Leveraging *ab initio* modelling approaches to link phys-chem nanomaterial (NM) properties to descriptors of bio-nano interaction

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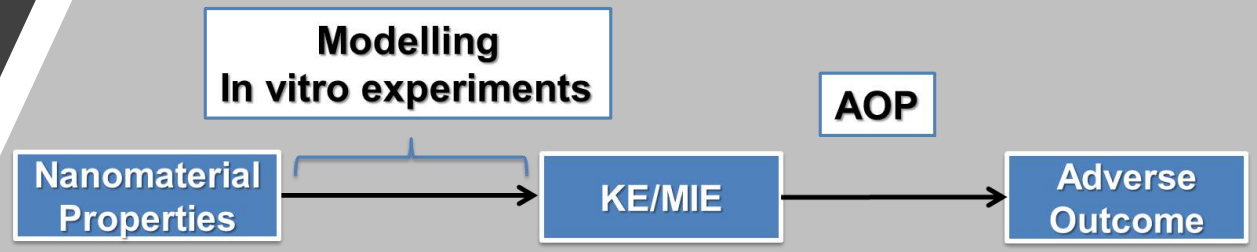
Pathway based approach

- We seek an explanation based on molecular level interactions between nanomaterials and membranes with a **molecular initiating event (MIE)** leading to an **adverse outcome** (toxic)

- Capture this in **software** that correlates nanomaterials properties and toxicity

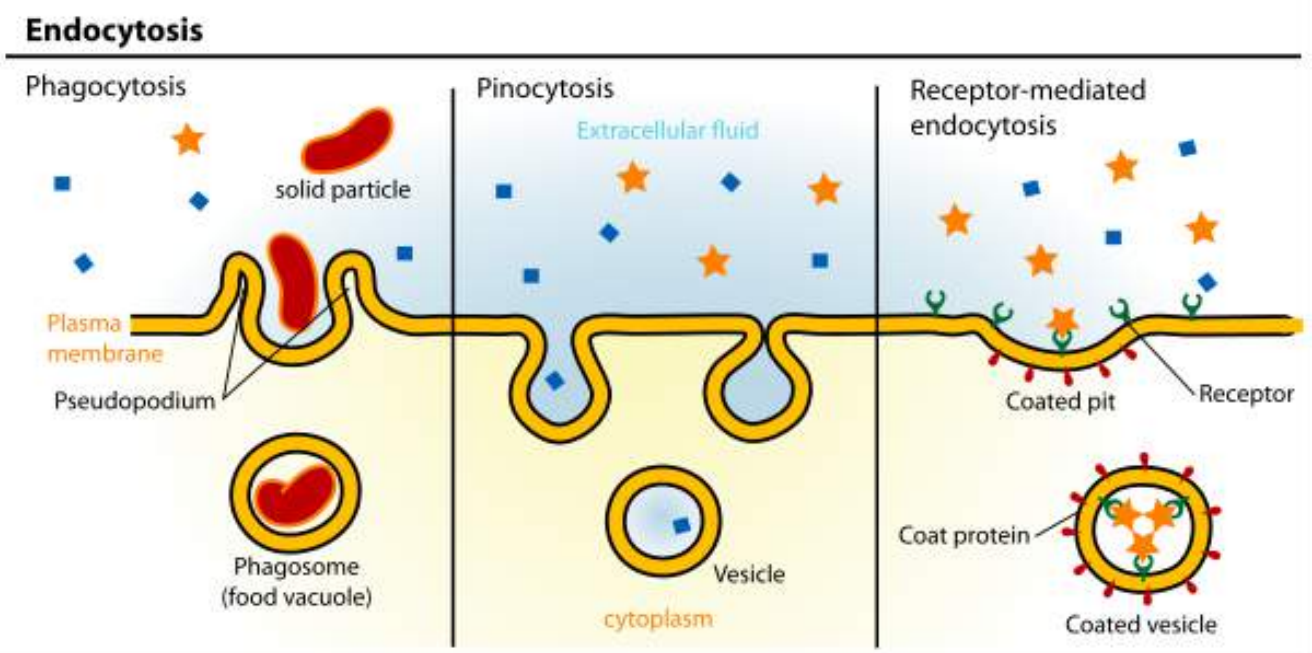
SmartNanoTox
Smart Tools for Gauging Nano Hazards

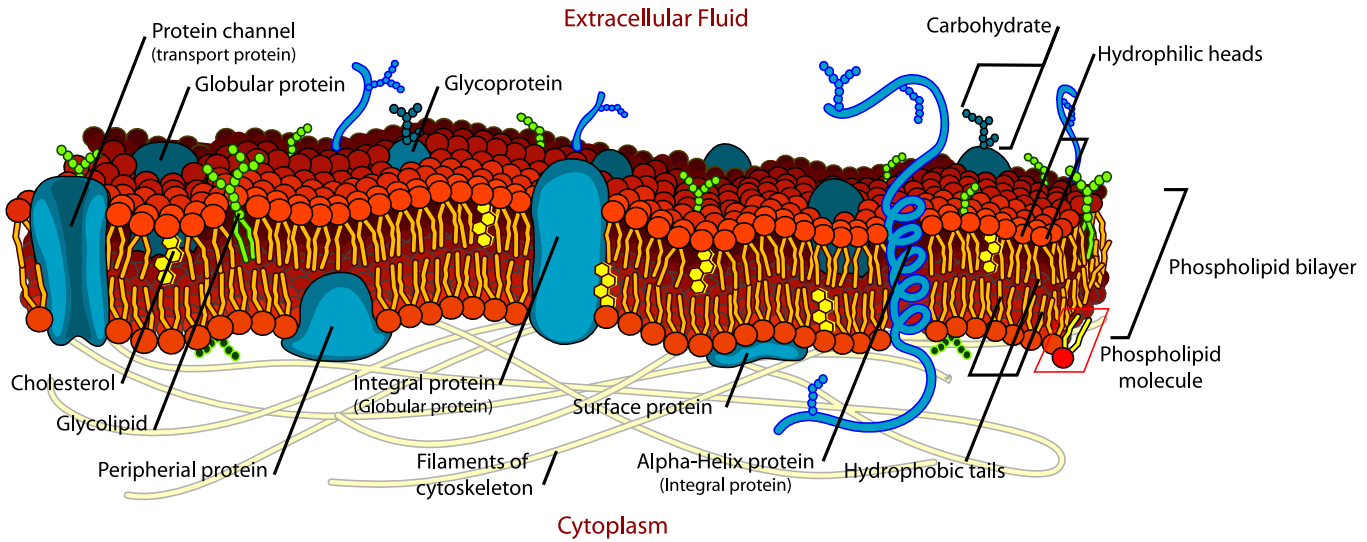


Getting stuff in….

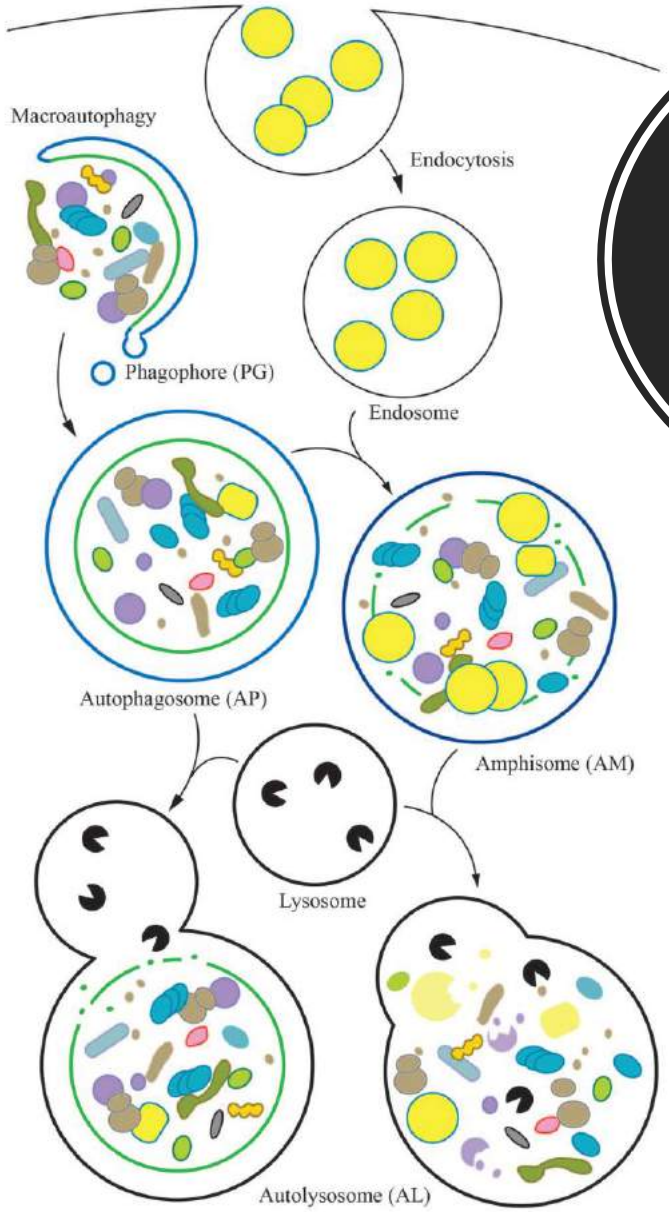
- Conventionally endocytosis is a cellular process in which substances are actively brought into the cell.
- BUT there are alternative routes: We assume passive diffusion across the membrane **and that once inside nanomaterials will be taken up by the autophagic pathway – the cell clearance mechanism**

Chem. Soc. Rev., 2017, 46, 4218





The (auto)lysosome is the primary organelle of the endocytic pathway



Nanomaterials enter the endocytic pathway

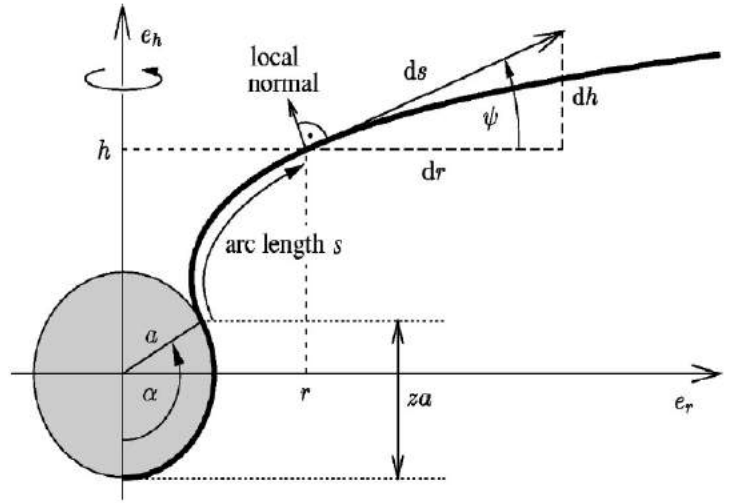
- High acidity could remove NP coronas

How to build a tractable model?

A tractable model: KIE is the penetration of the plasma membrane

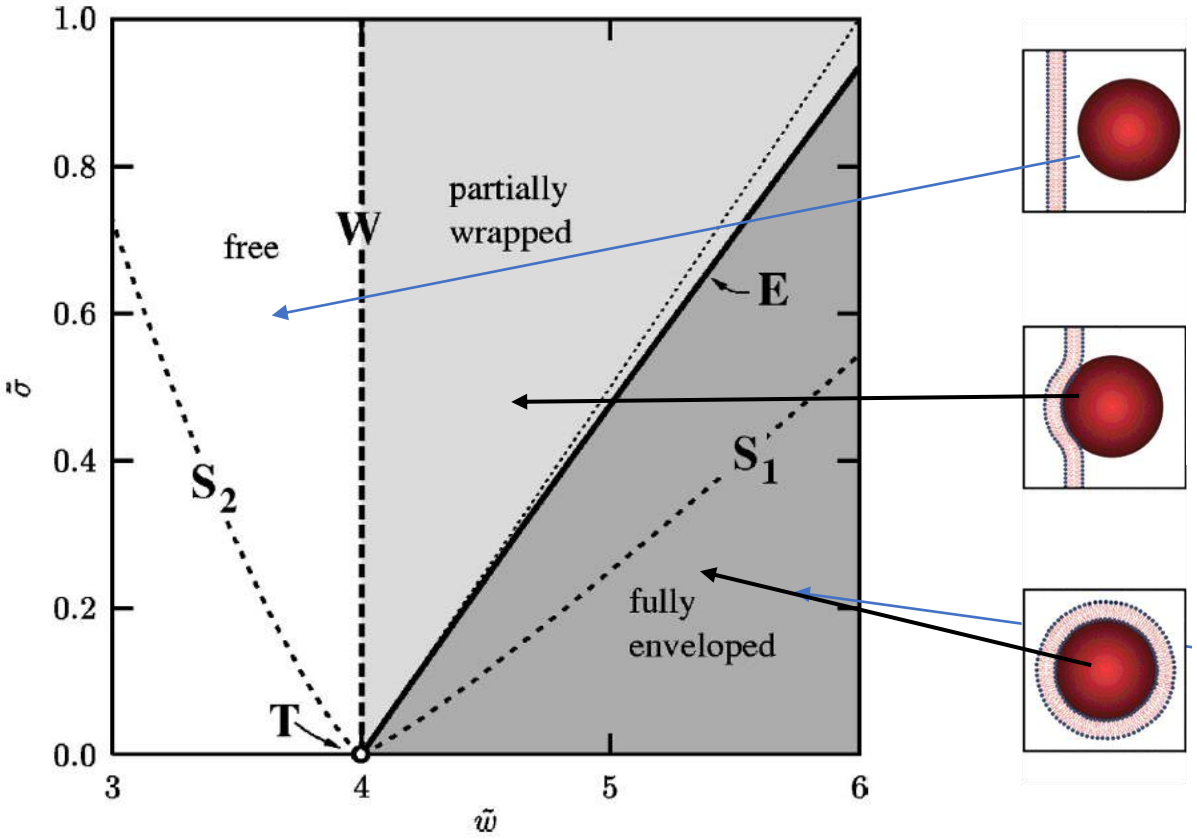
Base model is Elastic Theory: All the complication of the real plasma membrane turned into a 2d infinitesimally thin elastic sheet
The properties of which will be obtained from simulation of pure lipid membranes interacting with planar nanomaterial surfaces: we use a liposome/NP model of the plasma membrane

Three parameter theory



$$\tilde{E} = -(\tilde{w}-4)z + \tilde{\sigma}z^2 + \tilde{E}_{\text{free}}(z,\tilde{\sigma}),$$

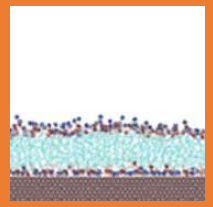
$$\tilde{E} := \frac{E}{\pi\kappa}, \quad \tilde{w} := \frac{2wa^2}{\kappa}, \quad \tilde{\sigma} := \frac{\sigma a^2}{\kappa} = \left(\frac{a}{\lambda}\right)^2,$$



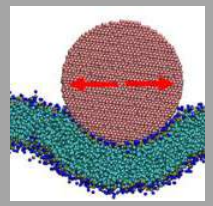
Predict wrapping (membrane disruption) if we know adhesion strength w or Kw (for planar surface)

Elastic theory has the potential for fast screening of nanoparticles for toxicity using the inflammation pathway

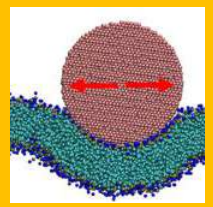
Major Challenges



No data for adhesion strength w – find from simulation?



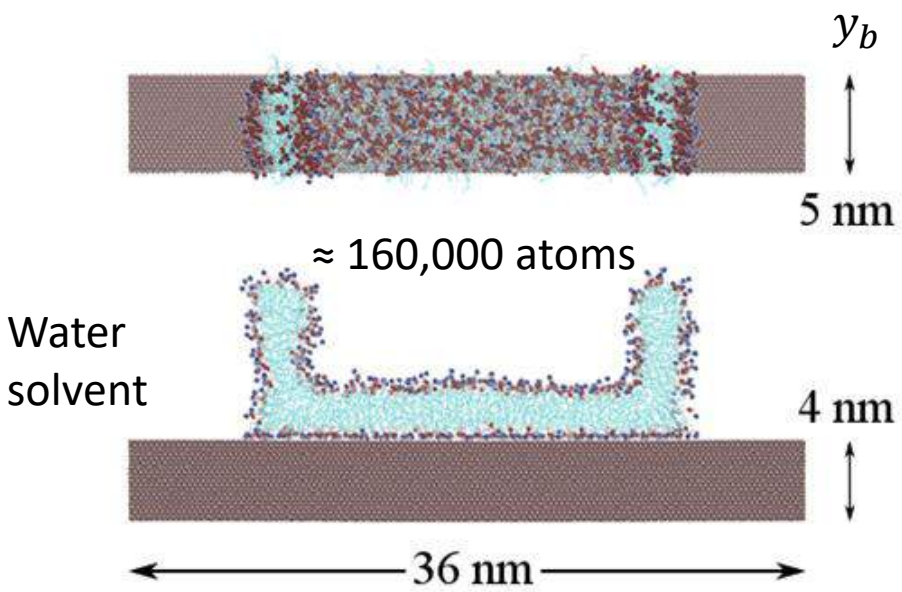
Assumes infinitesimally thin membrane: for nanoparticles membrane thickness (4nm) ~ NP radius

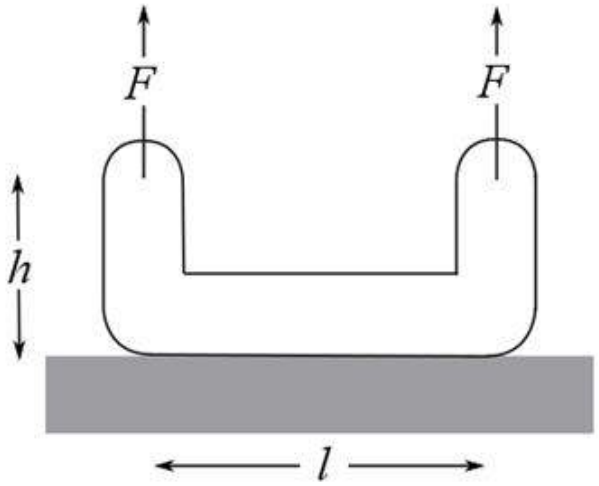


Assumes w, σ, κ independent of curvature

Atomistic molecular dynamics for *planar* interface k_w =W

The ribbon method: a novel way to predict free energies of adsorption





The 18 lipids at each end of the ribbon form 2 pull groups applying a force F. The pull groups are recalculated periodically (0.5 ns) to allow migration of lipids between leaflets: F resists the formation of new adsorbed bilayer

Adsorption free energy/adhesion strength ($mN.m^{-1}$)

$$k_w = \Delta G_\infty = F\frac{dh}{dA} = -F\frac{a_{fm(F)}}{y_b a_{slb}}$$

We need to know the lipid specific area in the adsorbed bilayer and in the free membrane

M. Schneemilch, N. Quirke, Chemical Physics Letters 664 (2016) 199–204

Formula	Polymorph	M. index	% ion	k_w mN.m^{-1}	ΔH_{adh} mN.m^{-1}	TΔS mN.m^{-1}	ApL nm	sep nm	thick nm
Au	Fcc	111	-	-42(2)	-118(7)	-76(9)	0.84	0.36	2.83
Au[a]	Fcc	111	-	-40.7(4)	-77.8(6)	-37(1)	0.85	0.47	2.83
Au[b]	Fcc	111	-	-21.6(6)	-93(10)	-71(11)	0.66	0.77	3.49
SiO_2	quartz	100	0	-2.1(4)	2.1(12)	4.2(16)	0.65	0.76	3.15
SiO_2	cristobalite	$10\bar{1}$	0	-3.2(4)	-2.8(17)	0.4(21)	0.67	0.54	3.08
SiO_2	amorphous	-	0	-7.5(5)	-10.0(11)	-2.5(16)	0.71	0.52	2.98
SiO_2[c]	cristobalite	$10\bar{1}$	-	-2.0(3)	-3.4(21)	-1.4(24)	0.64	1.01	3.18
SiO_2	quartz	100	9	-2.5(4)	3.0(16)	5.5(20)	0.65	0.76	3.17
SiO_2	cristobalite	$10\bar{1}$	9	-4.4(7)	10.7(13)	15.1(19)	0.65	0.69	3.18
SiO_2	amorphous	-	9	-5.3(4)	-4.4(11)	0.8(15)	0.66	0.93	3.14
SiO_2	cristobalite	$10\bar{1}$	18	-2.4(1)	2.8(20)	5.2(2.1)	0.62	0.74	3.23
SiO_2	amorphous	-	18	-4.3(2)	4.4(14)	8.7(16)	0.63	0.88	3.21
TiO_2	Rutile	110	-	-1.8(2)	1.6(10)	3.5(12)	0.62	1.18	3.20
TiO_2	Rutile	100	-	-3.6(5)	2.6(11)	6.1(16)	0.65	0.89	3.03
TiO_2	Rutile	101	-	-4.0(5)	1.4(14)	5.4(20)	0.66	0.85	3.04
TiO_2	Rutile	001	0	-0.3(1)	1.9(13)	2.2(15)	0.62	0.96	3.16
TiO_2	Anatase	101	-	-1.0(4)	-6.5(14)	-5.5(18)	0.64	1.00	3.08
TiO_2	Anatase	100	-	-1.1(7)	-3.7(12)	-2.6(18)	0.63	1.53	3.22
TiO_2	Anatase	001	0	-0.6(2)	-1.7(15)	-1.2(16)	0.64	1.10	3.14
TiO_2	Anatase	110	-	-4.0(3)	-13.8(23)	-9.8(25)	0.64	0.81	3.12

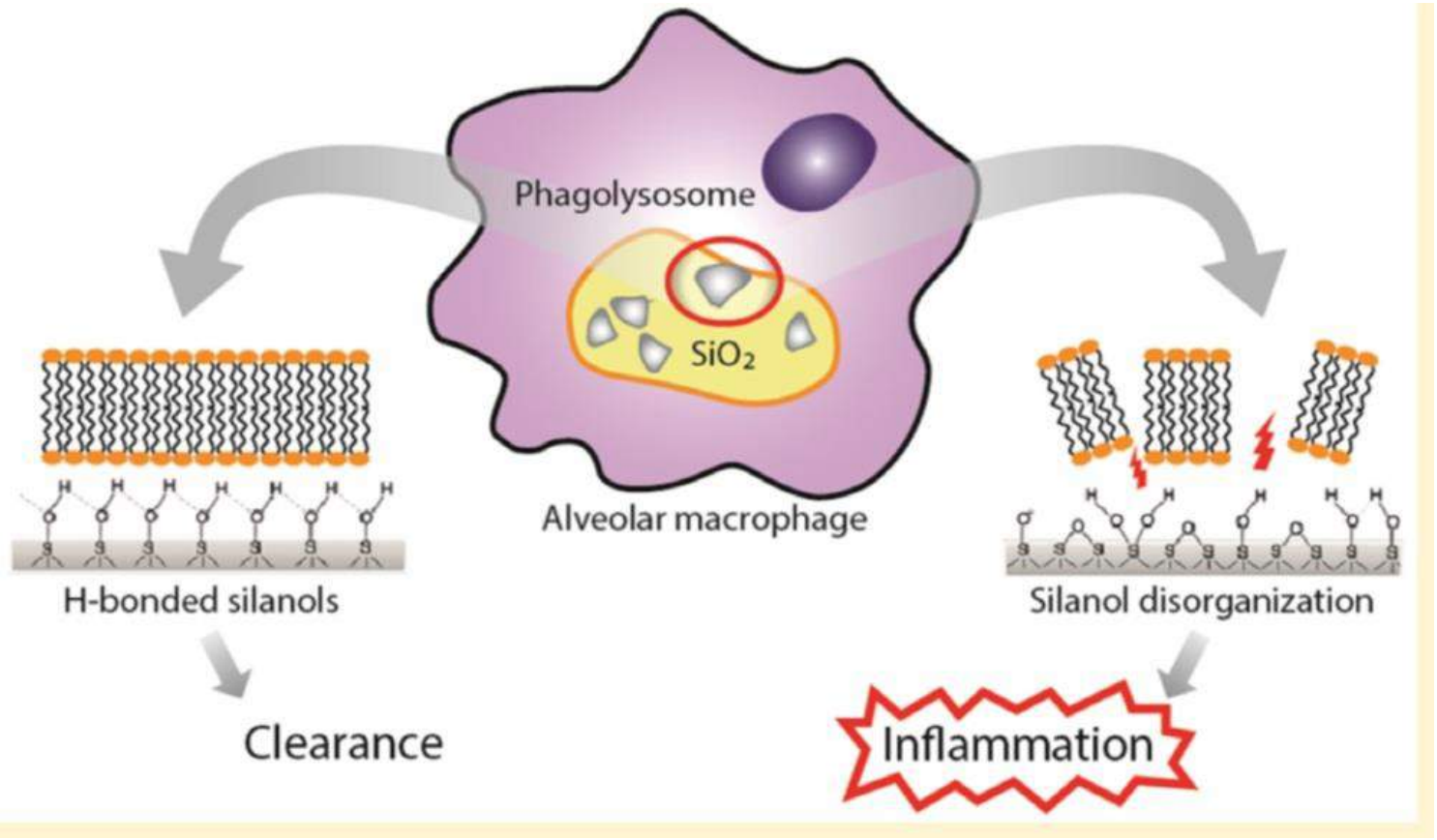
Table 1. Results from atomistic adhesion simulations using the ribbon geometry. Numbers in brackets denote the uncertainty expressed in terms of the least significant digit. Where no error is recorded the uncertainty is smaller than the least significant digit. [a] bilayer mixture with no cholesterol. [b] bilayer mixture with cholesterol. [c] Q^4 surface silanols have been condensed to form siloxane bridges.

DMPC Bilayer on Au, SiO_2, TiO_2 at 310K

*Note mixed lipid layers and effect of Chol**

Gold has highest Kw , followed by neutral amorphous silica, titania has weaker adsorption

*we have also simulated a mixed membrane containing 50% POPC, 25% DOPE, 15% BMP, and 10% SM (18) with and without cholesterol (in equal molar quantities to POPC when present)



Consistent with silica toxicology: disordered surfaces associated with inflammation

- Our simulations suggest that disordered silica surfaces have a much stronger adsorption free energy w than ordered surfaces
- Kw = -7.5

C Pavan, and B Fubini, ' Unveiling the variability of 'quartz hazard' in light of recent toxicological findings',Tox,30, 469, (2017)

Coarse graining the atomistic membrane/nanomaterial

From 118 site Slipids force field to 10 bead implicit solvent

Magic software

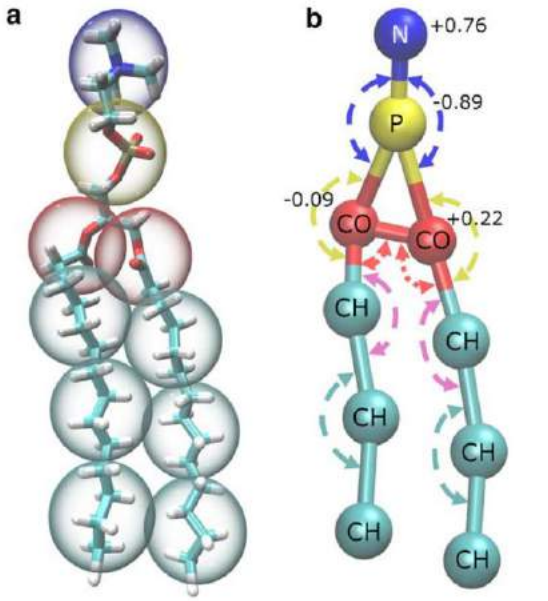
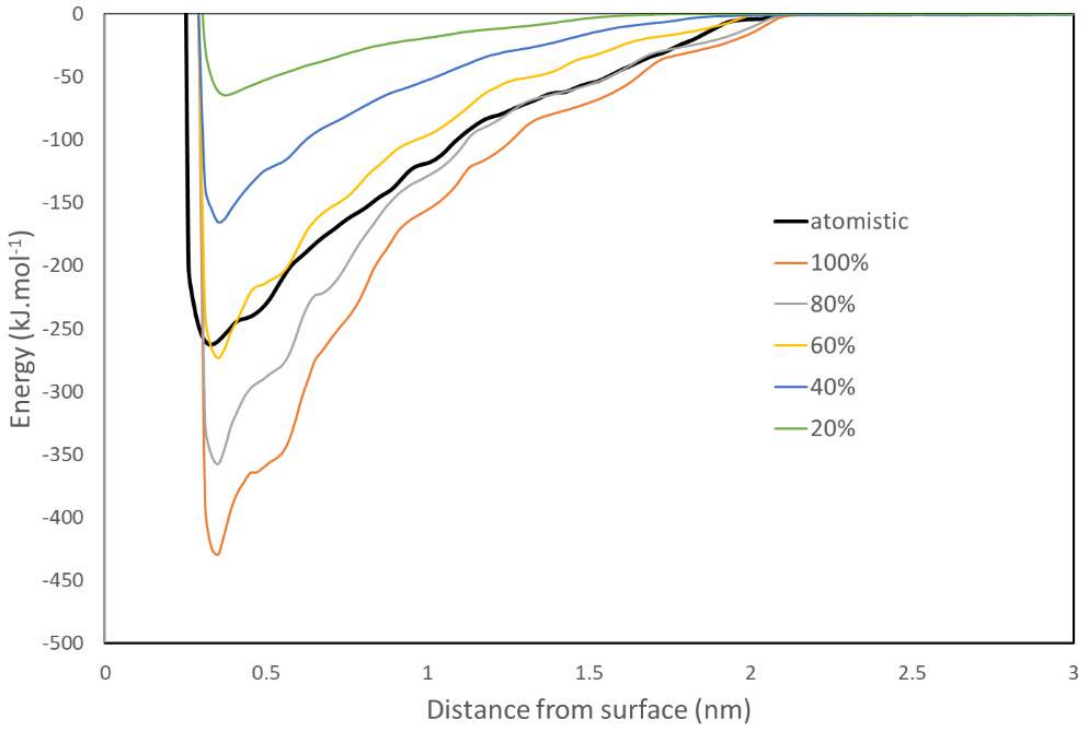
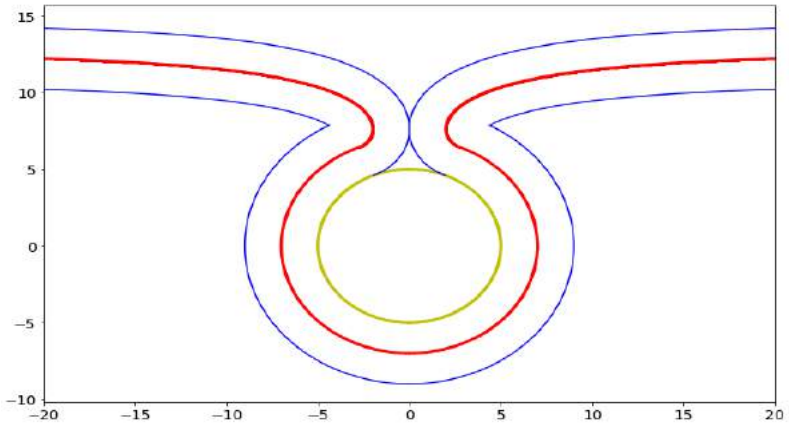


Figure 1. (a) Coarse-grained representation of a DMPC molecule. Blue bead—choline, yellow bead—phosphate group, red beads—ester groups, and cyan beads—hydrocarbon tails. Beads of the same color belong to same bead type. (b) Intramolecular bonds introduced in the CG model. Solid lines denote covalent bonds, dashed arrows denote angle bending interactions. Bonds of the same color are assumed to be identical. Positive and negative numbers indicate partial charges of the head-beads. [Color figure can be viewed in the online issue, which is available at wileyonlinelibrary.com.]

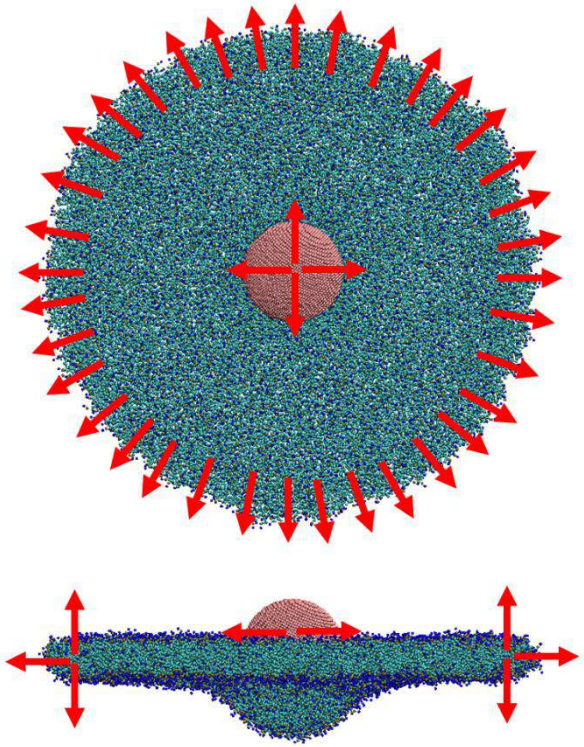
X-potential : PMF for CG lipid interacting with a gold surface in water: Scale CG ε in Steele 10-4-3 potential: 60% = AMD



Note need to modify elastic theory (MET)

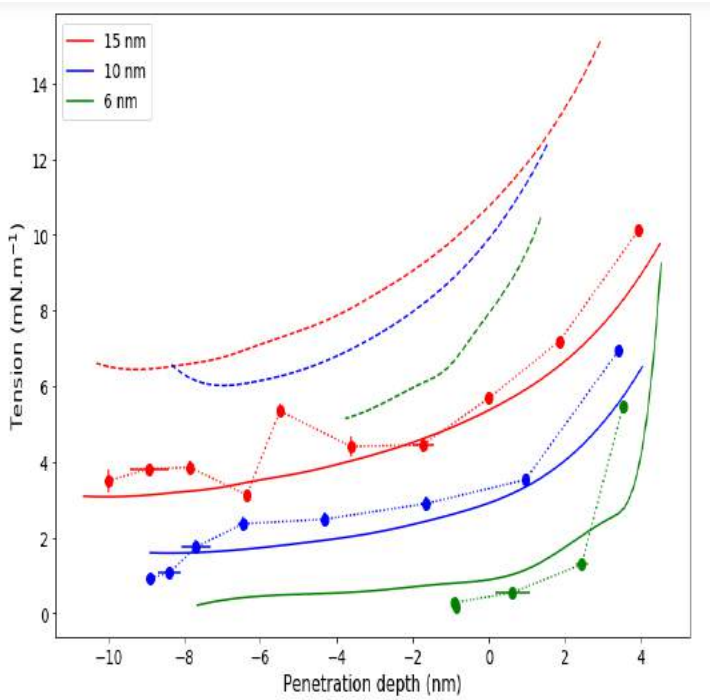


CG MD



k_w = -10 mN.m^{-1}

CGMD. v. ET and MET



Membrane tension as a function of NP penetration

From the CG simulations and MET we are able to predict minimum diameters for complete wrapping/membrane penetration

$$w_{SLB}^{s} = -k_w + \frac{m}{a} - \frac{2\kappa_{SLB}}{a^2} \quad (15)$$

$$w_{SLB}^{c} = -k_w + \frac{m}{2a} - \frac{\kappa_{SLB}}{2a^2} \quad (16)$$

Critical sizes for complete wrapping using fitted w

formula	polymorph	M. index	% ion	k_w mN.m^{-1}	sphere d_{crit}^{s} nm	cylinder d_{crit}^{c} nm
Au	Fcc	111	-	-42(2)	0	0
SiO_2	quartz	100	0	-2.1(4)	41.2	18.6
SiO_2	cristabolite	$10\bar{1}$	0	-3.2(4)	25.1	10.6
SiO_2	amorphous	-	0	-7.5(5)	7.5	1.8
SiO_2*	cristabolite	$10\bar{1}$	-	-2.0(3)	43.5	19.8
SiO_2	quartz	100	9	-2.5(4)	33.7	14.9
SiO_2	cristabolite	$10\bar{1}$	9	-4.4(7)	16.8	6.4
SiO_2	amorphous	-	9	-5.3(4)	13.0	4.5
SiO_2	cristabolite	$10\bar{1}$	18	-2.4(1)	35.4	15.7
SiO_2	amorphous	-	18	-4.3(2)	17.3	6.6
TiO_2	Rutile	Sphere	-	-2.48	34.0	-
TiO_2	Anatase	Sphere	-	-1.47	61.1	-
TiO_2	Rutile	Cylinder	-	-2.89	-	12.2
TiO_2	Anatase	Cylinder	-	-1.11	-	39.3

Table 1. Predicted critical diameters for a range of materials for both cylindrical and spherical particles.

$d_{crit} = 2*r$ m, κ fitting parameters

Amorphous silica binds more strongly in an acid environment: Cylinders wrap more easily

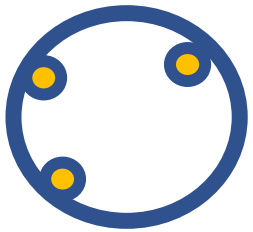
Not the only consideration!

If we consider a system of **liposomes** in a solution of particles it is possible to use these data to determine a maximum particle concentration for complete wrapping to occur (*cells may have lipid reservoirs, hence no tension change*)

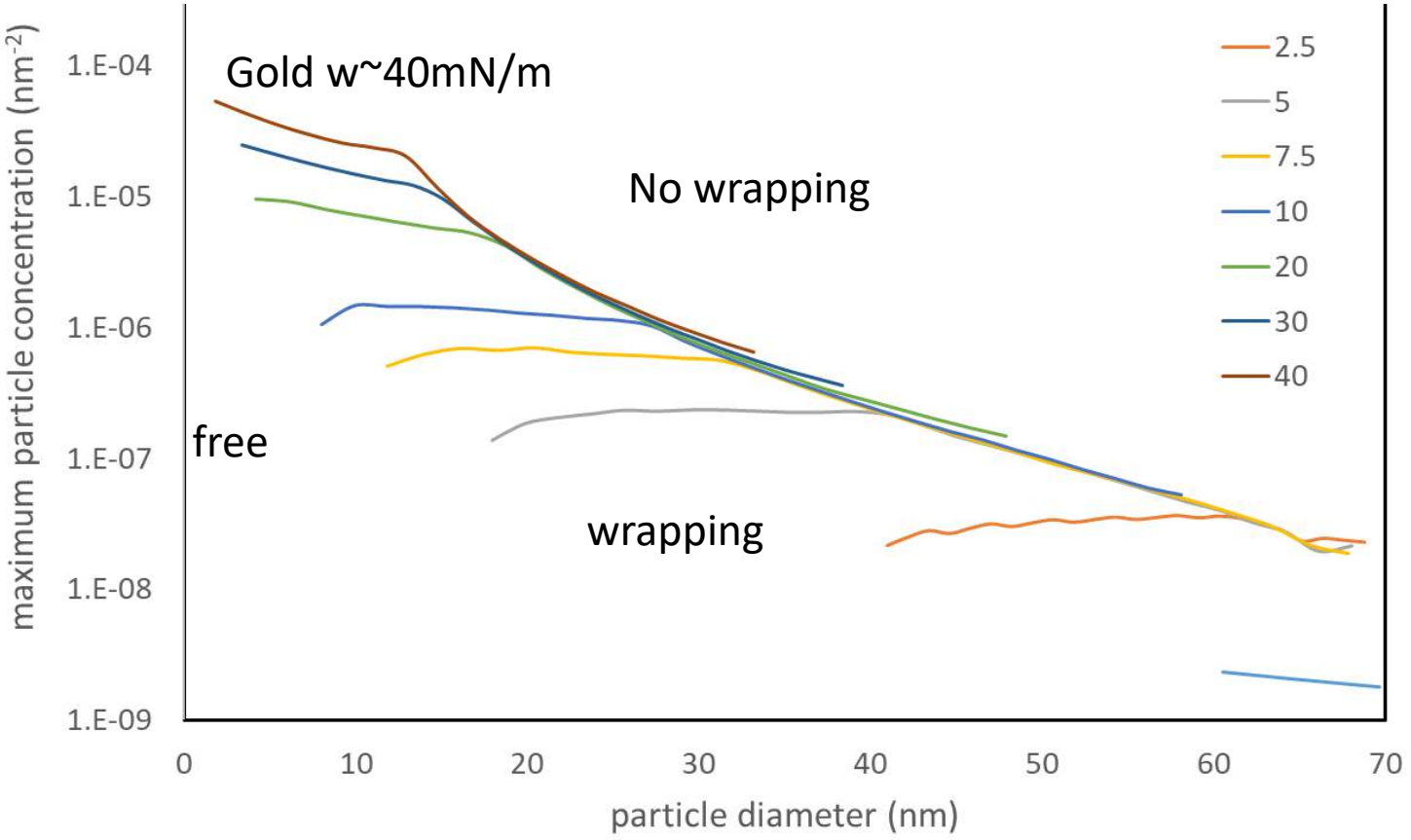
At greater particle concentrations only partial wrapping will occur as the membrane deformation induced by the particles will increase the tension beyond the maximum before complete wrapping can occur.

Particles per square nm of the liposome surface:
ApL0=tensionless membrane
Each particle consumes $4\pi a^2$ at the max wrapping tension

After this concentration only no or partial wrapping can occur



$$c_{max}^{sphere} = \frac{\left({ApL_{max}}/{ApL_0} - 1\right)}{4\pi a^2}$$



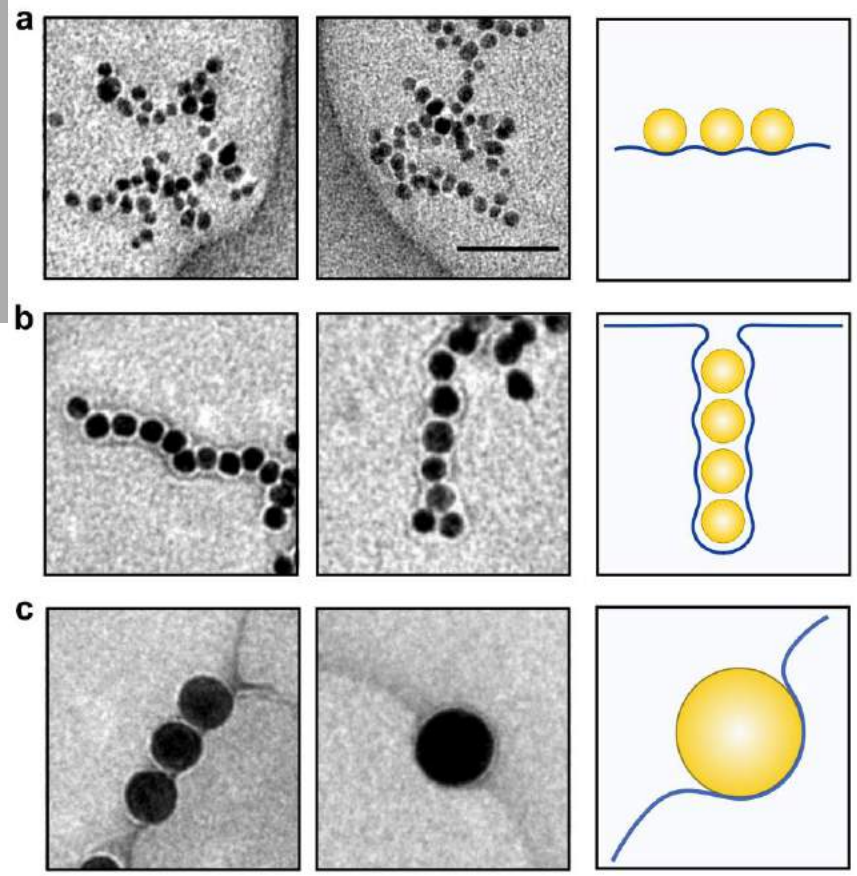
Maximum spherical particle concentration, expressed in particles per square nm of liposome bilayer, for complete wrapping over a range of values of adhesion strengths

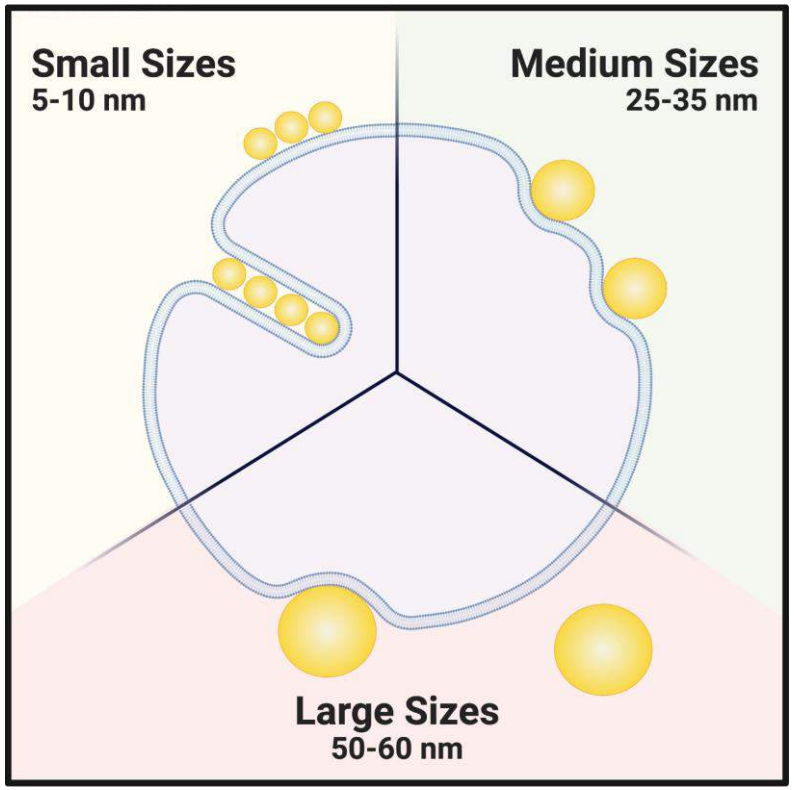
complete wrapping is halted above these NP/Liposome concentrations

For larger particles , low concentrations stop wrapping completely

Experiment: no complete wrapping for larger gold NPs

Theory
<d_{lipo} > ~200nm
d_{np} =5nm, cmax~10-4 nm2,
Complete wrapping
d_{np} 25 nm, cmax ~10-6,
no complete wrapping
Larger, no wrapping





Smaller NPs prefer to form 'cylinders'

C. Contini, J. W. Hindley, J. T. Macdonald, O. Ces, N. Quirke, 'Size matters: Size dependency of gold nanoparticles interacting with model membranes', Nature Communications Chemistry (submitted, **2019**)

SmartNanoTox
Smart Tools for Gauging Nano Hazards

- New physical data (W) for empirical correlations
- New coarse grained simulation model for pure liposomes
- First steps in extending elastic theory to nanoscale systems
- First description of concentration effects and size effects(descriptors) on NP wrapping
- For cells need to correlate with expt data

All encapsulated in a code ‘Nanolip’ to be available soon

M Schneemilch, N Quirke, ‘Predicting the fate of nanoparticles in contact with biological membranes’, in preparation