

IMAG Theme 3

Top Down Mesoscale Simulations and Free Energy Calculations: Experimentally Validated (Minimal) Models for Receptor Trafficking and Nanocarrier Adhesion

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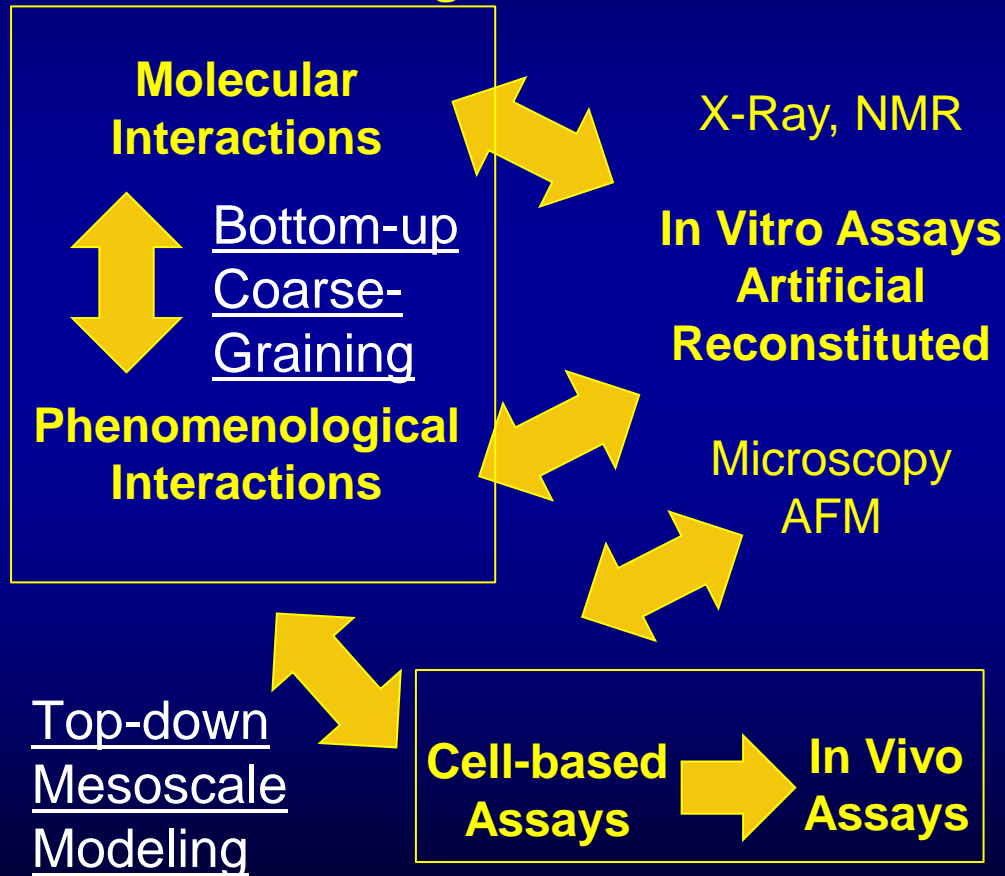
Collaborators: V. M. Muzykantov, T. Baumgart

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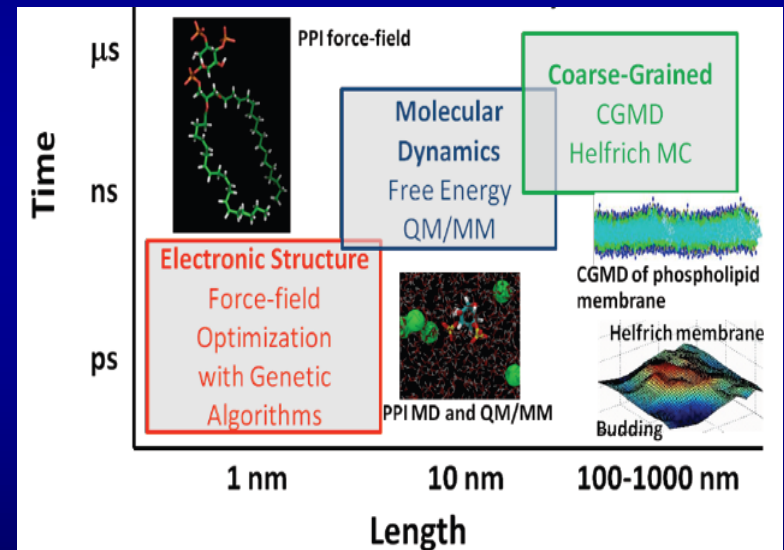


Physically Based Multiscale Models

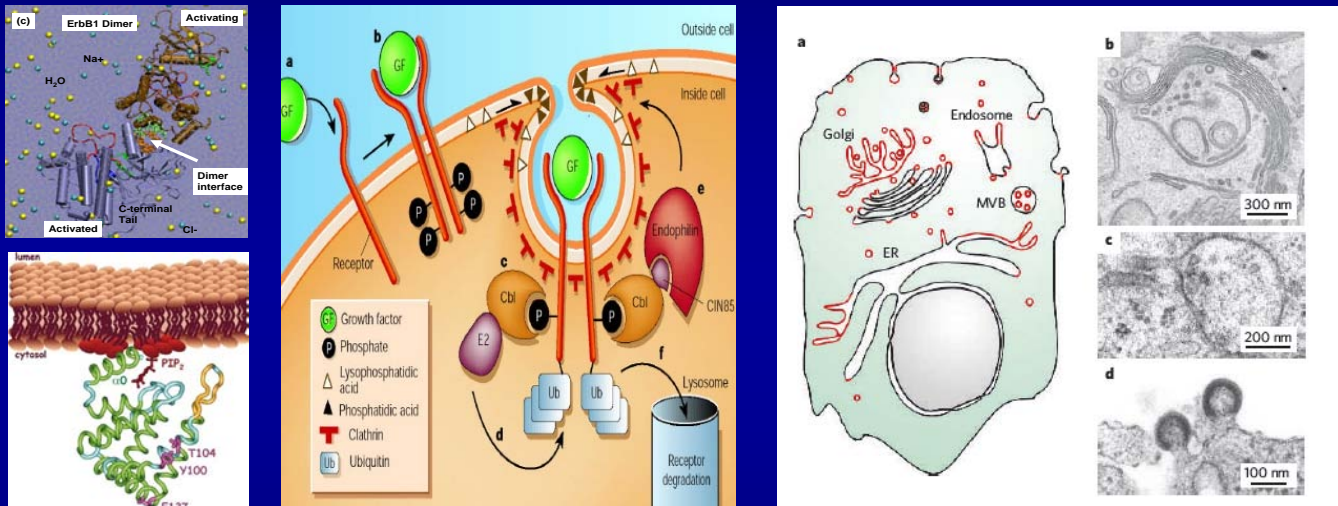
Molecular Modeling



Physical Interactions \leftrightarrow
Mechanisms



Minimal Models for Intracellular Trafficking



Predictive Biophysical Models for Protein-Mediated Membrane Processes at the Mesoscale



Mesoscale Elastic Model for Membranes

Helfrich Free Energy

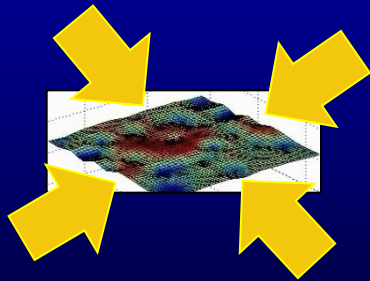
Nelson, Piran, Weinberg, 1987

$$f_c = \frac{1}{2}k(H - H_0)^2 + \bar{k}K.$$

H_0 : Intrinsic curvature

k : Bending Modulus

\bar{k} : Gaussian Curvature Modulus

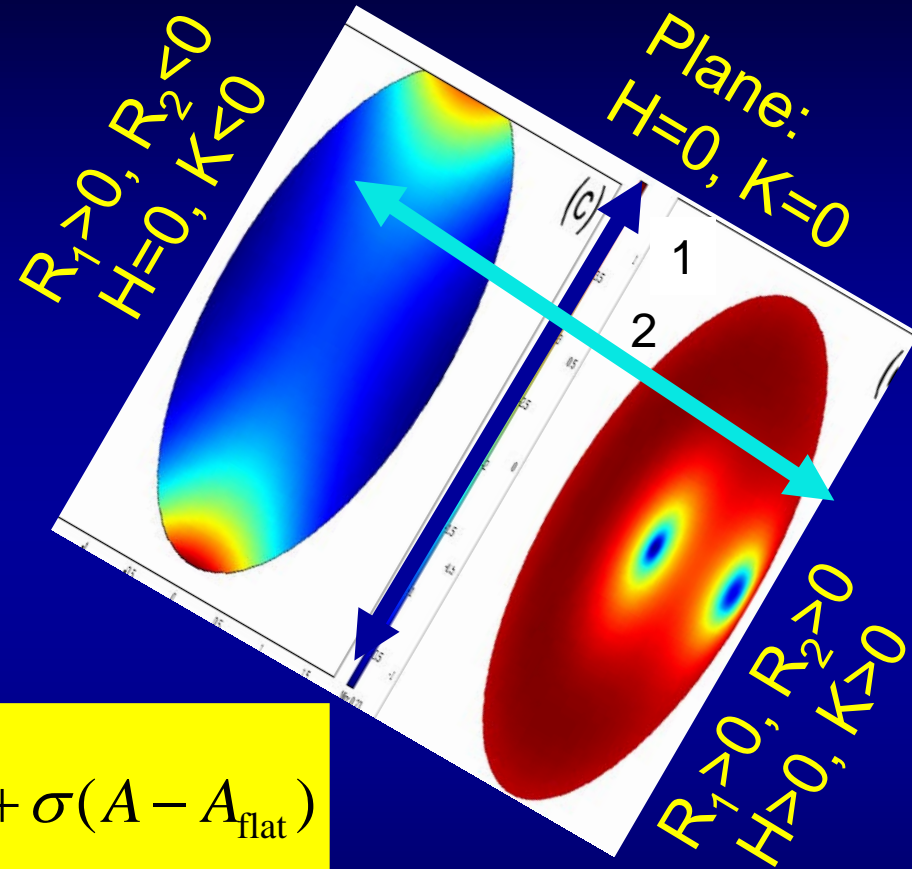


$$E = \int_A \frac{K}{2} (H - H_0)^2 dA + \sigma (A - A_{\text{flat}})$$

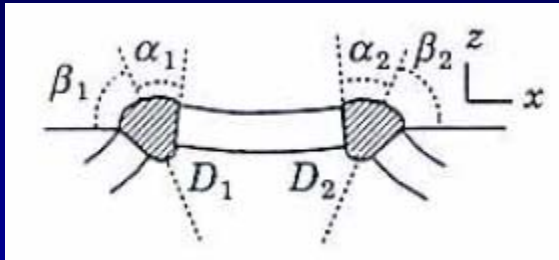
Elastic free energy including frame tension on a membrane patch

$$H \Rightarrow 1/2[1/R_1 + 1/R_2]$$

$$K \Rightarrow 1/R_1 \times 1/R_2$$



Coarse-Grained Representation of Protein-Membrane Interaction



Integral Membrane Proteins

Membrane is attached to the protein at a fixed contact angle

Goulian M, Bruinsma R, Pincus P (1993)

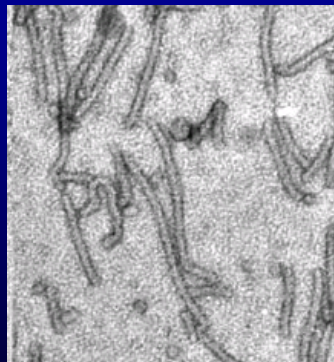
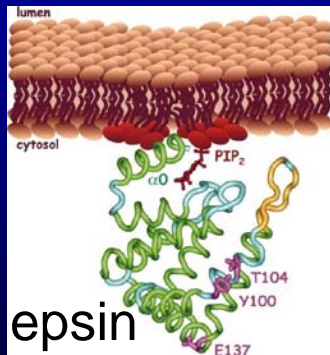
Lubensky T (1997)

Kim KS, Neu J, Oster G (1998)

LOCAL CURVATURE INDUCER MODEL

McMahon, 2003, 2005

Tubule diameter=20 nm; $\Delta E_{\text{binding}} = -14 \text{ k}_B\text{T}$

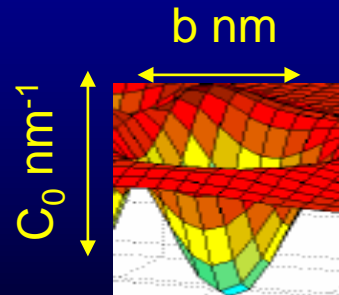


Seifert, et. al. 2006;

Weinstein, Radhakrishnan, 2006;

Agrawal, Weinstein, Radhakrishnan, 2008

$C_0 = 0.1 \text{ nm}^{-1}$; $b = 8 \text{ nm}$; $\kappa = 20 \text{ k}_B\text{T}$



$$H_0 = C_0 e^{-s^2/b^2}$$



Generalized Langevin Dynamics

Helfrich Hamiltonian for fluid lipid membrane [Helfrich (1973) *Z. Naturforsch* **28c** 693]

$$E = \int_s \left[\frac{\kappa}{2} (H - H_0)^2 + \bar{\kappa} K \right] dA$$

$$H = c_1 + c_2 \quad \text{Mean curvature}$$

$$K = c_1 c_2 \quad \text{Gaussian curvature}$$

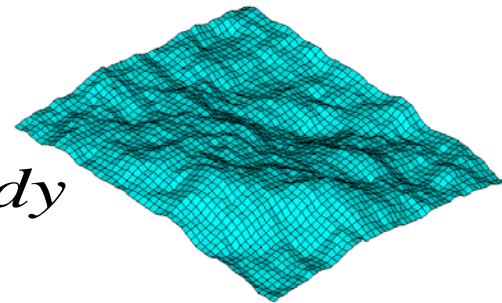
κ : bending rigidity $\bar{\kappa}$: Gauss curvature modulus H_0 : instantaneous mean curvature

Monge (linear) model [Agrawal *et al.* (2008) *Mol. Phys.*]

$$z = z(x, y) \quad A_{ij} = A_{flat,ij} \left[1 + (\nabla z)^2 \right]^{\frac{1}{2}}$$

$$H \approx z_{xx} + z_{yy} = \nabla^2 z$$

$$E = \iint_R \left[\frac{\kappa}{2} (\nabla^2 z - H_0)^2 + \frac{\kappa}{4} H_0^2 (\nabla z)^2 \right] dx dy$$



$$\frac{\partial z(r, t)}{\partial t} = -M \frac{\delta E}{\delta z} + \xi(r, t)$$

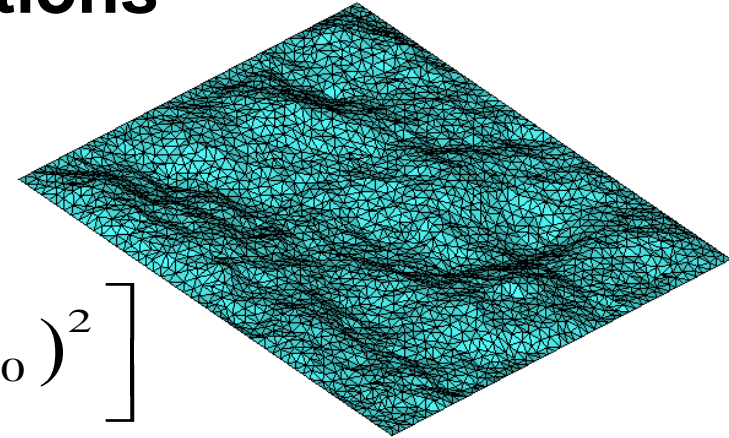
M : mobility term

ξ : thermal noise term



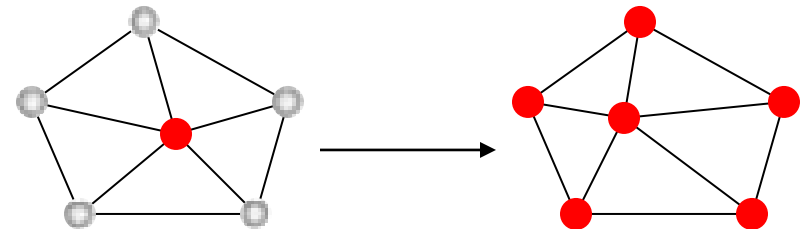
Monte Carlo Simulations

$$E = \frac{\kappa}{2} \int_s dA (H - H_0)^2$$
$$= \sum_{v=1}^N A(v) \left[\frac{\kappa}{2} (c_1(v) + c_2(v) - H_0)^2 \right]$$

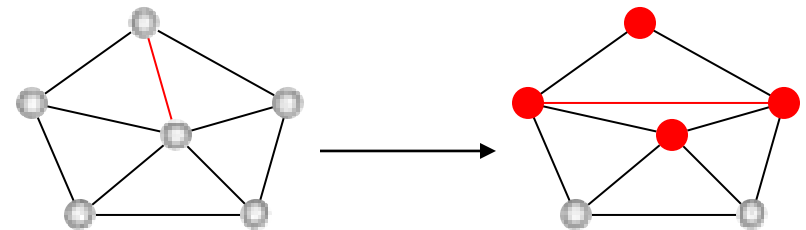


Curvatures are calculated through coordinate transformation between global and local Darboux frame. One of the two moves is randomly selected and the energy change is calculated.

Vertex shifts:



Link flip:



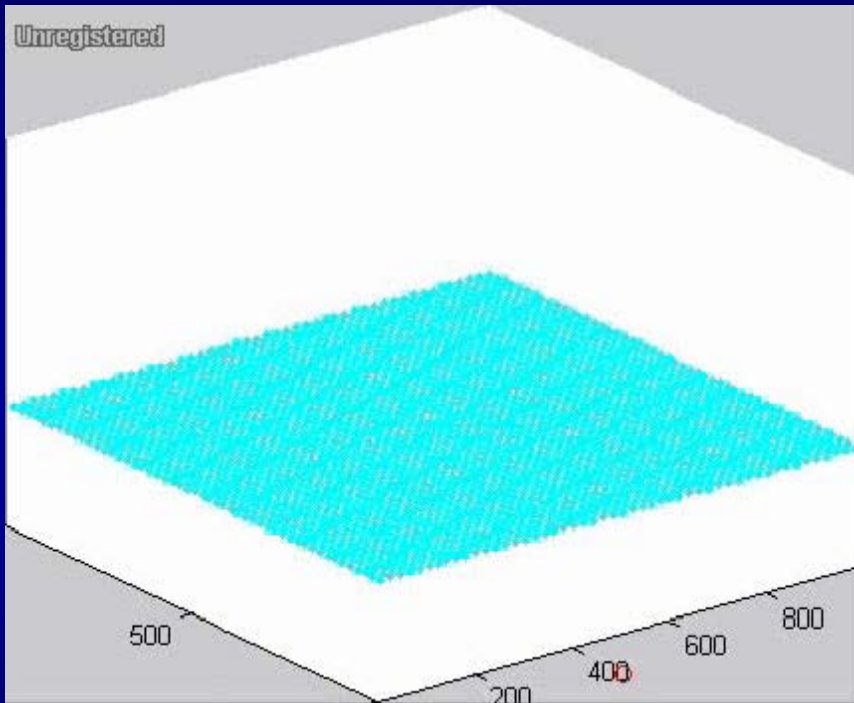
[Ramakrishnan *et al.* (2010)
PRE **81** 041922]



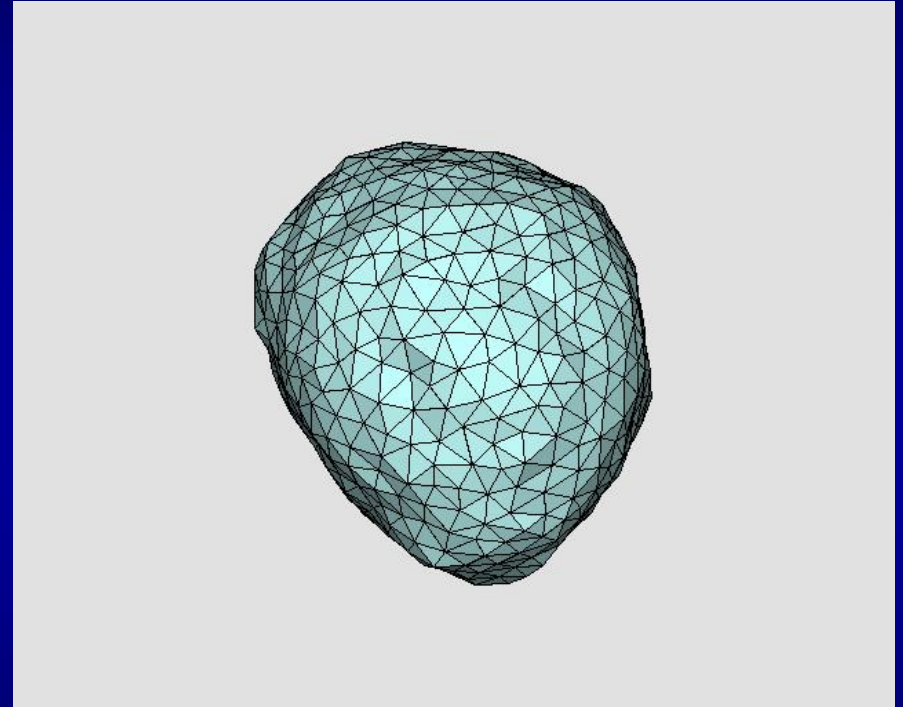
Protein-Mediated Membrane Fluctuations

Weinstein, Radhakrishnan, Mol Phys, 2006;
Agrawal, Radhakrishnan, Mol Phys, 2008

Langevin Dynamics



Monte Carlo



Proteins perform a random walk on membrane surface with a membrane mediated force field



Membrane-Mediated Potential of Mean Force (PMF) between Proteins

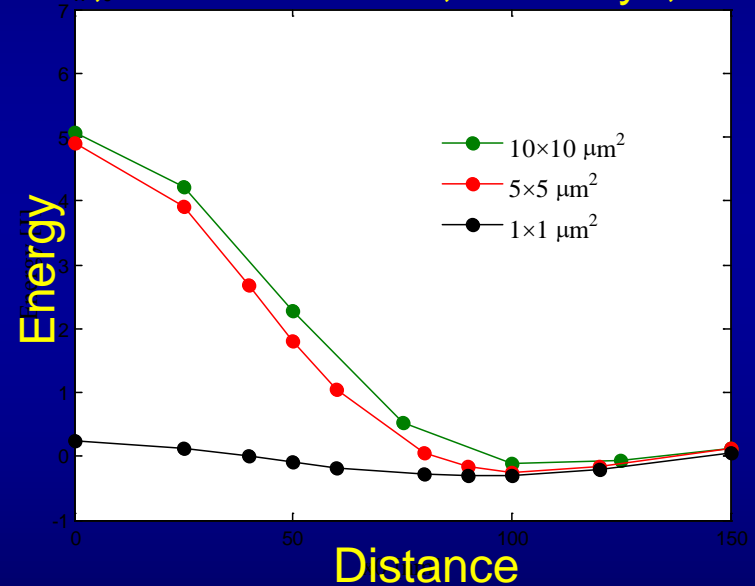
- PMF is dictated by both energetic and entropic components
- **Energy:** Epsin experience repulsion due to energetic component when brought close.
- **Entropy:**

$$\delta^2 E(\phi) = \iint_A \kappa (\nabla^2 \phi)^2 + \left(\frac{\kappa}{2} H_0^2 + \sigma \right) (\nabla \phi)^2 dx dy > 0$$

$\delta^2 E \sim$ spring constant; ϕ =test function

- Regions of non-zero H_0 assume increased stiffness and hence reduced membrane fluctuations
- The system can lower its free energy by localizing epsins on the membrane

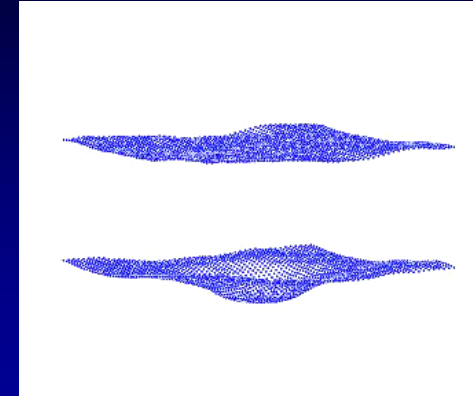
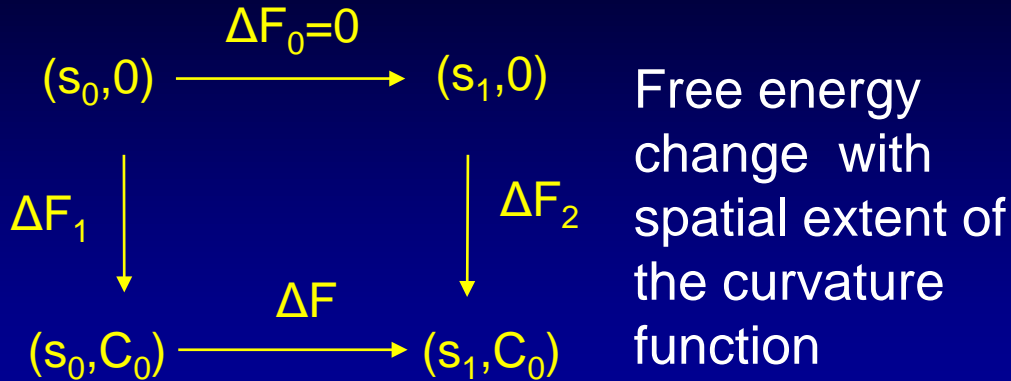
Agrawal, Radhakrishnan, Mol Phys, 2008



Need for Free Energy Calculations



Free Energy Calculations via Thermodynamic Integration



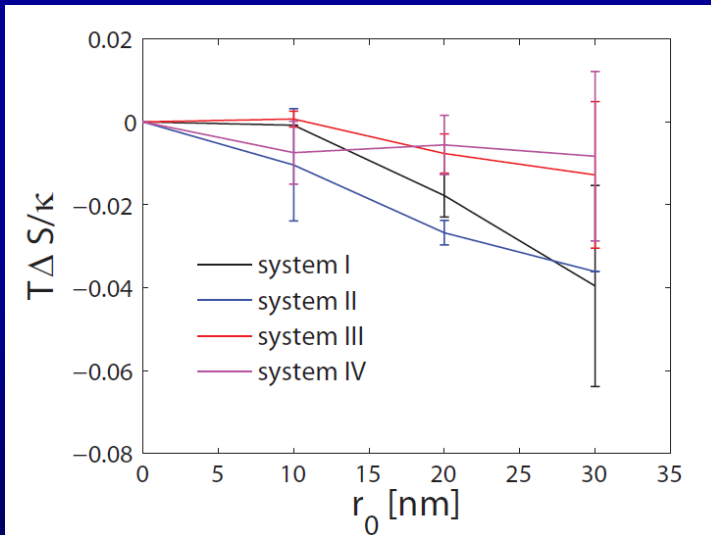
Agrawal, Radhakrishnan, PRE, 2009

$$\left(\frac{\partial F}{\partial \lambda} \right)_{N,V,T} = -\frac{1}{\beta} \frac{\partial}{\partial \lambda} \ln Q = \left\langle \frac{\partial E}{\partial \lambda} \right\rangle_{\lambda}$$

By choosing $\lambda = C_0$,

$$\frac{\partial F}{\partial C_0} = \left\langle \Gamma(r_0) \kappa \sum_N \left[-(\nabla^2 z_i - \lambda \Gamma(r_0)) + \frac{\lambda}{2} (\nabla z_i)^2 \right] (\Delta r_i)^2 \right\rangle$$

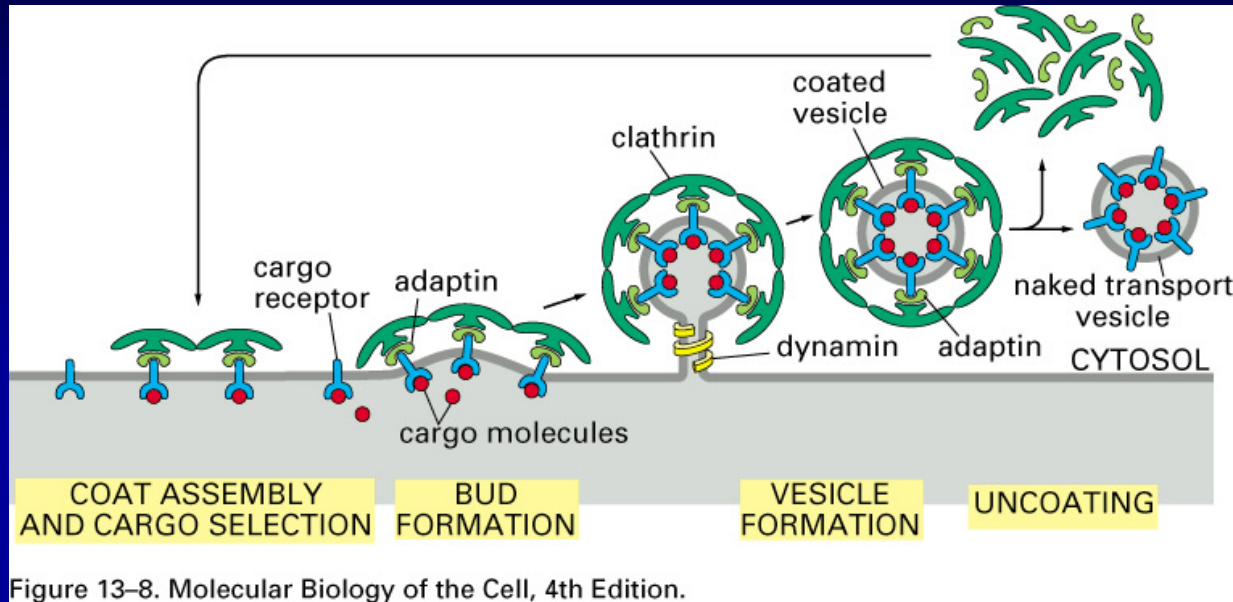
$$\Delta F = F(C_0) - F(0) = \int_0^{C_0} \frac{\partial F}{\partial C_0} dC_0$$



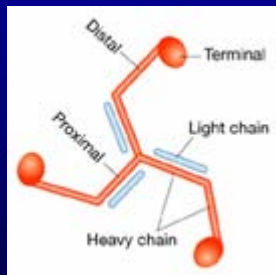
Entropy change is small (<5% of the Energy change) but is of order $k_B T$



Vesicle Nucleation in Clathrin-Mediated Endocytosis

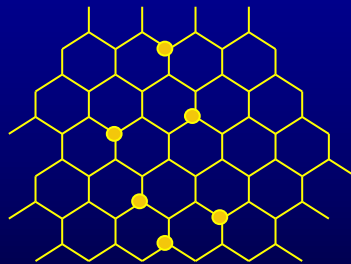


Clathrin



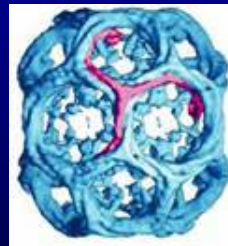
trimer

Clathrin Coat

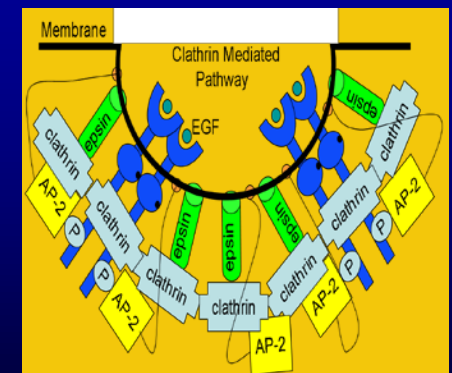


trimer:AP-2=1:1

Clathrin Cage



27 (50nm)

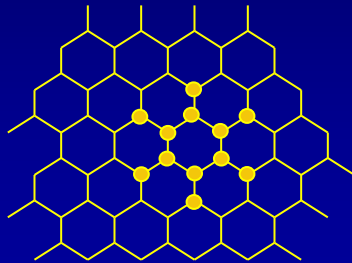


Bond-Orientational Patterning of Epsin on Clathrin Lattice Leads to a Mature Vesicle Formation

Kirschhausen (2000)

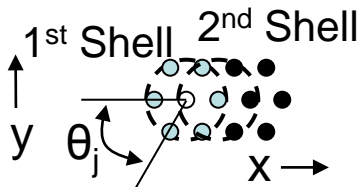
18.5 nm

Clathrin Coat



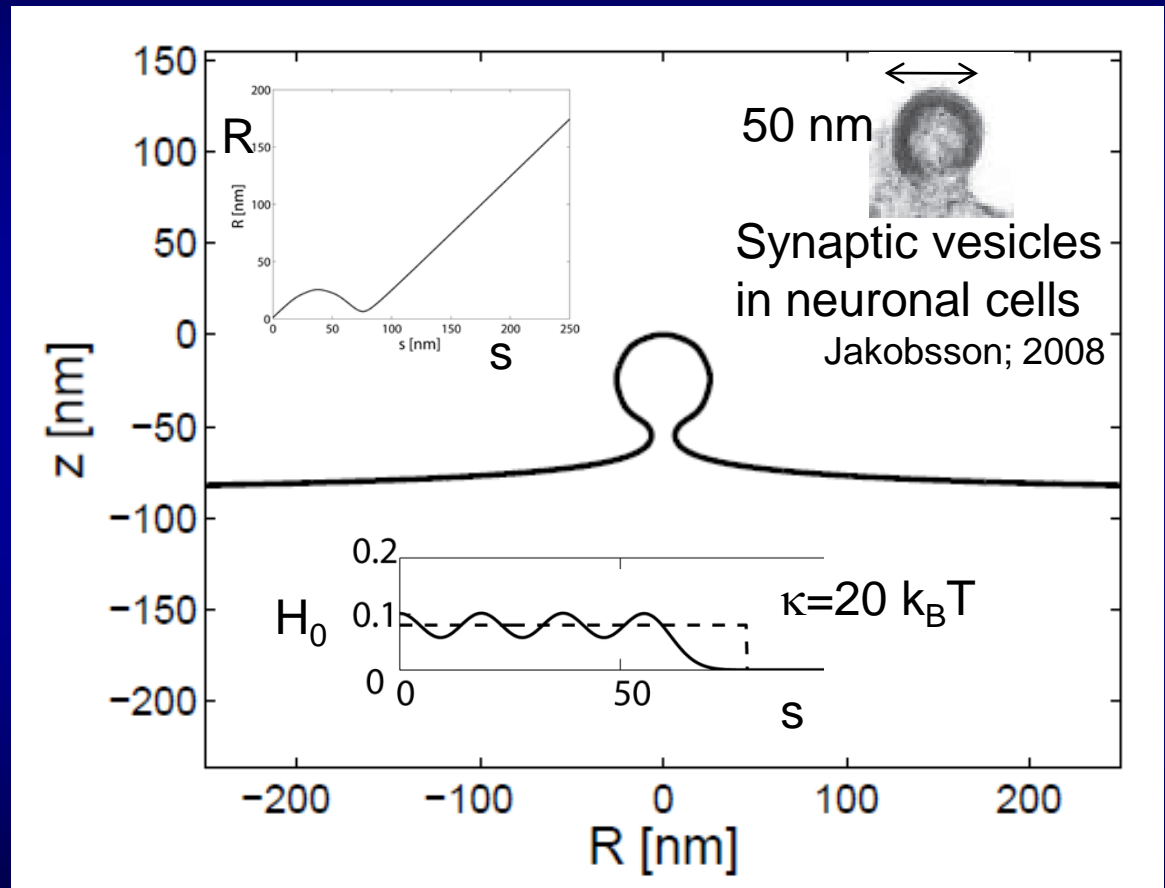
Area = A_a

Epsin arrangement



$$\langle \exp(i6\theta(r)) \rangle$$

$$\langle \Psi_6^*(0) \Psi_6(r) \rangle$$



[Agrawal, Radhakrishnan, Plos Comput Biol, 2010]



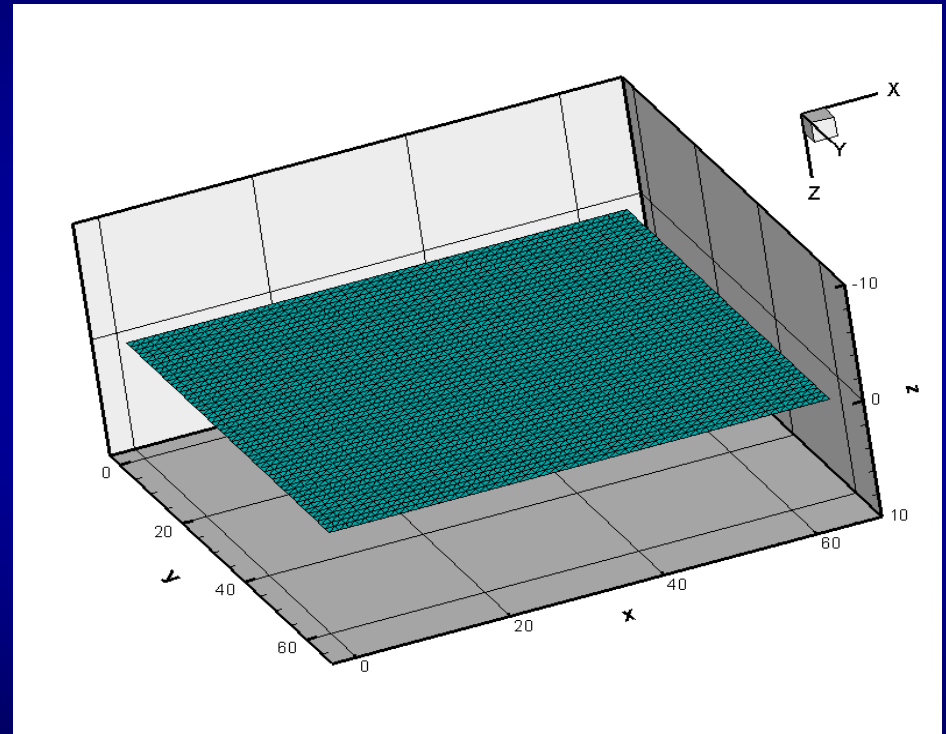
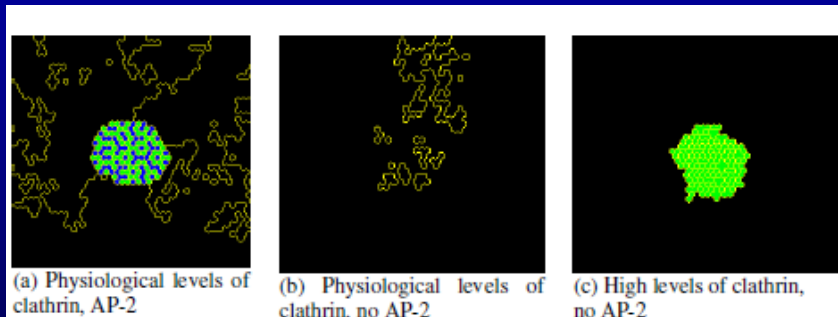
University of Pennsylvania

Department of Bioengineering

Receptor Trafficking: Bioenergetics of Clathrin Induced Membrane Vesiculation

Agrawal, Radhakrishnan, Plos Comput Biol, 2010

Ramanan et al, Integrative Biology, 2011

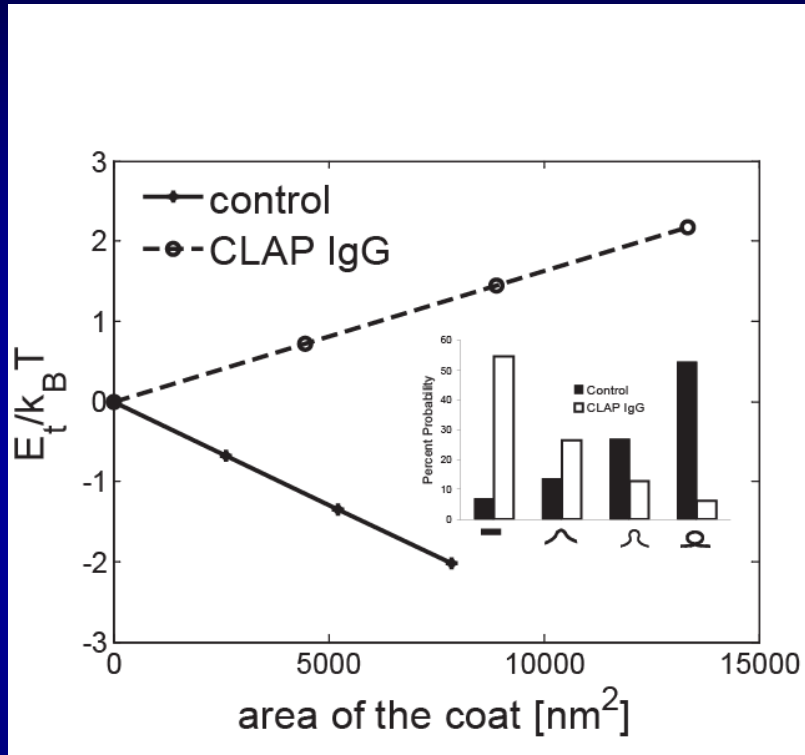


Weak hydrophobic interactions lead to self assembly of clathrin coat
Epsin-Induced Curvature Stabilizes Mature Vesicular Intermediates



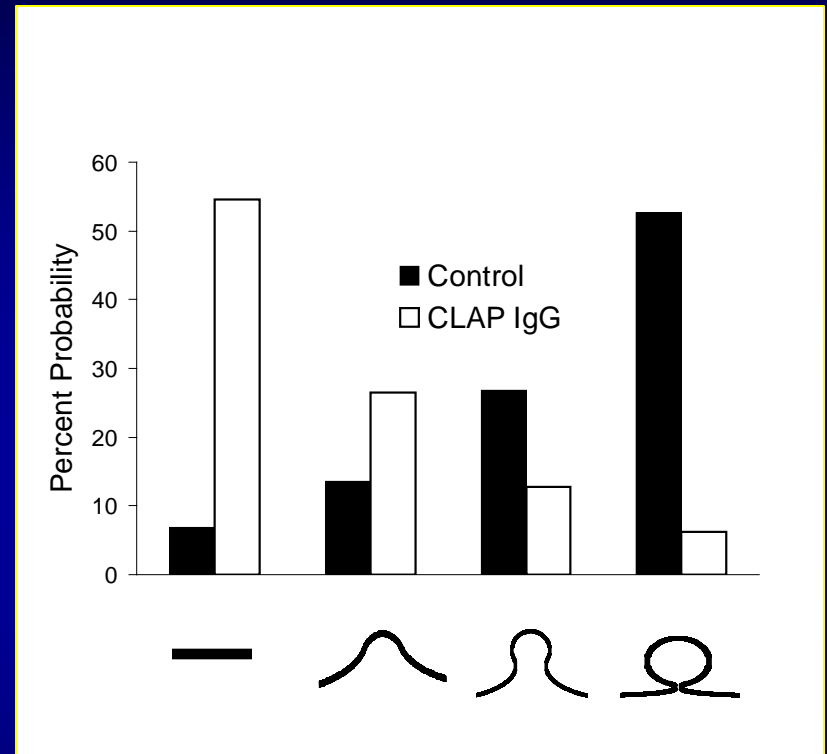
Free Energy Considerations in Clathrin Induced Membrane Vesiculation

Calculated



Agrawal, Radhakrishnan, Plos Comp Biol, 2010

Experimental

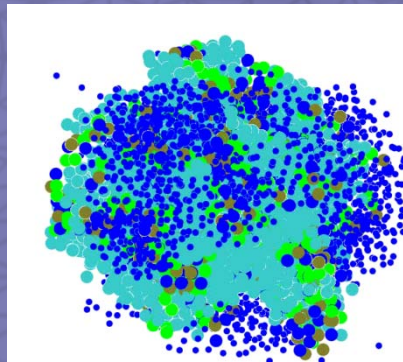
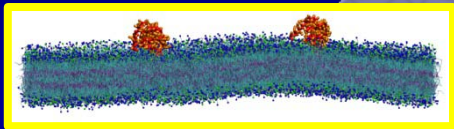


Jakobsson, J.; PNAS 2008, 6445.

Our results highlight the unique and central role played by epsin in the process of vesicle nucleation during endocytosis



**Ensemble of Epsin-Membrane
CGMD Simulations; each
replica with a unique lateral
pressure**



CGMD gives curvature field



**Mesoscale MC model
gives positions and frame
tension (lateral pressure)**

**Two-way multiscale coupling; epsin-
membrane simulations in will quantify:**

1. Curvature field
2. Curvature induction by multiple epsins
3. Local stress fields

**CG/CM Interface for
Coupling CGMD: coarse-
grained molecular
dynamics with CM:
continuum Helfrich MC**



**Model for predicting
protein partitioning
and vesicle budding
events under various
conditions:
different proteins,
cytoskeletal tension,
ECM stiffness**

