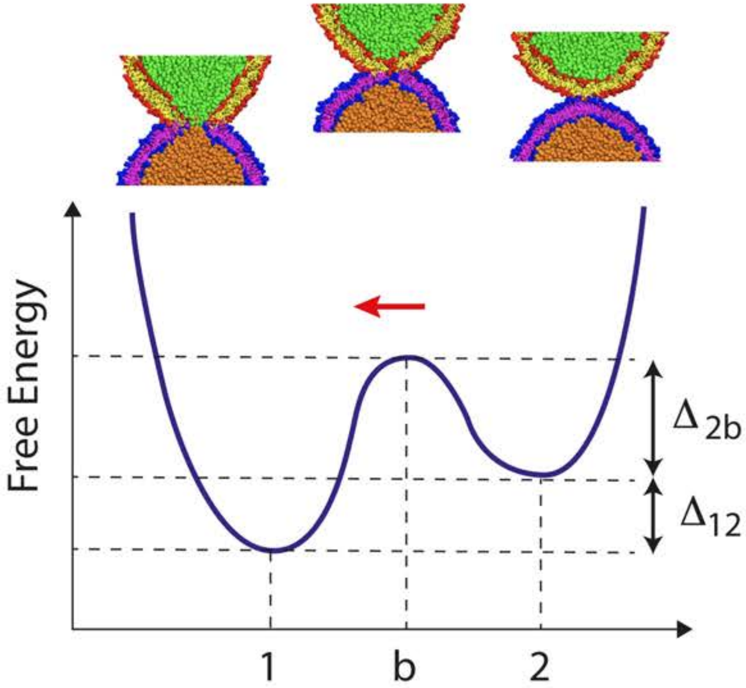


Fusion of Nanovesicles

Shillcock, RL, *Nature Materials* (2005) Grafmüller et al, *Phys. Rev. Lett.* (2007)
Gao et al, *Soft Matter* (2008)

- Two nanovesicles under mechanical tension $\Sigma \sim (A - A_0)/A_0$
- After local contact, fusion pore opens within 2 μs





- Important role of tension in both fusion and fission processes !

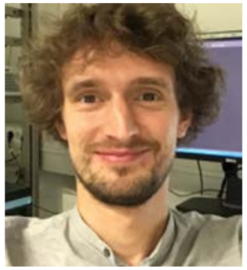
Coworkers

Experiment

Tripta Bhatia

Rumiana Dimova

Yonggang Liu



Jan Steinkühler

Ziliang Zhao

Joint projects with:

Petra Schwille
Thomas Litschel
Seraphine Wegner
Solveig Bartelt

Simulation

Rikhia Ghosh

Andrea Grafmüller

Markus Miettinen

Vahid Satarifard

Aparna Sreekumari

Theory

Simon Christ

- Ongoing projects: Joachim Spatz, Petra Schwille, Tony Hyman, Titus Franzmann

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Experiment

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