Physiologically Validated Models for Adhesion of Functionalized Nanocarriers to Endothelium in Targeted Drug Delivery

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Targeted Drug Delivery using Functionalized Nanocarriers

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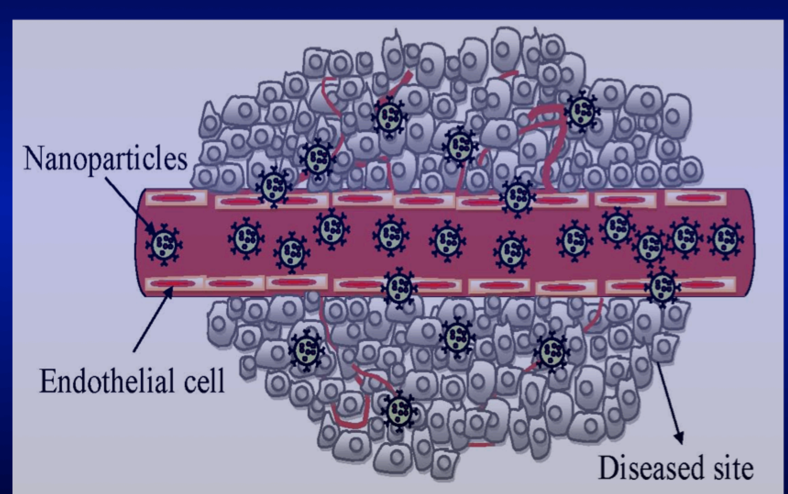
Specific receptor proteins expressed on pathological cells provide good target. Coating specific ligands onto the nanocarrier surface helps enhance the efficiency of binding.

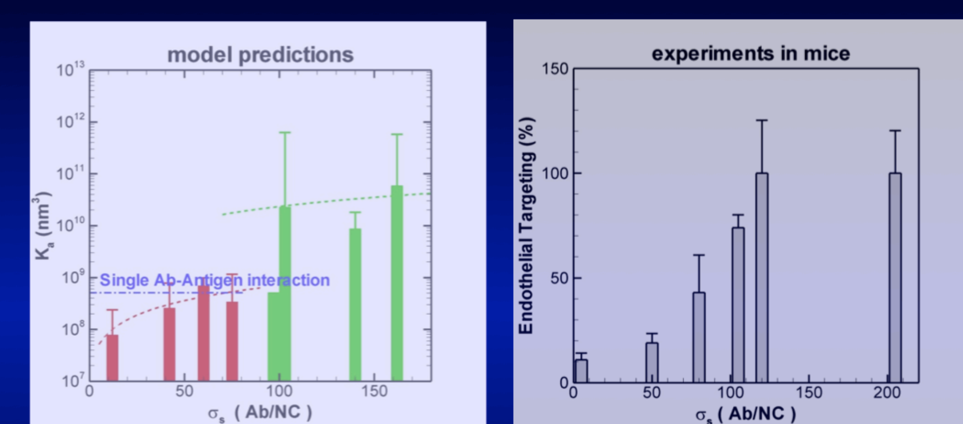


Time scales (near-wall):
1. Hydrodynamic
2. Brownian relaxation
3. Cell Membrane
4. Adhesion

Length scales:
1. Hydrodynamic
2. Cell Membrane
3. Antigen-antibody interaction
4. Antigen flexibility

How do the nanocarrier (NC) size, shape, ligand surface coverage etc. affect efficiency of targeting in vitro and in vivo?

Effect of Surface Coverage σ_s



- A threshold at σ_s ~45% (100Ab/NC), the binding affinity abruptly drops below that of single antibody to antigen
- Linear dependence below and above the threshold at fixed multivalency, dotted lines.
- Exponential reduction because of the multivalency change (from 3 to 2) around σ_s ~45%

Model predictions are consistent with results of in vivo mice experiments

Role of Cell Membrane Undulations in Nanocarrier Adhesion

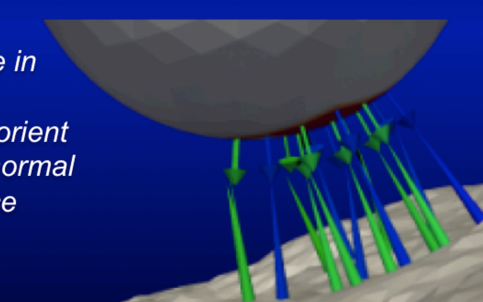
Undulating membrane is modeled as a thin, elastic, fluid surface with energy

$$\mathscr{H}_{\rm mem} = \int \frac{\kappa}{2}\left(2H - C_0\right)^2 ds$$

Bending rigidity — Mean curvature — Spontaneous curvature

Membrane undulations are simulated using dynamically triangulated Monte Carlo.

Periodic membrane in a simulation box; Unflexed antigens orient along the surface normal Triangulated surface representation of membrane

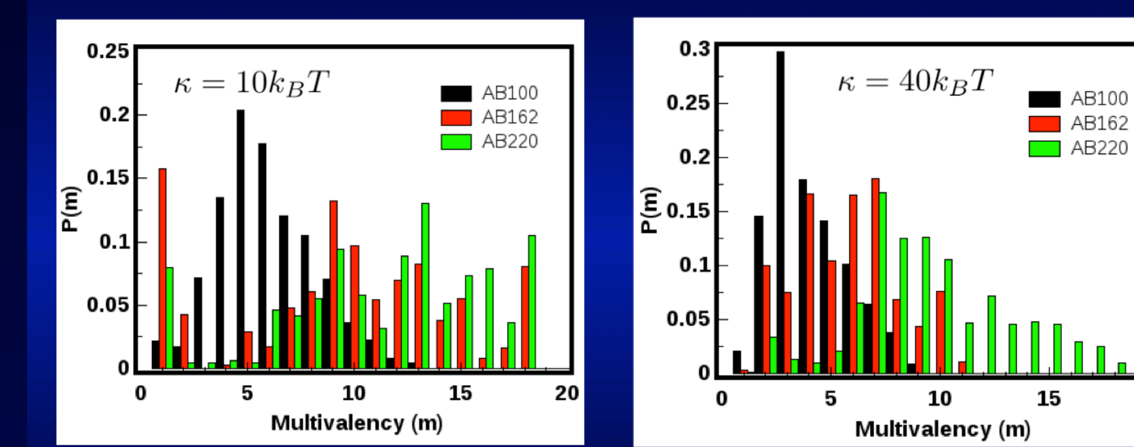


Membrane Properties
(a) Bending rigidity
(b) Surface tension
(c) Excess surface area

How does membrane mobility and surface curvature affect nanocarrier (NC) binding ?

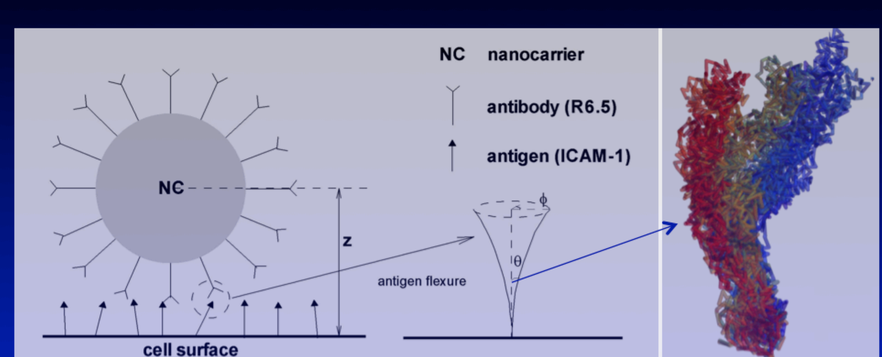
Effect of Membrane Bending Rigidity on Multivalency

lower bending stiffness $\left\langle \frac{A}{A_p} \right\rangle = 1.06$ higher bending stiffness



Systematic decrease in multivalent interactions is observed with increasing stiffness; the multivalency approaches the statistics of flat substrate as $\kappa \rightarrow \infty$

Minimal Model for Nanocarrier (NC) Adhesion



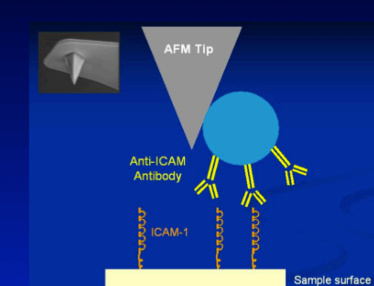
- Spherical 100nm NCs coated with uniformly distributed N_{ab} antibodies (Ab). The saturation Ab surface coverage (100%) σ_s=220Ab/NC in experiments.
- Antibody-antigen interaction is treated using the Bell model:

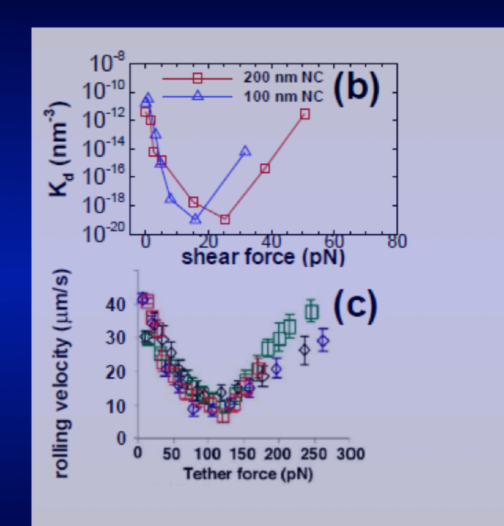
$\Delta G_r(d)=\Delta G_0+k_0d^2/2$ k_0: bond force constant

- Antigen flexure accounted for by orientational-bias MC sampling of θ and ϕ
- Calculation of Potential of Mean Force leading to absolute binding free energy

Objective: How do we quantify NC binding to cells mediated by antibody-antigen interactions?

AFM Binding/Rupture and Effect of Flow on NC Adhesion: Shear Enhanced Binding and Rolling Behavior



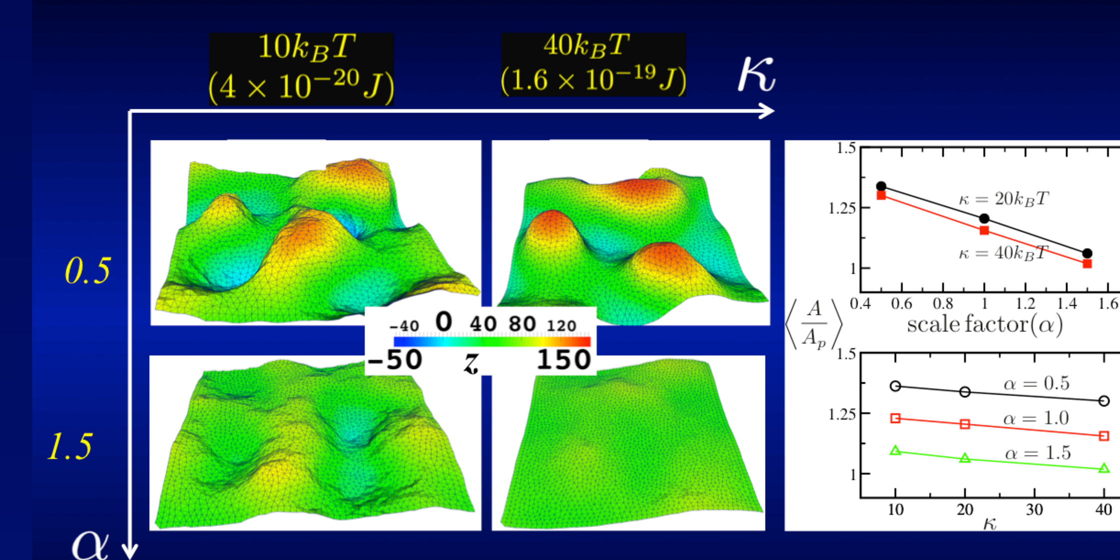


Model predicts: 230±41pN
AFM experiments (89 trials): 316±48pN

Model prediction of rupture force distribution is consistent with AFM experiment

Model achieves simultaneous thermodynamic, physiological, and mechanical consistency in the presence as well as absence of flow. This shows potential for minimal models to guide the design of nanocarriers for targeted drug delivery

Excess Area Fluctuations in Membrane



Excess area of the membrane decreases with increasing bending rigidity (κ) and is controlled by length scale factor (α).

Membrane Curvature-Undulation Coupling

When a nanocarrier binds to the membrane it induces a preferred curvature and this couples to the surface fluctuations and modulates its physical properties.

Energy of a heterogeneous planar membrane:

$$\mathscr{H}_{\rm mem} = \int \frac{\kappa(\mathbf{x})}{2}\left\{\nabla^2 h(\mathbf{x}) - C_0\right\}^2 ds.$$

In the Fourier space, the average energy becomes

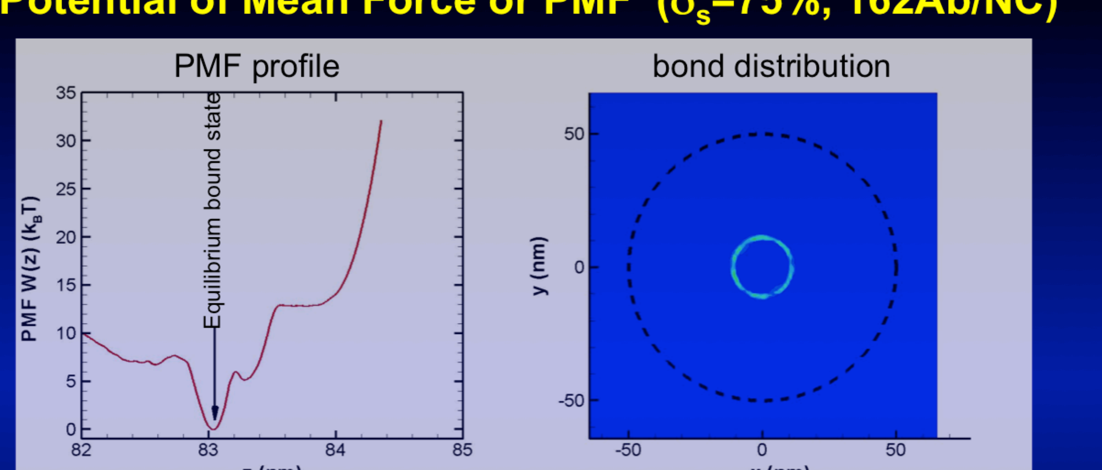
$$\langle \mathscr{H}_{\rm mem}\rangle = \frac{1}{2A_p}\sum_{\mathbf{q}}\sum_{\mathbf{q}'}\left\{\mathbf{q}^2\mathbf{q}'^2\left\langle h_{\mathbf{q}}h_{\mathbf{q}'}\right\rangle - \mathbf{q}^2\left\langle h_{\mathbf{q}}C_{0,\mathbf{q}'}\right\rangle - \mathbf{q}'^2\left\langle C_{0,\mathbf{q}}h_{\mathbf{q}'}\right\rangle + \left\langle C_{0,\mathbf{q}}C_{0,\mathbf{q}'}\right\rangle\right\}\kappa(\mathbf{q}+\mathbf{q}')$$

The modes of the homogeneous membrane obeys the scaling relation

$$A_p\langle h_q h_{-q}\rangle = (\kappa q^4)^{-1} \quad \text{when } \kappa(q+q') = \delta_{qq'}$$

Multivalent binding of NC to membrane induces curvature, which couples to (and modulates) membrane undulations. This coupling can have an impact on emergent membrane morphology and on the initiation of intracellular signaling complexes mediated by curvature sensing proteins

Potential of Mean Force or PMF (σ_s=75%, 162Ab/NC)



- 3 firm bonds formed with an energy well ~$32k_BT$
- Annulus (bond) distribution with outer radius r_o and inner radius r_i
- The areas can be calculated as:
- Association constant is given by:

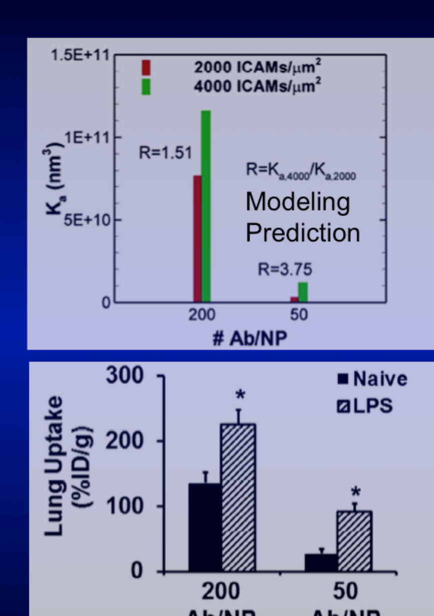
$$K_a = \frac{(N_{ab}/N_b)\Delta\omega}{8\pi^2}\times\frac{A_{R,b}^{(1)}\times A_{R,b}^{(2)}\times\ldots\times A_{R,b}^{(N_b)}}{A_{R,ub}^{(1)}\times A_{R,ub}^{(2)}\times\ldots\times A_{R,ub}^{(N_b)}}\times A_{NC,b}\int e^{-\beta W(z)}\,dz$$

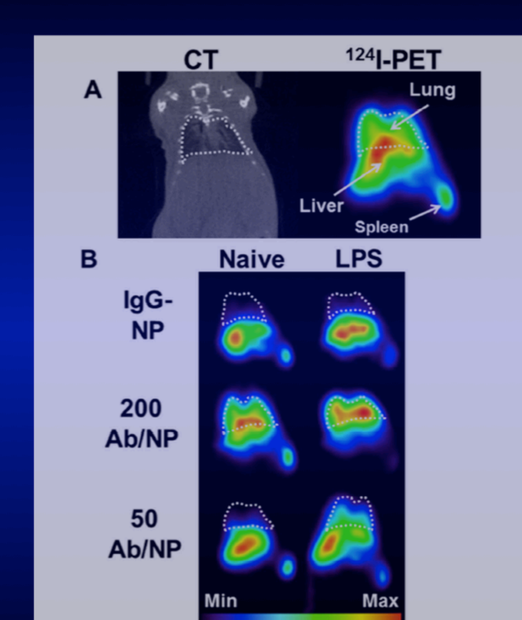
$$A_{R,b}^{(1)} = \pi r_o^2 \qquad A_{R,b}^{(2)} = \pi\left(r_o^2 - r_i^2\right)$$

$$A_{R,b}^{(3)} = A_{NC,b} = \left(r_o - r_i\right)^2$$

$$A_{R,ub}^{(1)} = A_{R,ub}^{(2)} = A_{R,ub}^{(3)} = \pi r_o^2$$

Tissue Selectivity Predictions

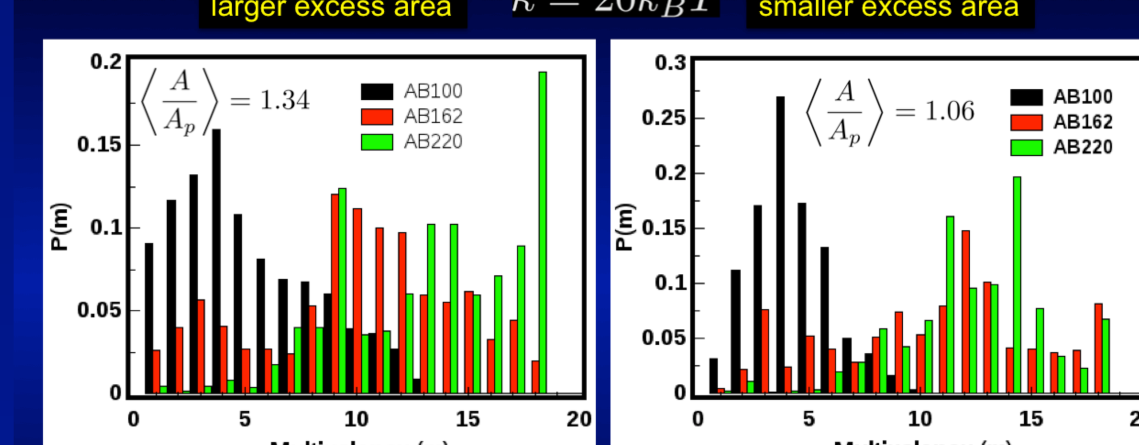




Model correctly predicts the reduction of nanoparticle avidity enhances the selectivity of vascular targeting and PET detection of pulmonary inflammation in mice

Effect of Excess Membrane Area and Undulations on Multivalency

larger excess area $\kappa = 20k_BT$ smaller excess area



Excess surface area in the membrane promotes higher multivalent binding

Presence of excess membrane area can significantly impact nanocarrier binding to the cell membrane and can promote wrapping of cargo which is extremely important in uptake of nanocarriers by cells.

Bibliography Summarizing Current Work

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