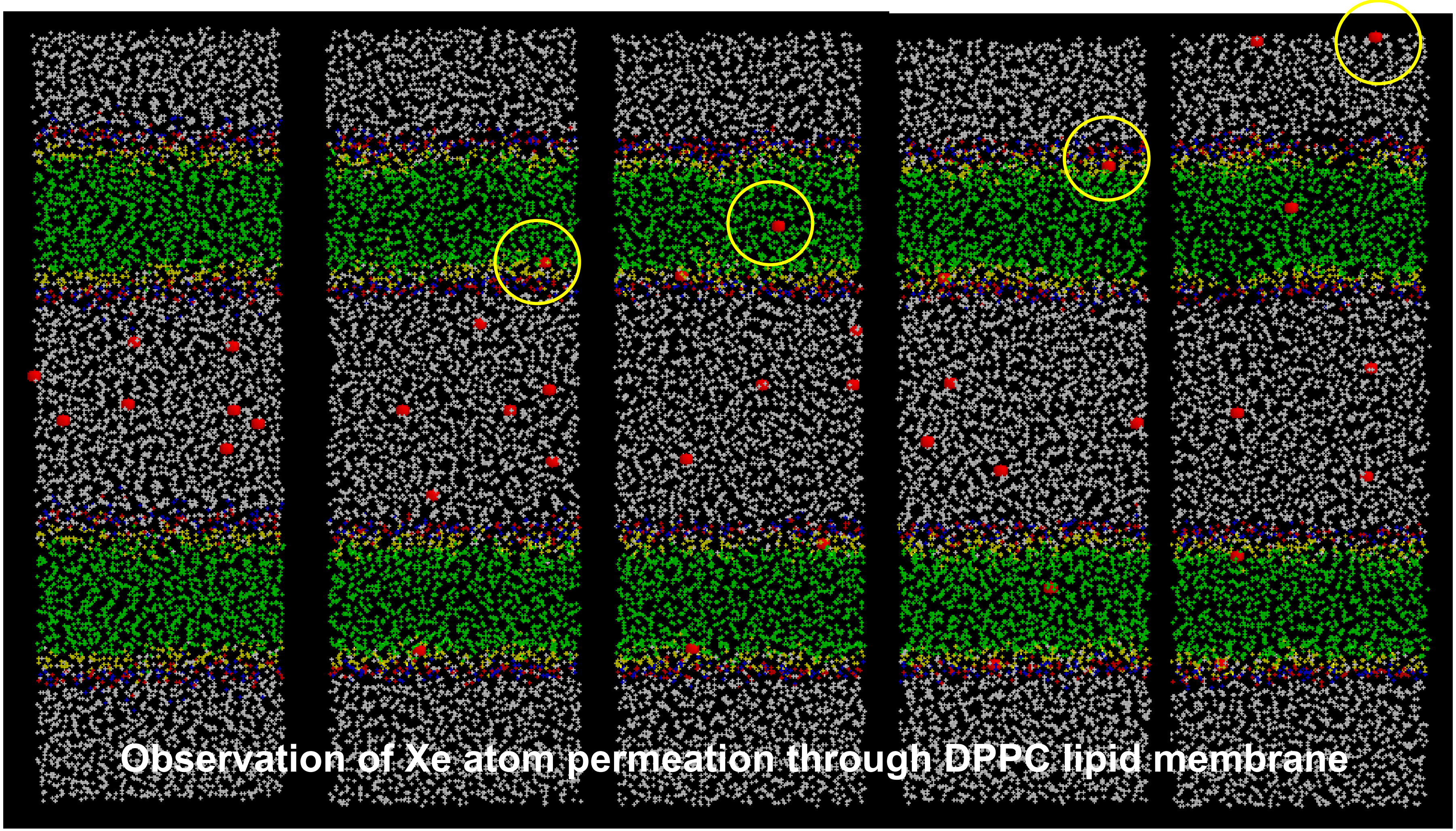
Transport of Molecules and Ions Across Model Membranes



Observation of Xe atom permeation through DPPC lipid membrane

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University of Illinois at Chicago

Molecular Dynamics studies of transport

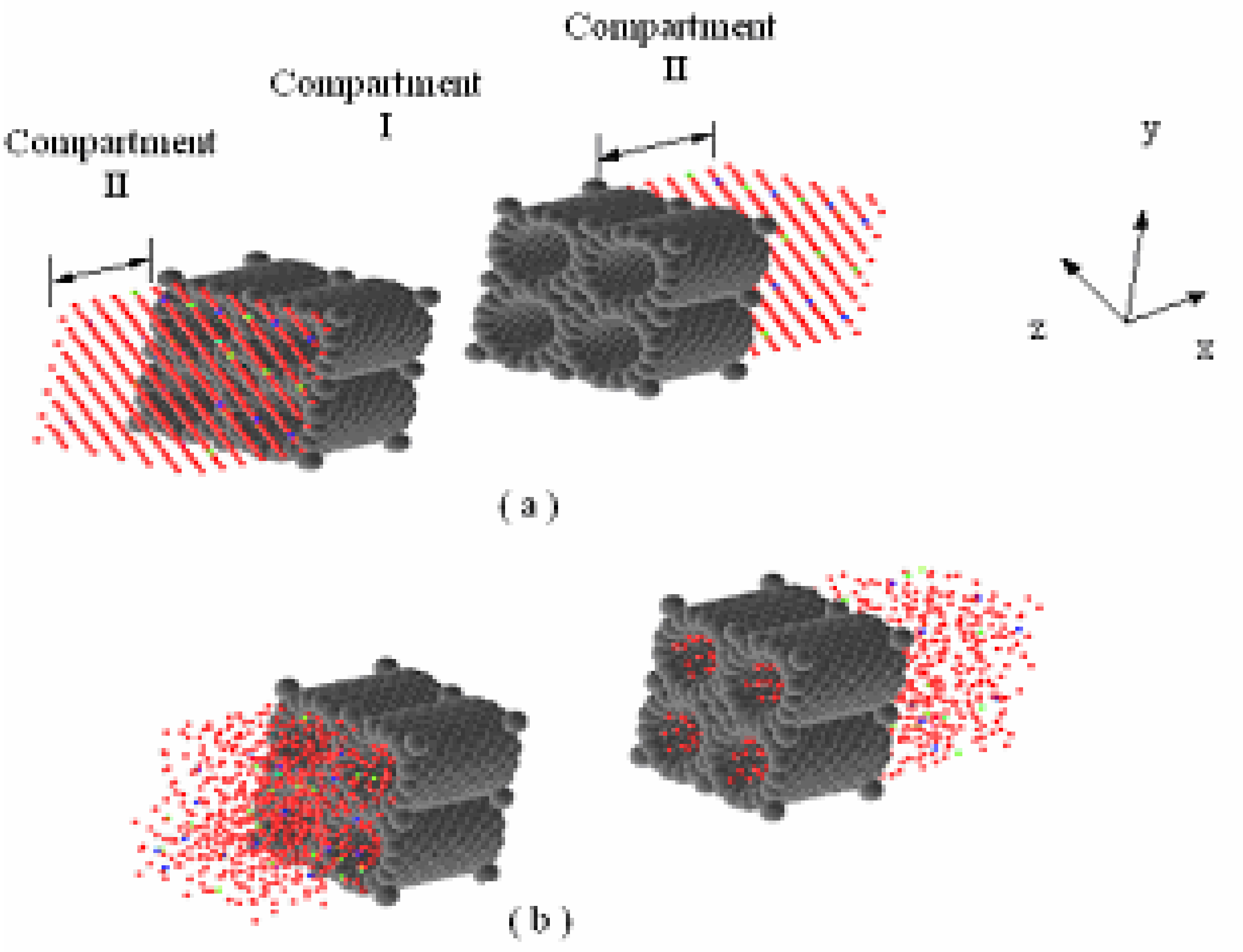
- Permeation of ions into carbon nanotubes
- Selectivity of nanochannels for K^+ vs. Na^+ ions in aqueous solution
- Permeation of ions into surface-charged tubes
- Permeation of lipid bilayer model membranes by small molecules: Xe, O_2, CO_2

PERMEATION OF IONS INTO CARBON NANOTUBES

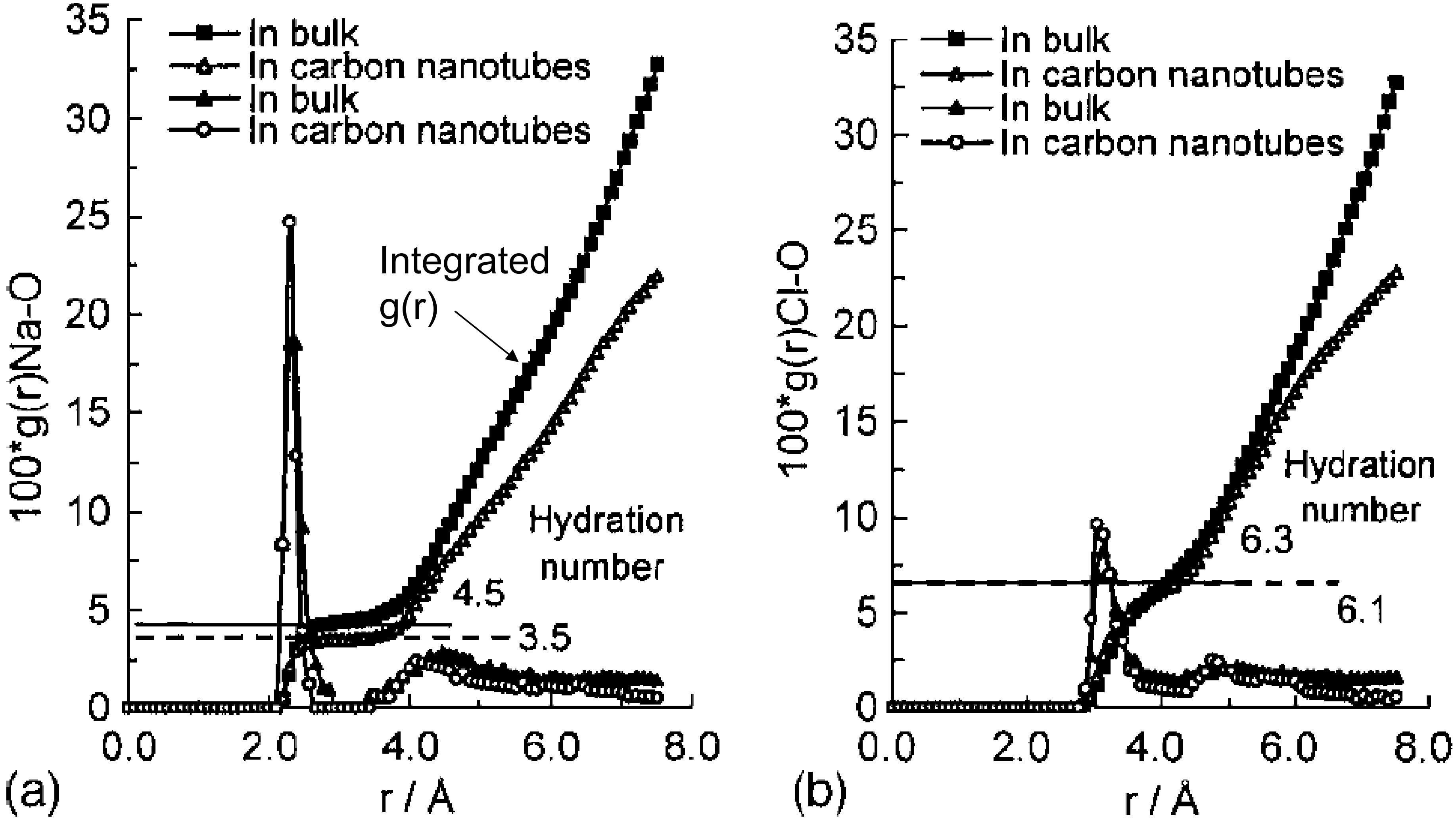
- characteristics of hydrated ions in nanotubes compared to bulk: radial distribution functions
- particle density profiles
- probability contours of H_2O and ions at the tube surface
- trajectories of positive and negative ions through the nanotubes
- effects of flexibility of the tube

with Hongmei Liu

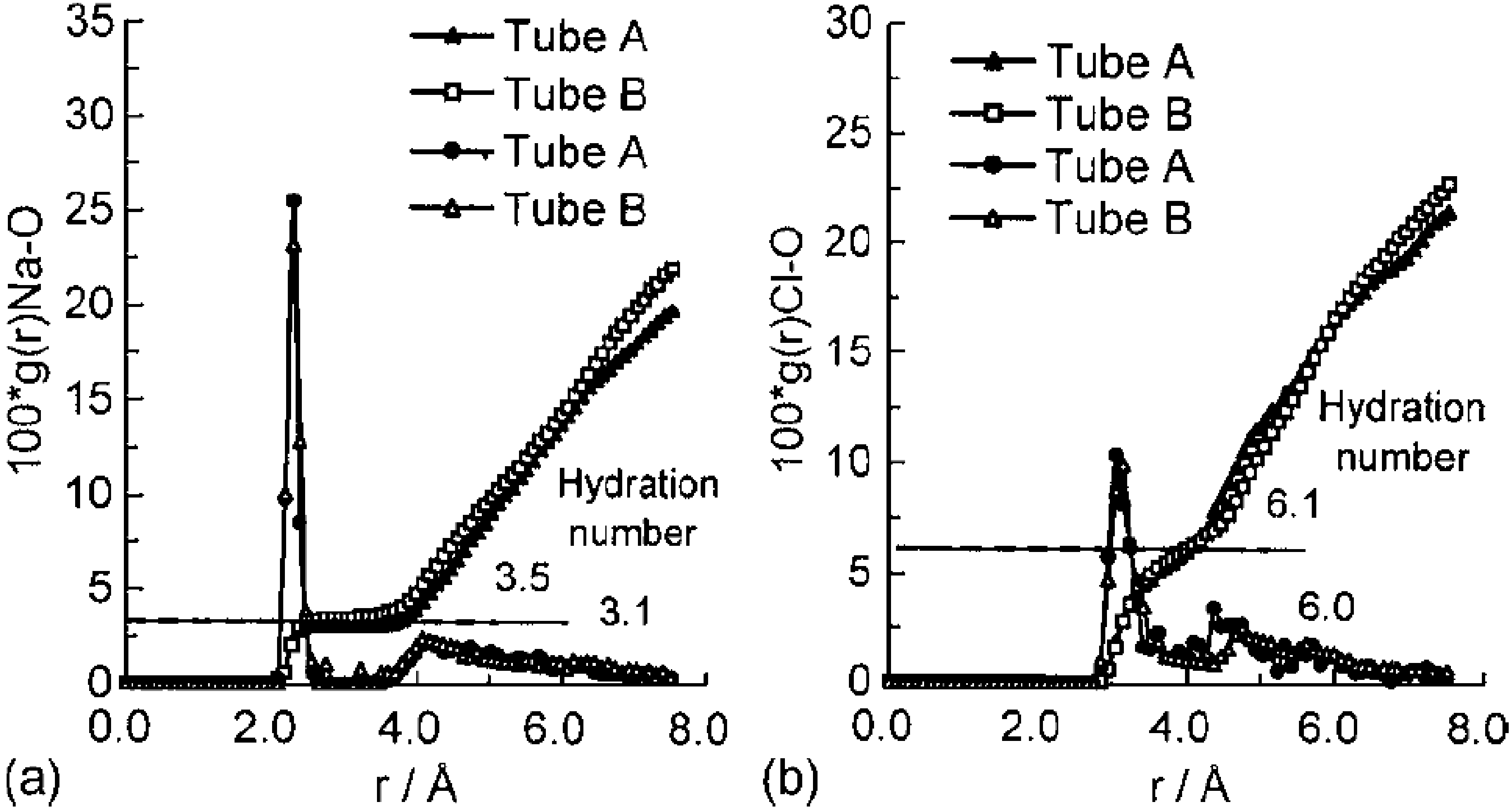
our simulation box



Radial distribution functions and hydration numbers inside nanotubes compared with bulk solution

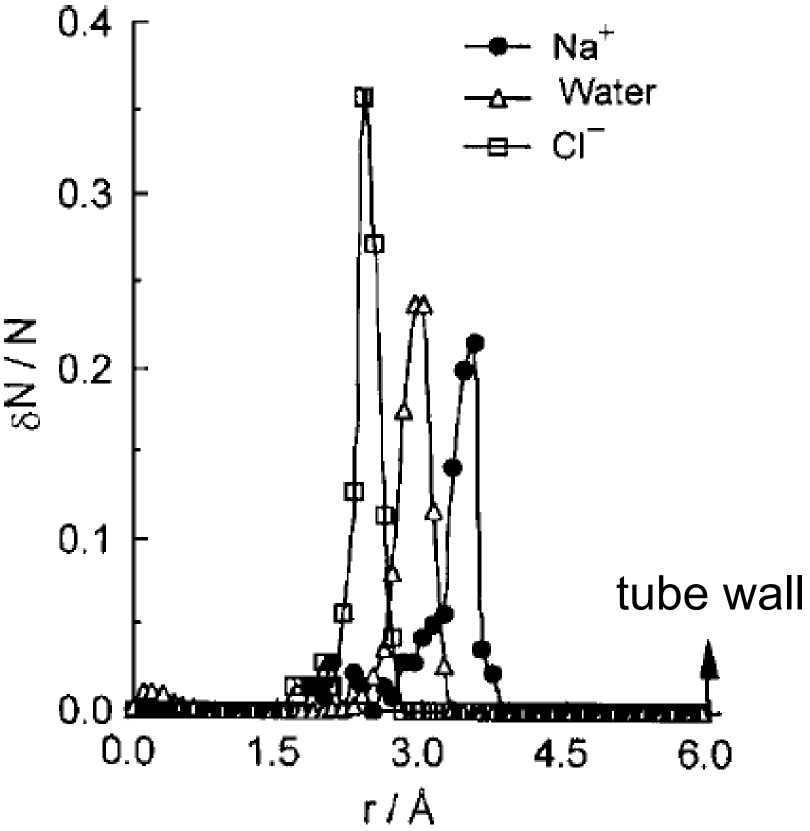


Radial distribution functions inside nanotubes



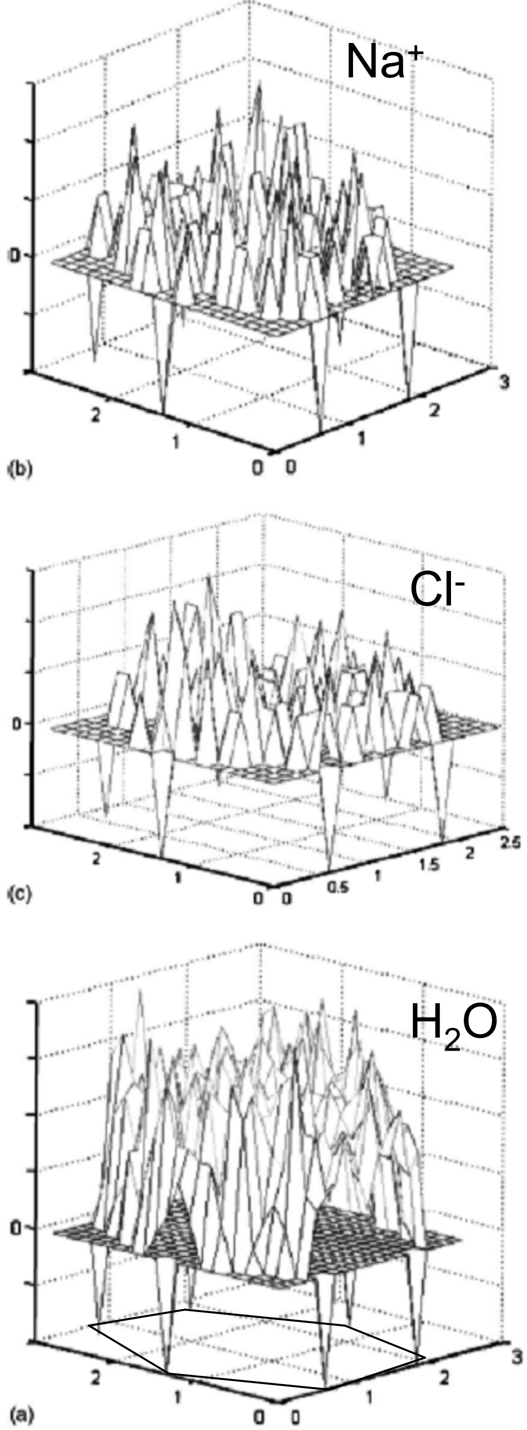
Effective diameters: tube A = 0.82 nm, tube B = 0.90 nm

Normalized radial particle distribution profiles δ*N*/*N* in a 0.90 nm tube



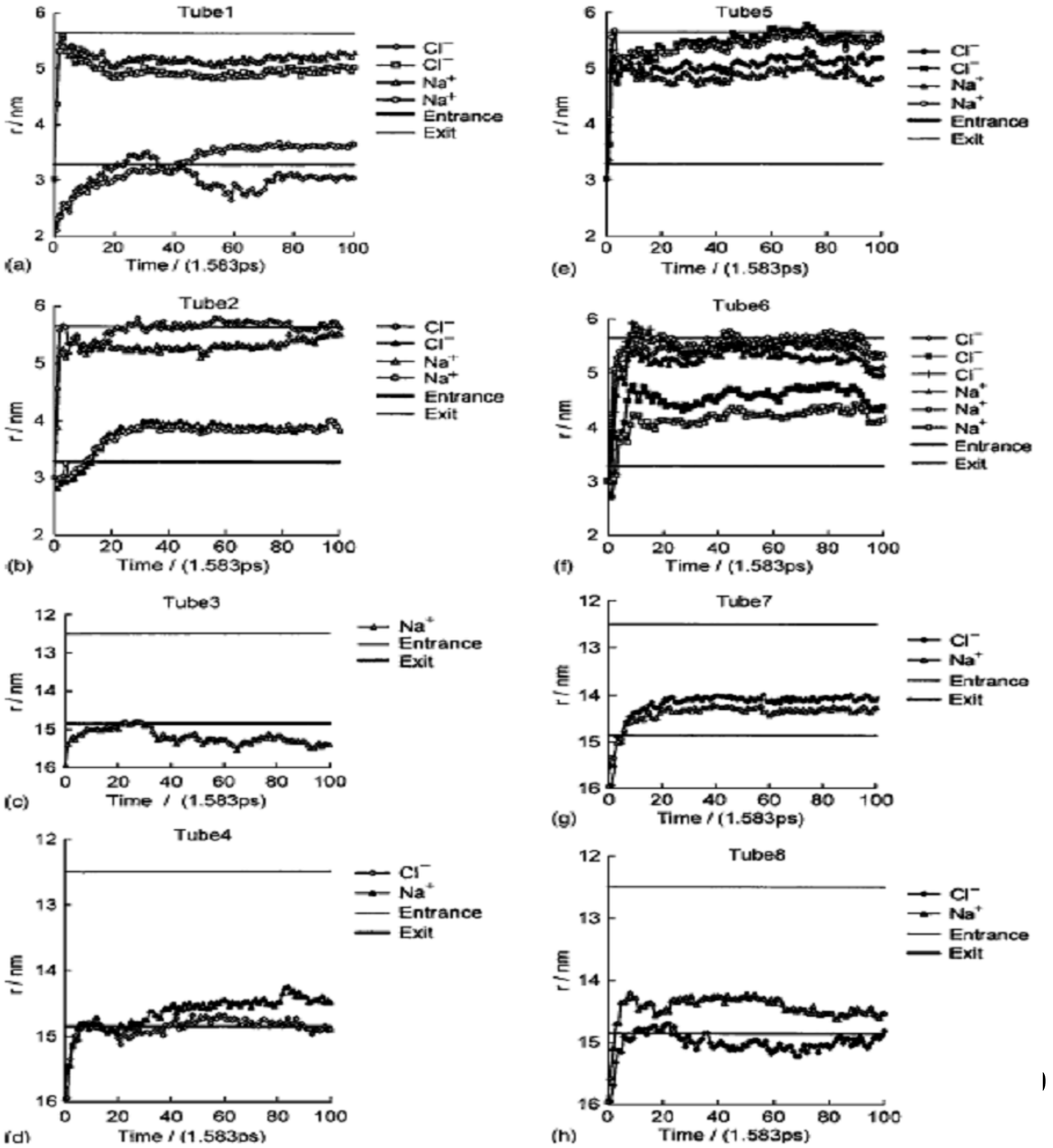
We study how these profiles change with number of ions in the same tube

Effect of surface corrugation

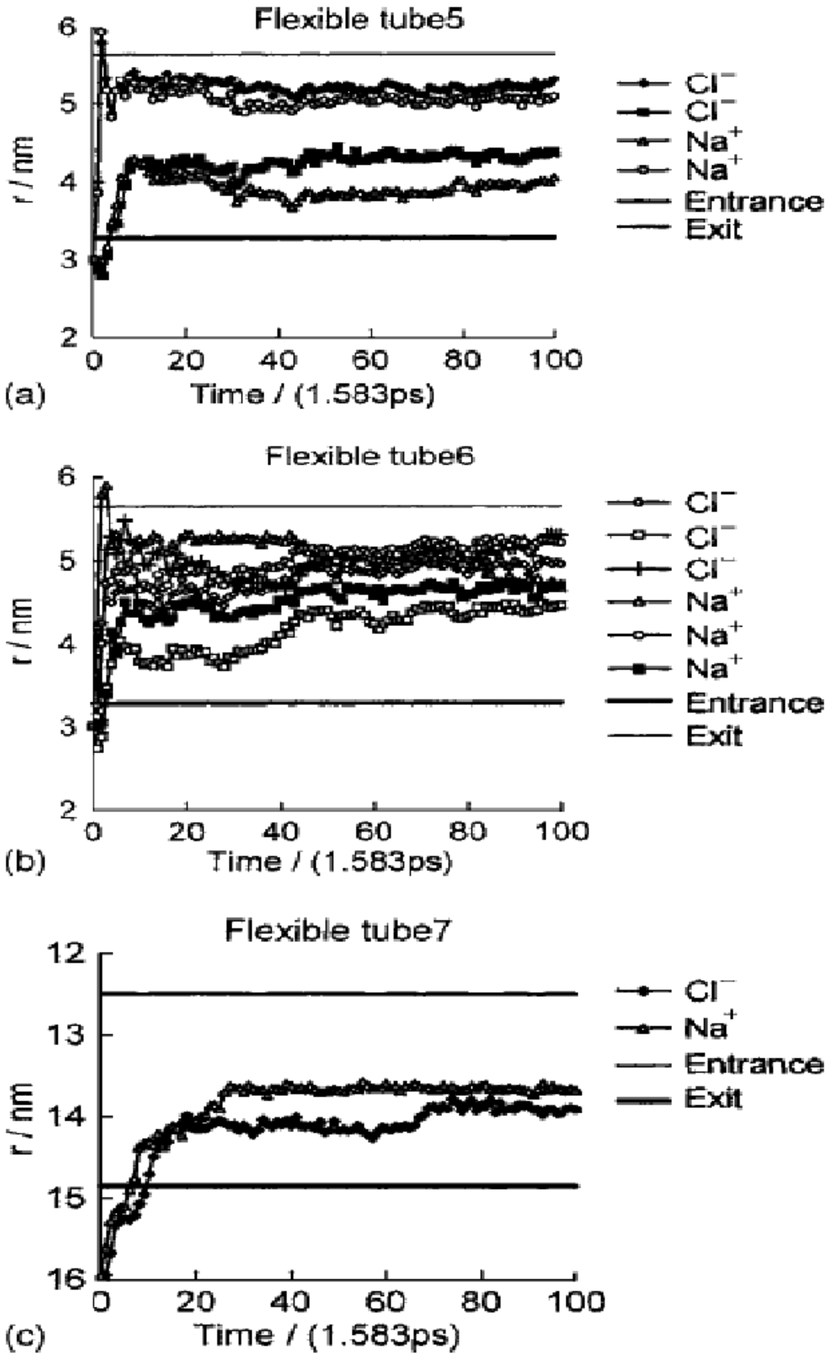


Probability density of ions or water projected on to a 6-membered ring of the carbon nanotube

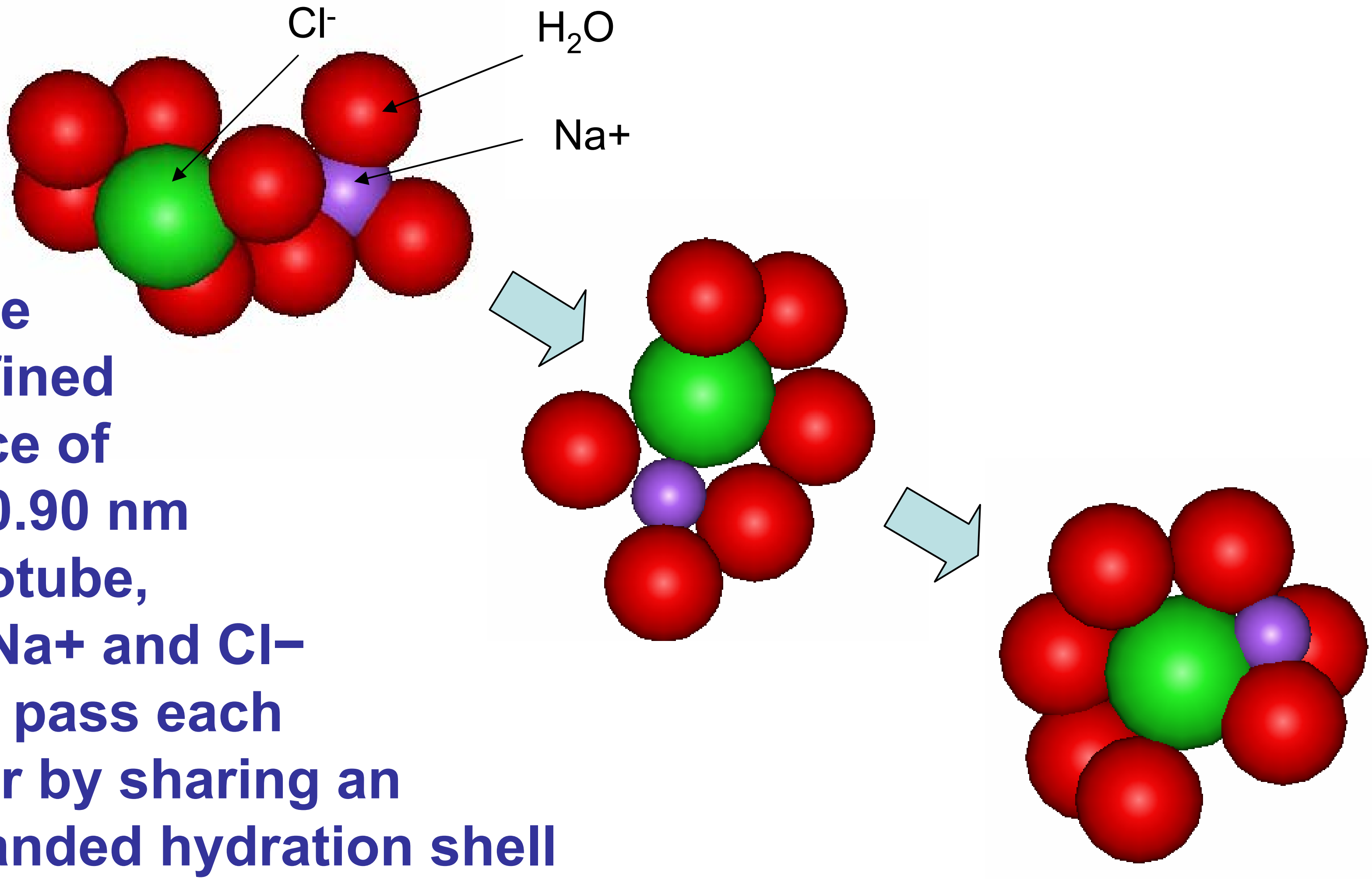
Trajectories of ions in each of 8 tubes



Trajectories of ions in more flexible tubes



Snapshots of solvation shells of Na^+ and Cl^- ions as they pass each other in a carbon nanotube



In the confined space of the 0.90 nm nanotube, the Na+ and Cl− ions pass each other by sharing an expanded hydration shell which re-forms around each ion as they move away from each other.

FINDINGS

- Tubes which have smaller diameters such as to be able to support only a column of water along the axis of the tube cannot permit ions to enter because there is no space enough to accommodate the hydration shell; the desolvation of such ions imposes a large energy as well as entropic penalty.
- Exchange times and reorientation times are longer in confinement than in the bulk solution.
- Although hydrated ions of the same charge move in single file in the tube, ions of opposite charge can pass each other in the tube.

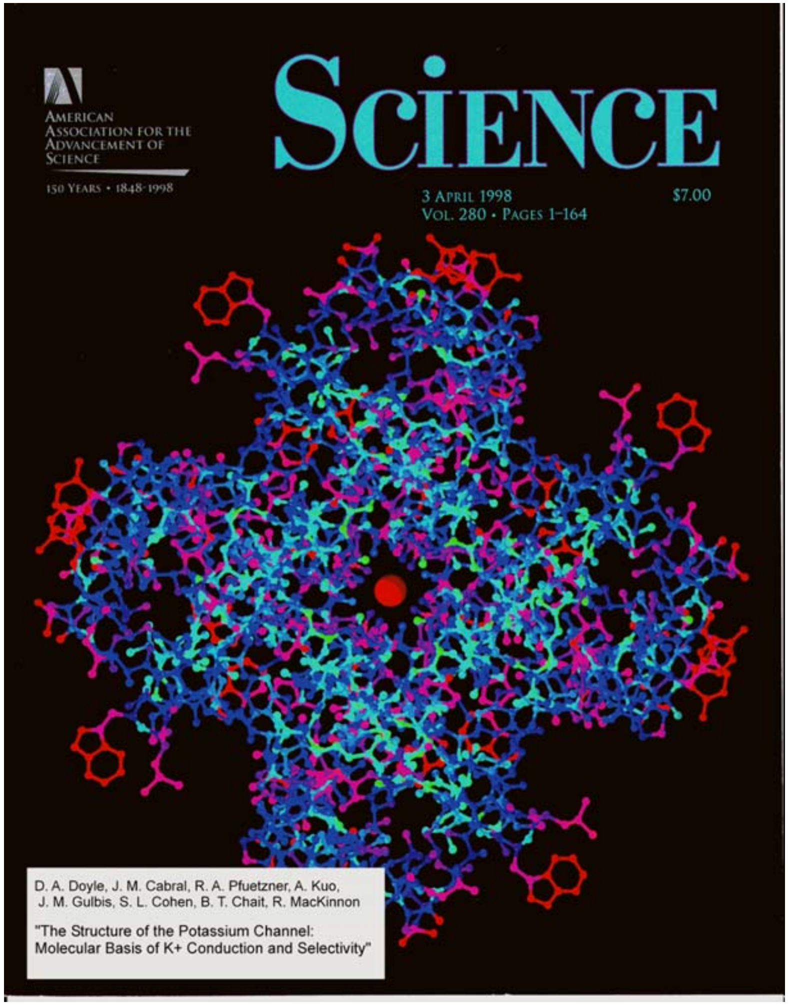
FINDINGS

- The Na^+ ion profile appears to change with tube diameter in such a way as to lie between the water profiles so as to maintain its first hydration shell.
- As the number of ions in the tube increases, the Cl^- ion radial profile does tend to spread slightly, whereas the Na+ radial profile dramatically spreads towards the center line. This is how ions of opposite charge can pass each other; the Na^+ ion can move towards the center of the tube to accommodate having a Cl^- ion in the same cross-sectional plane.

FINDINGS

- The first hydration shell of Na^+ ion can have as much as one or more H_2O molecule fewer compared to that in bulk solution
- The average hydration numbers for both ions are greater for the more flexible tube: 3.9 compared to 3.5 for Na+ and 7 compared to 6.1 for Cl−. Upon entering the more flexible tube, the ions do not lose as many waters from their first hydration shell as they do in the less flexible tube.

One of the most striking properties of potassium ion channels is their remarkable ability to conduct K$^+$ ions near the diffusion limit while simultaneously maintaining a selectivity of K$^+$ over Na$^+$ of over 1000 to 1.



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Science

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$7.00

D. A. Doyle, J. M. Cabral, R. A. Pfuetzner, A. Kuo,
J. M. Gulbis, S. L. Cohen, B. T. Chait, R. MacKinnon

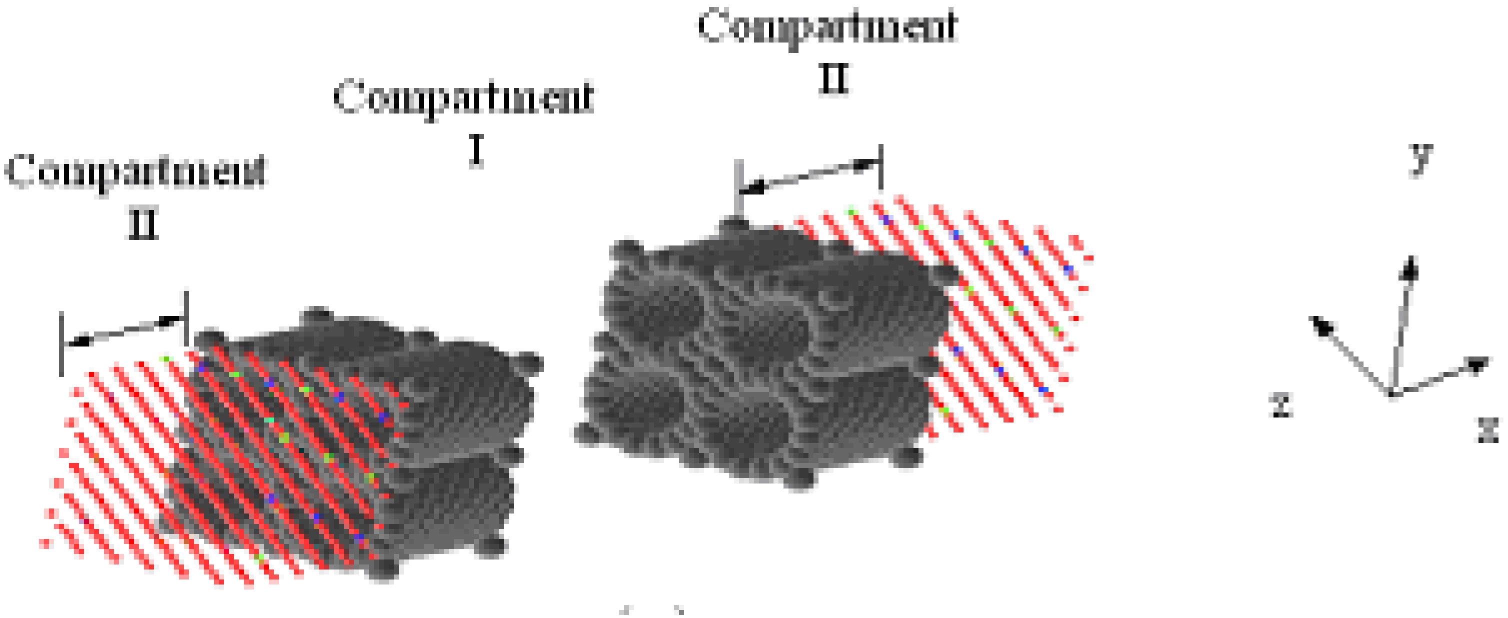
"The Structure of the Potassium Channel:
Molecular Basis of K+ Conduction and Selectivity"

Selectivity of nanochannels for K^+ vs. Na^+ ions in aqueous solution

- Does the selectivity of a nanochannel for a specific ion require specific coordination sites (selectivity filters) or can the channel diameter itself impose selectivity?

Goal of this work

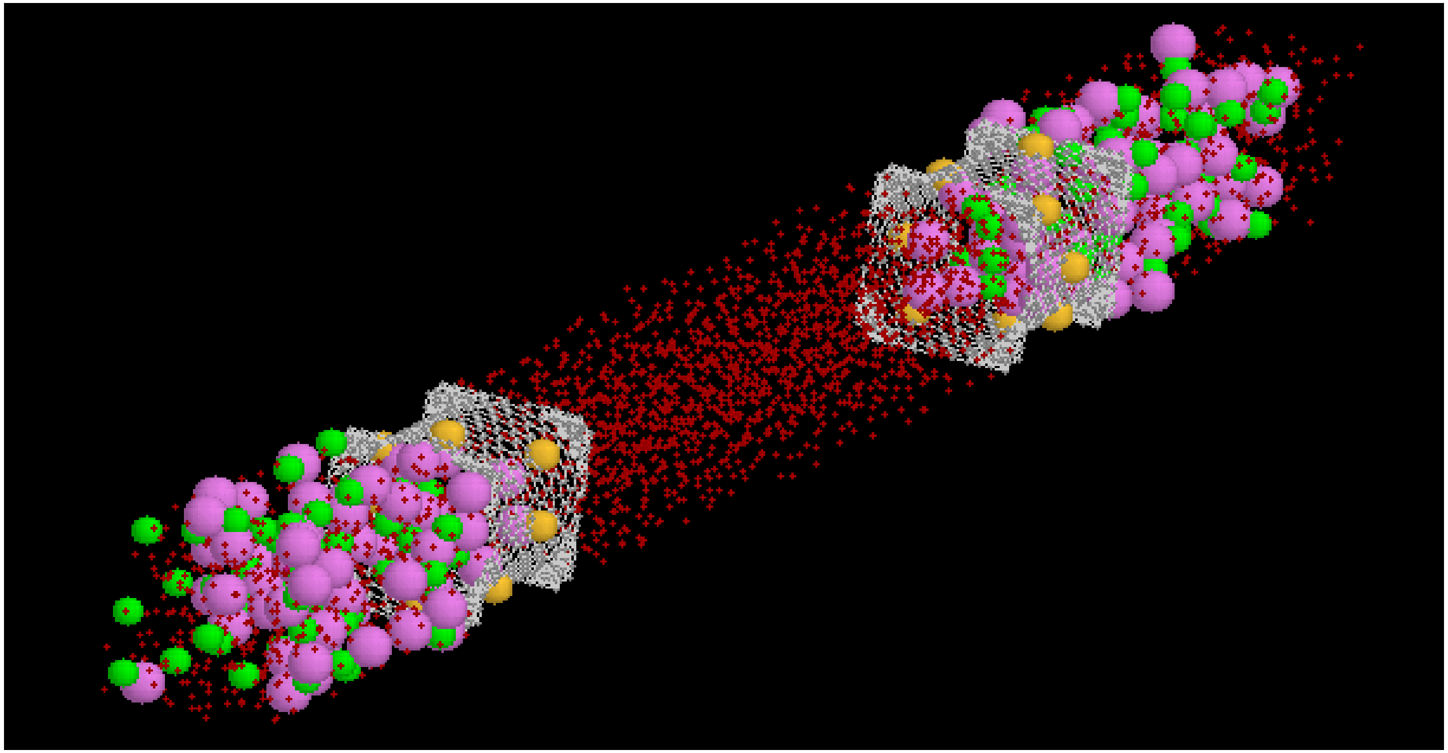
- To investigate relative selectivities of uniform atomistic cylindrical nanochannels to Na^+ and K^+ ions, in order to discover a general molecular mechanism for ion selectivity by nanochannels, a mechanism which does not require the presence of dynamic coordinating chemical groups at particular sites, the so-called biological selectivity filters.



Simulation box, earlier work.

Present work starts with equilibrated water in compartment I and equilibrated solution in compartment II

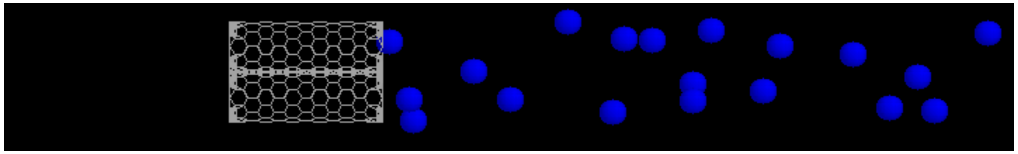
The simulation box



- ● K^+ ion
- ● Cl^- ion
- ● surface charges (present in other studies, not in this one)
- ● H_2O

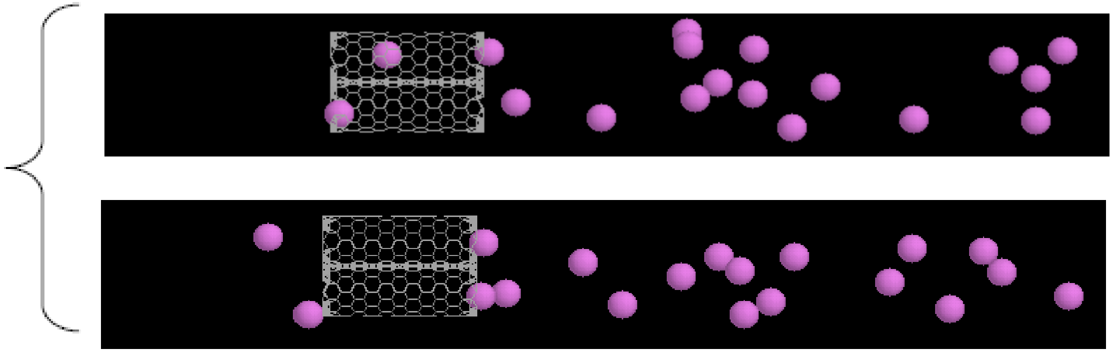
Uncharged carbon atoms of nanotube and dummy atoms are white

(10,0) carbon nanotube

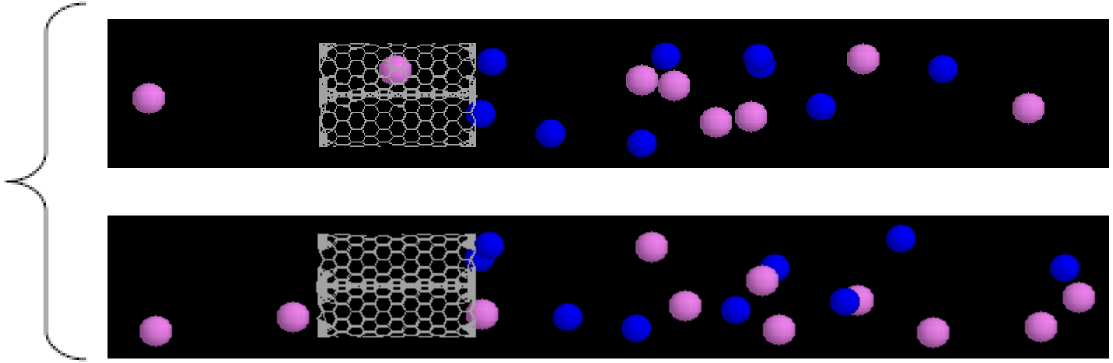


NaCl solution

Permeation direction



KCl solution



NaCl+KCl solution

0.4374 nm diam (similar to size of selectivity filter of potassium ion channel)

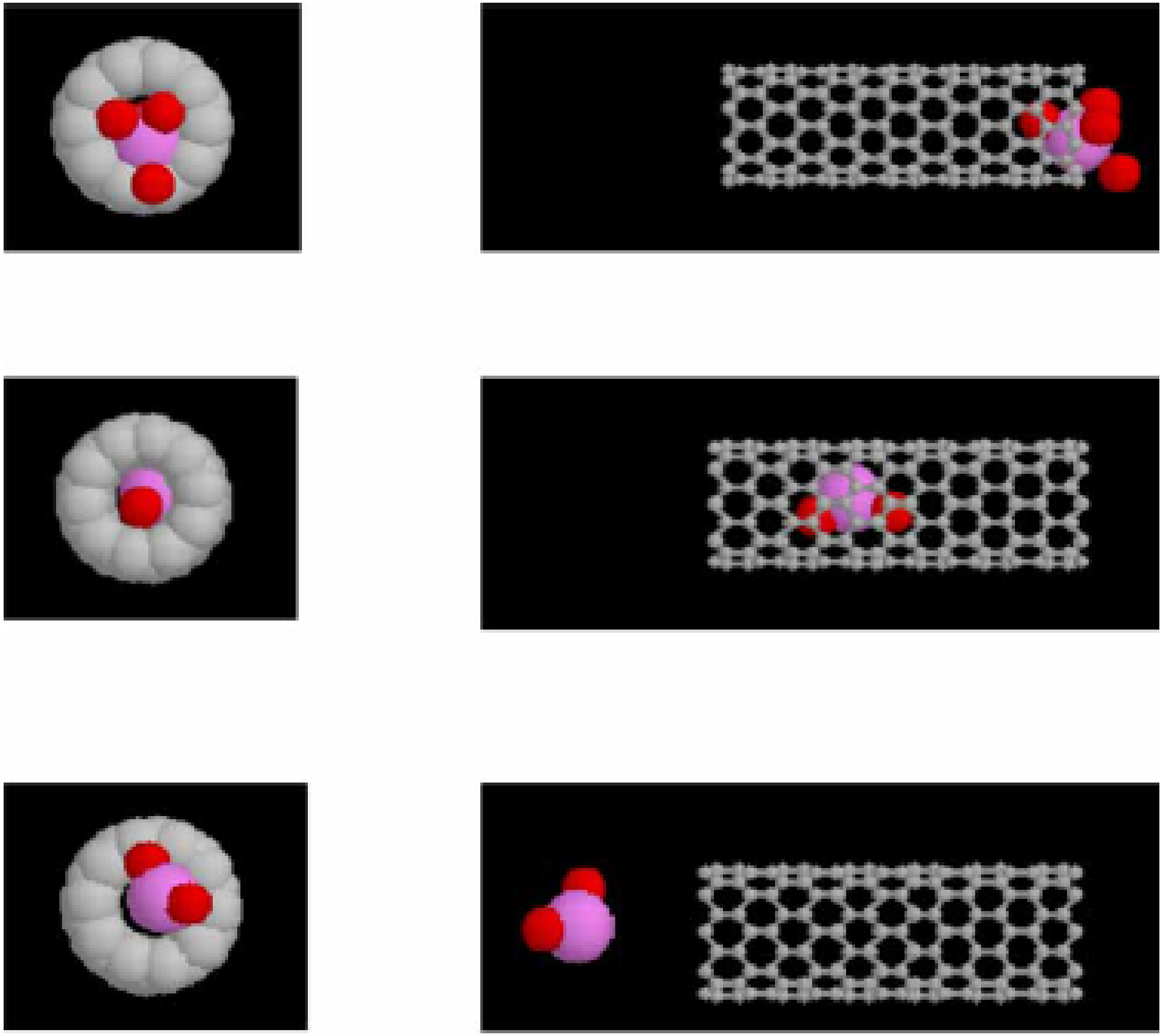
FINDINGS

- At 298K and an initial pressure of 5MPa with a concentration of 1.85 mole % (either NaCl, or KCl solution in water) show that one K^+ ion entered the membrane and was able to travel the entire length of the membrane and exit it. In contrast, none of the Na^+ ions were able to even enter the membrane.

FINDINGS

- When the solution contained equal amounts of NaCl and KCl (total concentration unchanged), once again one K^+ ion entered and exited the membrane, and once again none of the Na^+ ions were able to permeate or exit the membrane at all.
- Membranes with the smaller diameter (10,0) CNTs can be effective selectivity filters. The 0.4374 nm diam is similar to size of selectivity filter of potassium ion channel.

Snapshots of a K^+ ion permeating through a (10,0) carbon nanotube



Mechanism for permeation of hydrated K^+ ion

- The hydration shell of K^+ can be partially stripped off to make the solvated ion effectively linear, so that the K^+ ion can not only enter the membrane, but can also be readily transported across the entire length of the membrane.
- Only two waters are stripped off but these can be relegated to the position water molecules normally occupy in the second hydration shell.

The hydrated Na^+ ion, in contrast

- The higher solvation energy, and the corresponding higher energetic barrier to distorting the solvation shell, of Na+ ion in comparison with K^+ ions is primarily responsible for the observed difference.
- Only when both the pressure and temperature are increased in pure NaCl solutions (325 K, 3500 MPa), did we observe some permeation of Na^+ ions. At increased *T* and *P* the energetic cost of desolvating a Na^+ is available.

SUMMARY

- We actually followed at an atomistic level the permeation and transport of K^+ vs. Na^+ ions and thus has described the molecular mechanism of ion selectivity of nanochannels of diameter 0.437 nm. This mechanism does not depend on having dynamic, flexible coordinating groups acting as selectivity filters, and is therefore expected to operate in general in nanochannels of this approximate size.

CONCLUSIONS

- Our results in membranes consisting of uncharged CNT have shown that an ion filter can be effective even in the absence of Coulombic exclusion or coordinating chemical structures, based exclusively on the characteristics of the hydrated ion, and the corresponding energy of partial desolvation.

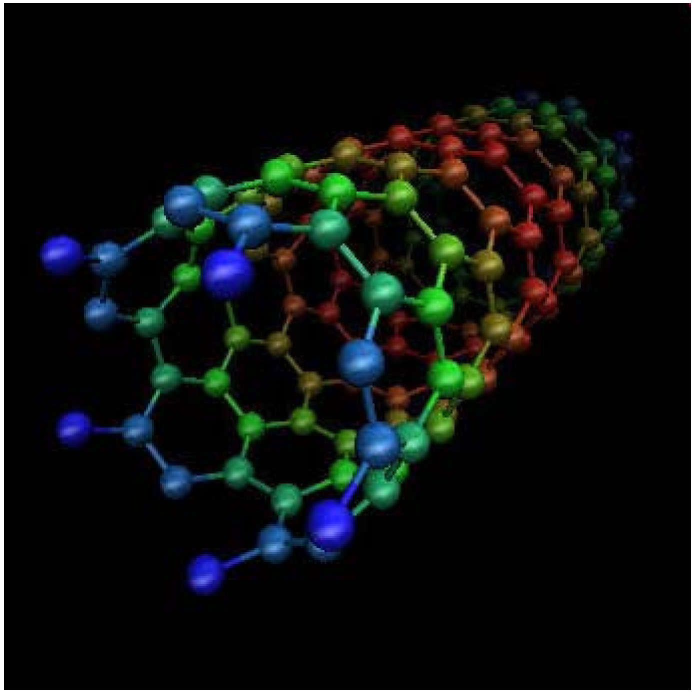
Permeation of ions into surface-charged tubes

- characteristics of hydrated ions in nanotubes with surface charges compared to uncharged tubes: radial distribution functions, hydration numbers
- particle density profiles in surface-charged tubes compared with electrical double layer model
- trajectories of ions in surface-charged tubes
- dependence of permeation rates on concentrations, tube diameter, surface charge density, and other factors
- comparison with experiments in silica channels

with Hongmei Liu

Surface-charged nanotubes

To study the effect of surface charges on permeation of ions into nanochannels, we arbitrarily assign charges on specific carbons of a nanotube and construct membranes with these.

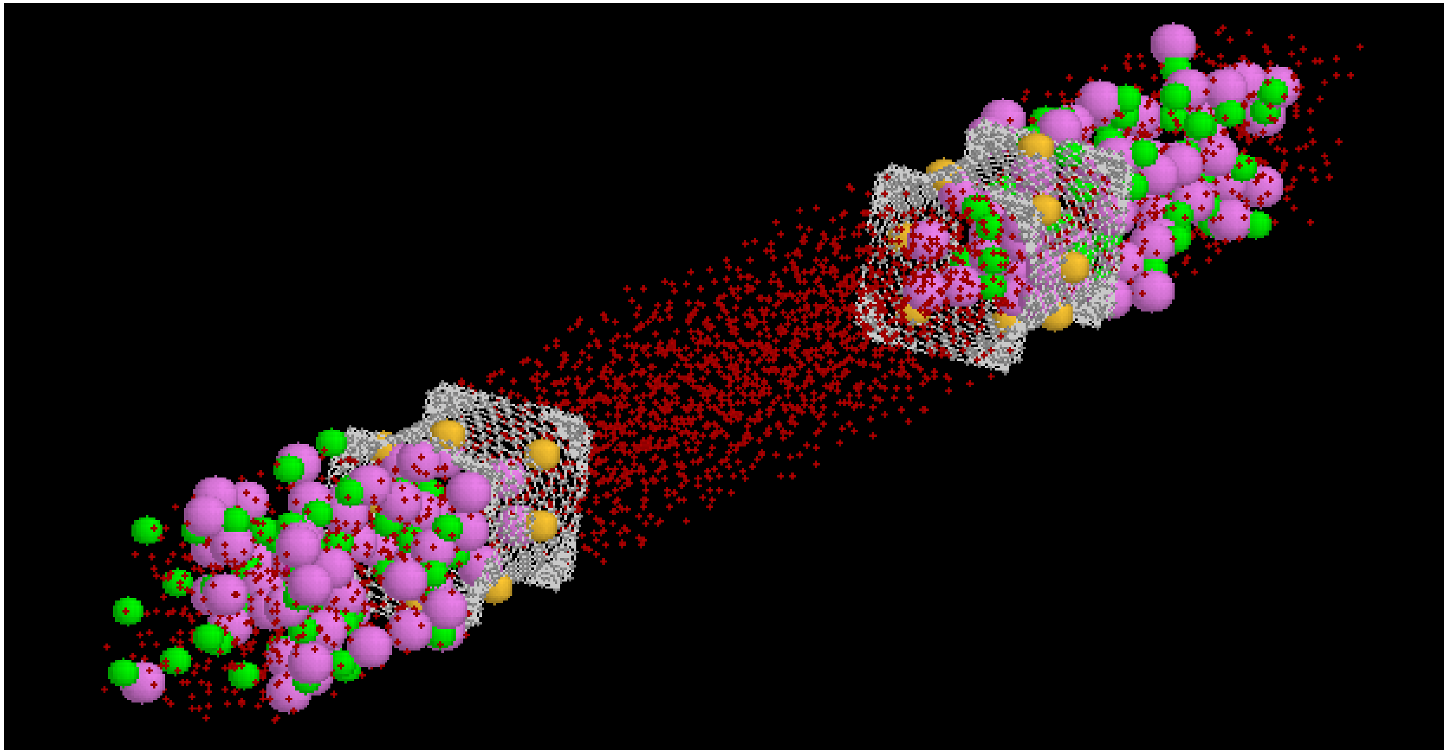


http://nanohub.org/site/media/images/nanotubes.jpg

Permeation of K^+ ions into a surface-charged tube. Our model

- zigzag type carbon nanotube (25,0), modified by symmetric distribution of charges on tube atoms
- 8 surface charges each -0.25e, net charge on each tube is -2e
- start with pure water in the middle compartment,
- 4.97 M KCl solution in outer compartment (3214 H_2O, 123 K^+, 119 Cl^-),
- show 10 snapshots, every 0.021 ns

The simulation box



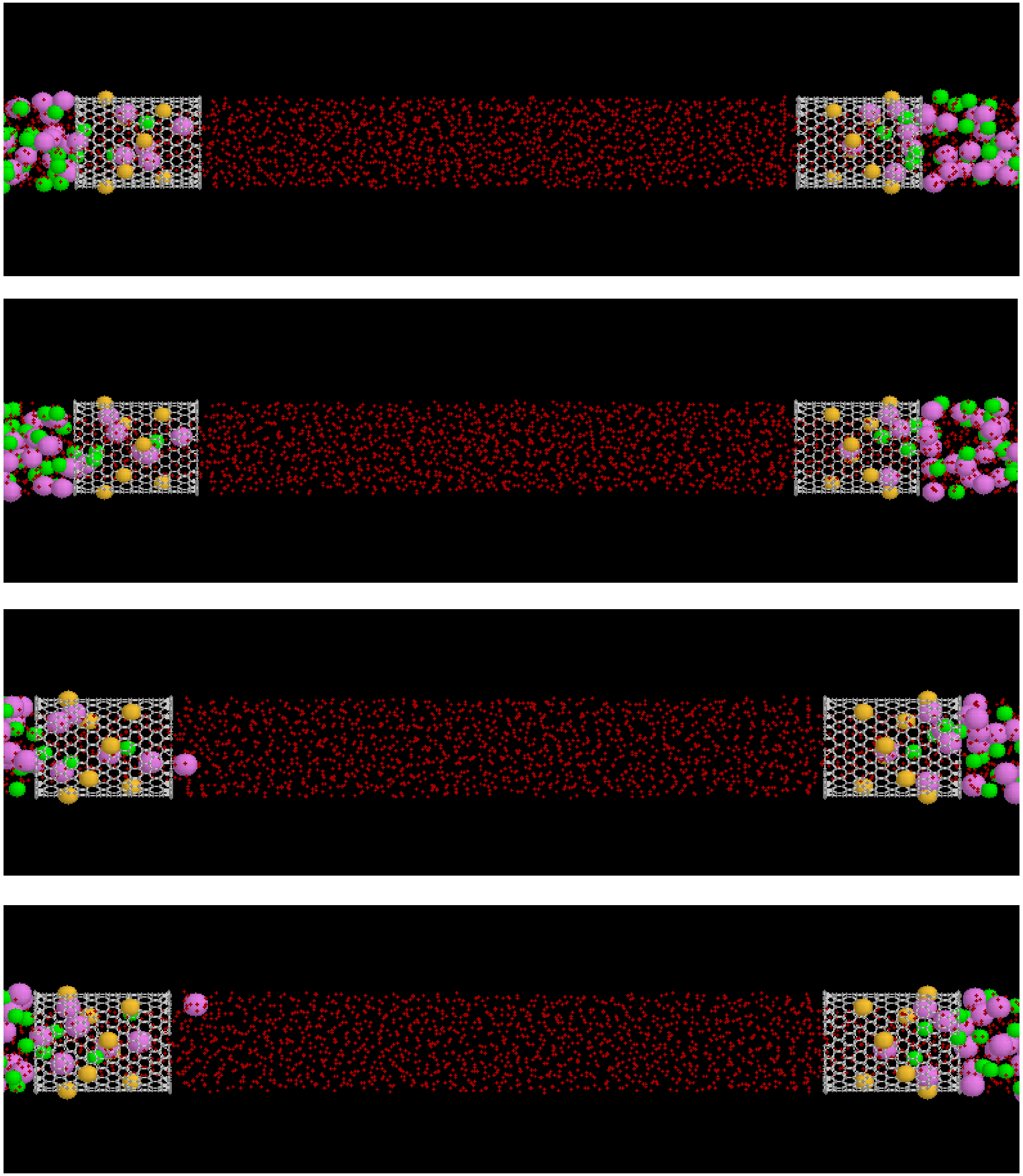
● K^+ ion

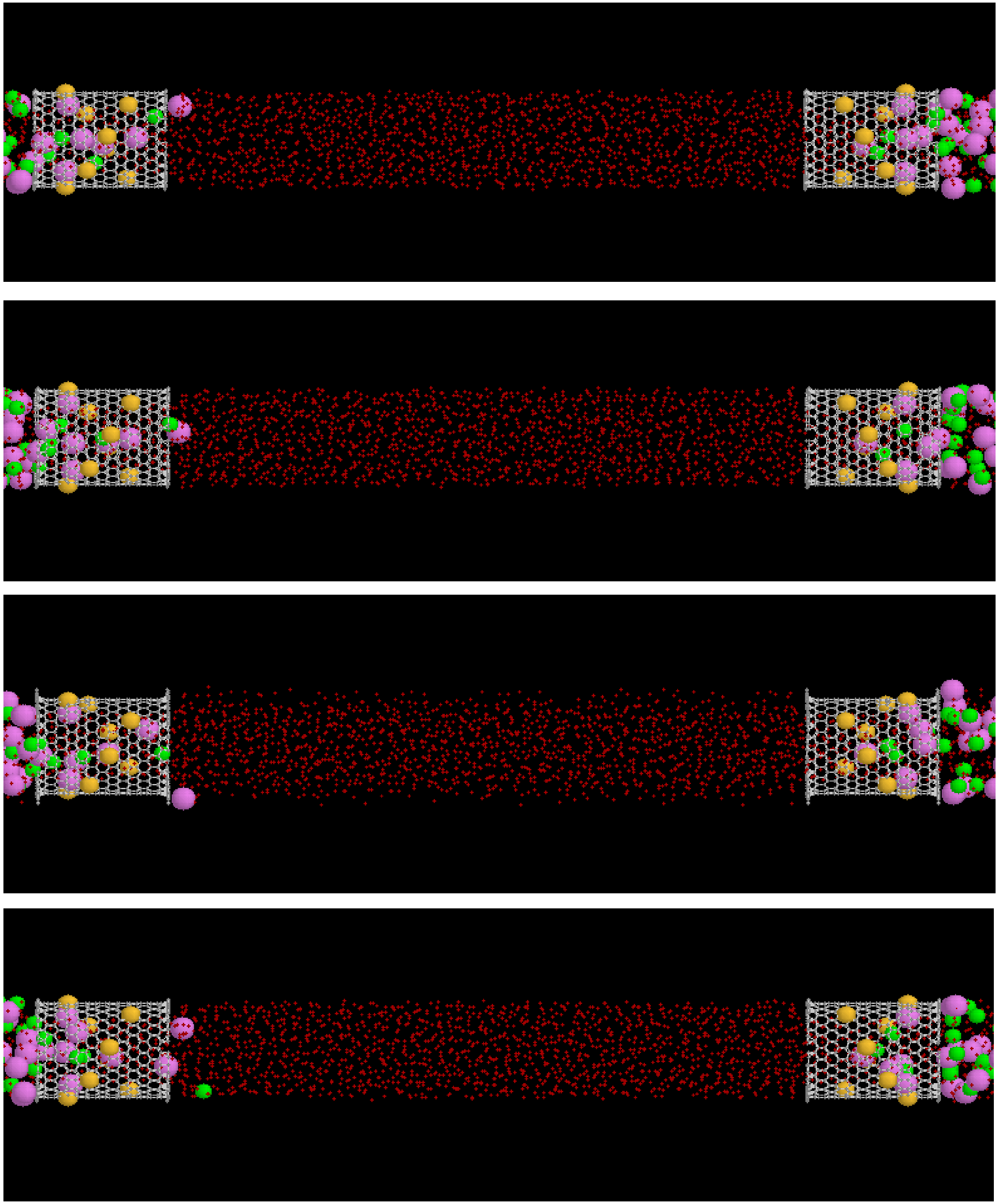
Uncharged carbon atoms of nanotube and dummy atoms are shown in white

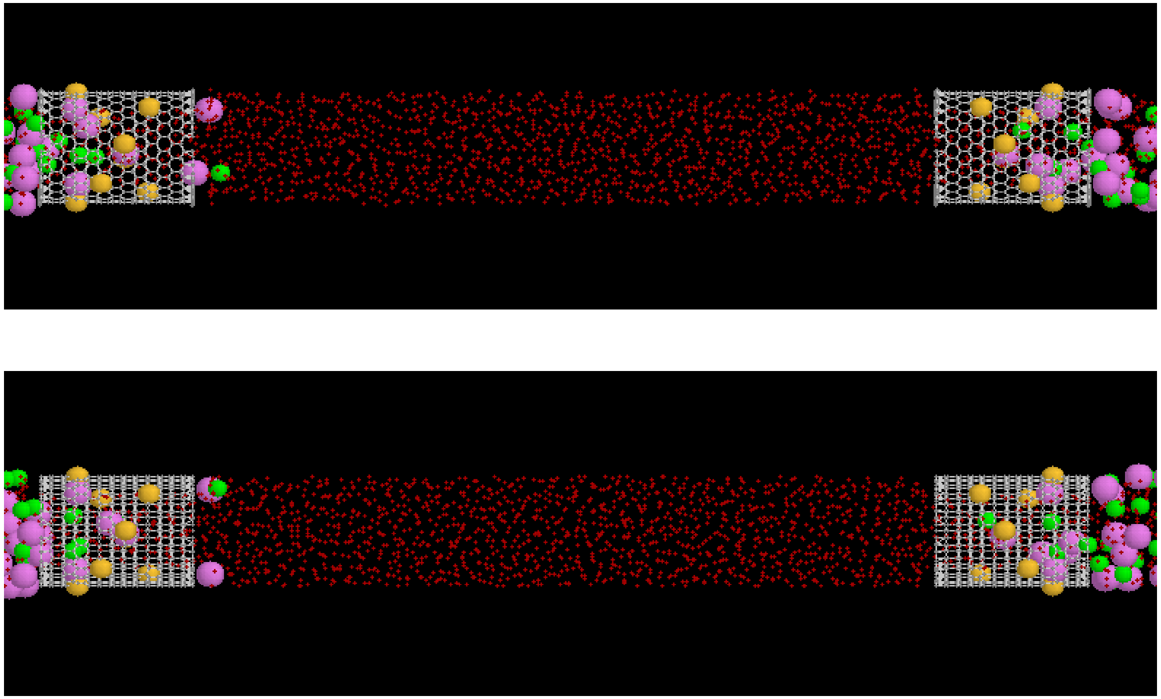
● Cl^- ion

● **surface charges**

● H_2O







K^+ ion distribution after 0.21 ns

8 surface charges each -0.25e, net charge on the tube is -2e

where	how many
entrance < ring1	3
ring1< ring2	4
ring2 < ring3	1
ring3 < ring4	2
ring4 < exit	0
>=exit	2

K^+ ion distribution after 0.21 ns

Increase charges:

8 surface charges each -1e,
net charge on the tube is -8e

where	how many
entrance < ring1	7
ring1< ring2	6
ring2 < ring3	4
ring3 < ring4	7
ring4 < exit	3
>=exit	0

CONCLUSIONS

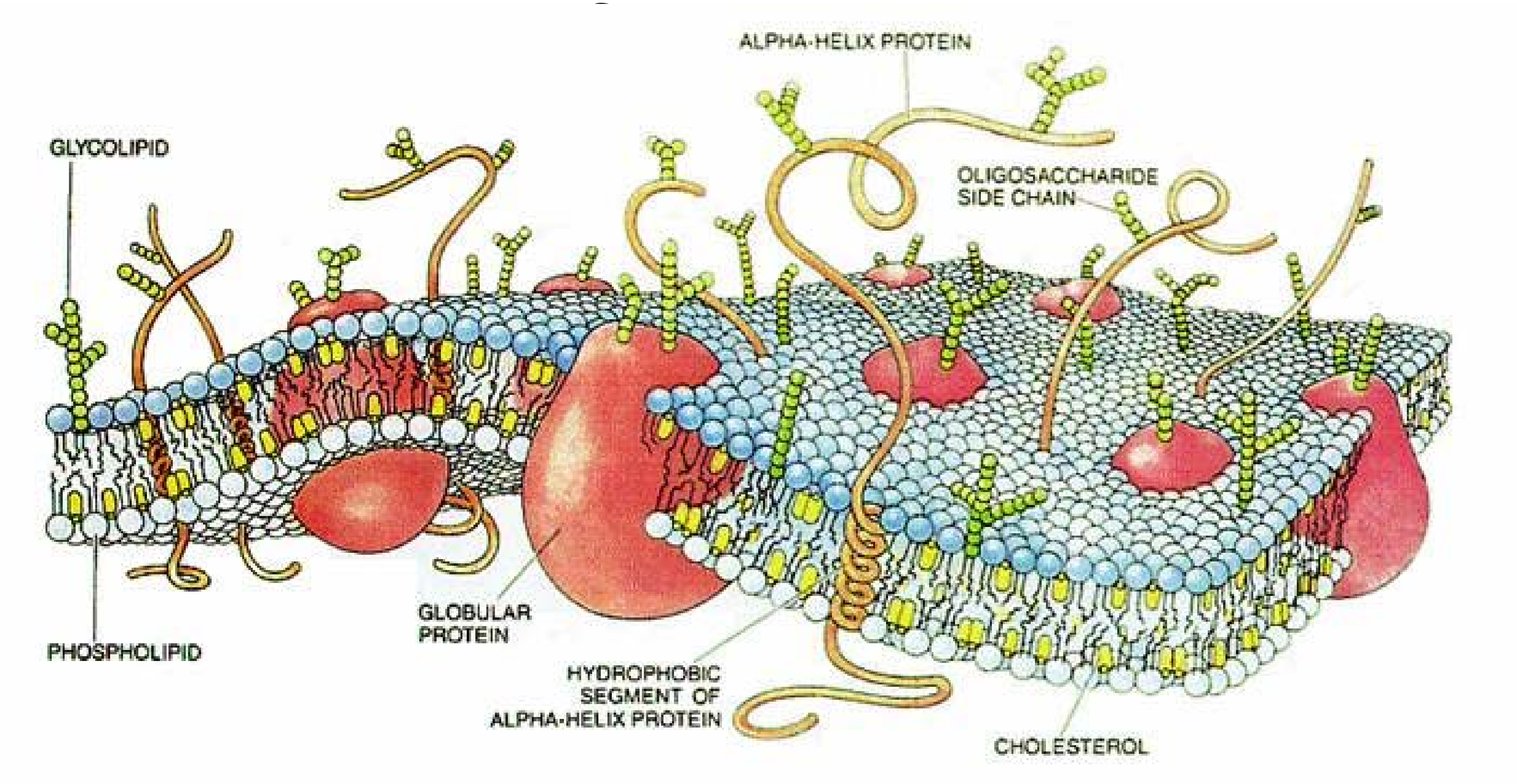
- Increased magnitude of surface charge led to greater number of K^+ ions permeating into the tube, but fewer ions exiting the tube
- Increased concentration of solution led to greater number of K^+ ions permeating into the same tube at the same stage of simulation.

TRANSPORT THROUGH LIPID BILAYER MODEL MEMBRANES

- Adopt a coarse-grain model using 4 to 1 mapping of 4 heavy atoms into 1 interaction site, use Marink force field
- Test the potential for the lipid bilayer itself. Does it self-assemble to the experimental thickness? Compare characteristic properties with experimental values.
- Introduce intermolecular interactions of the lipid with a solute, for example, Xe, O_2, CO_2 , define permeability in an MD simulation, compare permeabilities from simulations with experimental results
- Embed an outer membrane protein, OmpA into the lipid bilayer and observe transport of Xe, O_2, CO_2 .

with Huajun Yuan

Motivation



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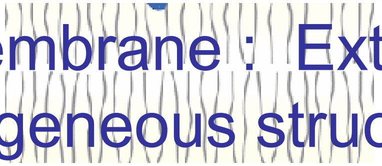


Biological membrane： Extremely Complex system, highly heterogeneous structure, displays biologically-relevant behavior.

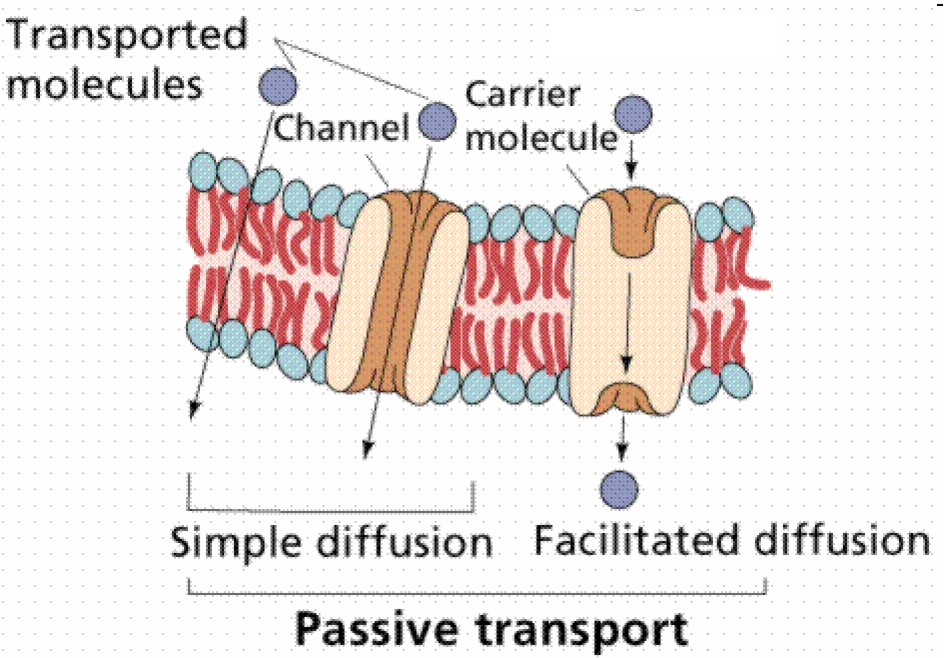
Membrane-Mediated Interaction of Celecoxib with COX-2
Ref: Atherosclerosis *2004*

A model membrane: lipid bilayer model system

- Start with general permeation of lipid bilayer by small molecules
- Later, simulate insertion of specialized pore-formers and permeation through pores as well

Gas Transport Through Biological Membranes: Important for life itself

Membrane Transport of Small Molecules:



Computer modeling of membrane transport of small molecules

- **Traditional Atomic-Level Model:** Accurate, but very expensive
- **Coarse-grained Model:** Significantly reduce number of interaction sites, simplified force field consists of beads interacting through effective interaction functions

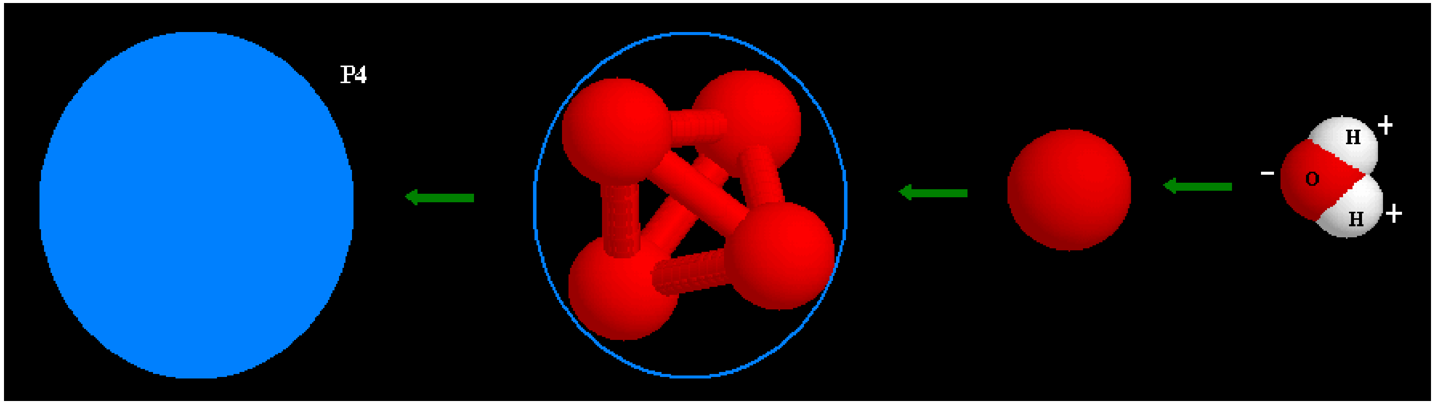
Permeation of lipid bilayer model membranes by small molecules: Xe, O_2, CO_2

- Use coarse-grained model
- Test the potential for the lipid bilayer itself. Does it self-assemble to the experimental thickness? Introduce intermolecular interactions of the lipid with a solute, for example, Xe, O_2, CO_2
- Test existing permeation theories, define permeability in an MD simulation
- Compare permeabilities from simulations with experimental results

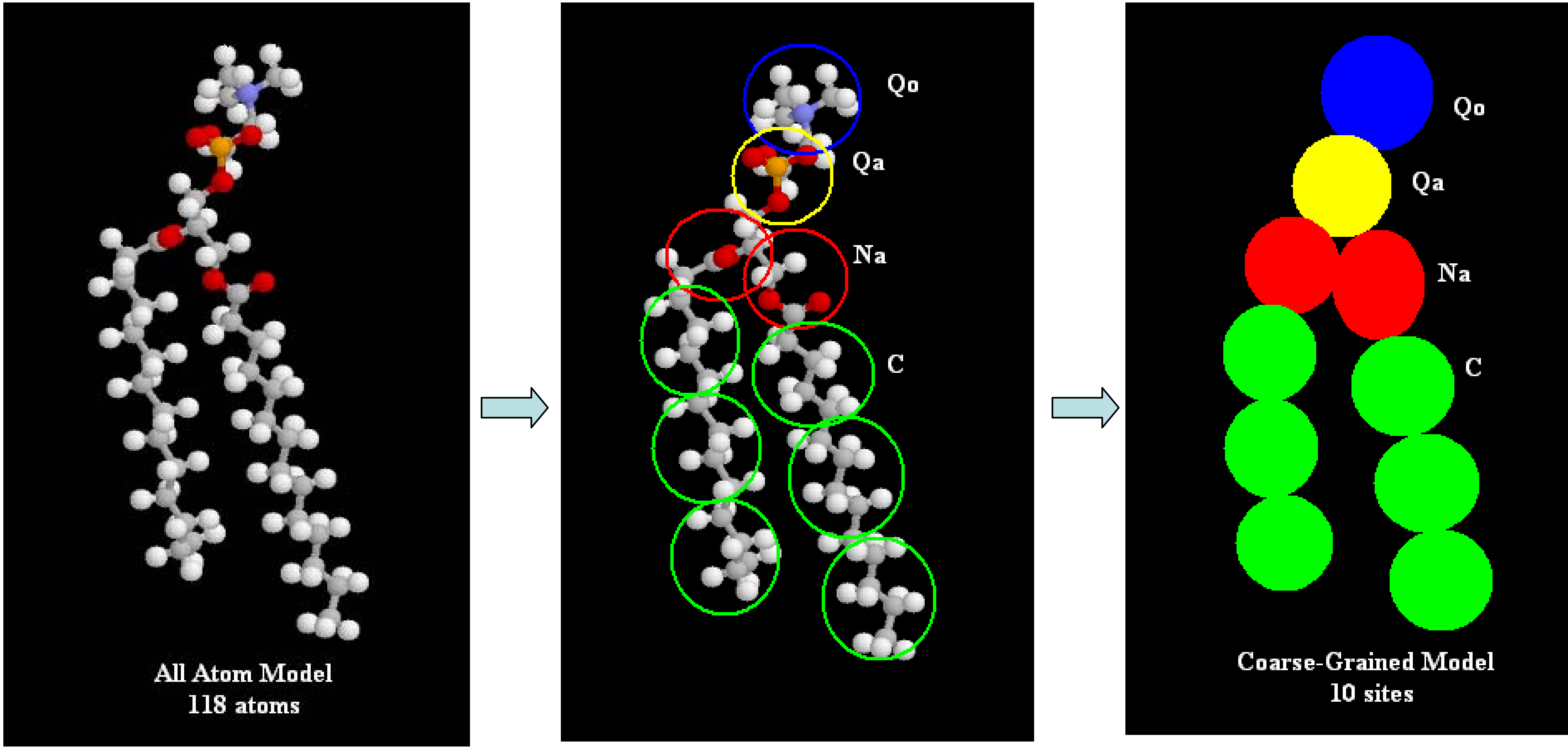
Coarse grain force field

MARTINI force field by Marrink et al J.Chem.Theory and Comput. 2008, 4, 819-834

Coarse grain mapping strategy
4 → 1



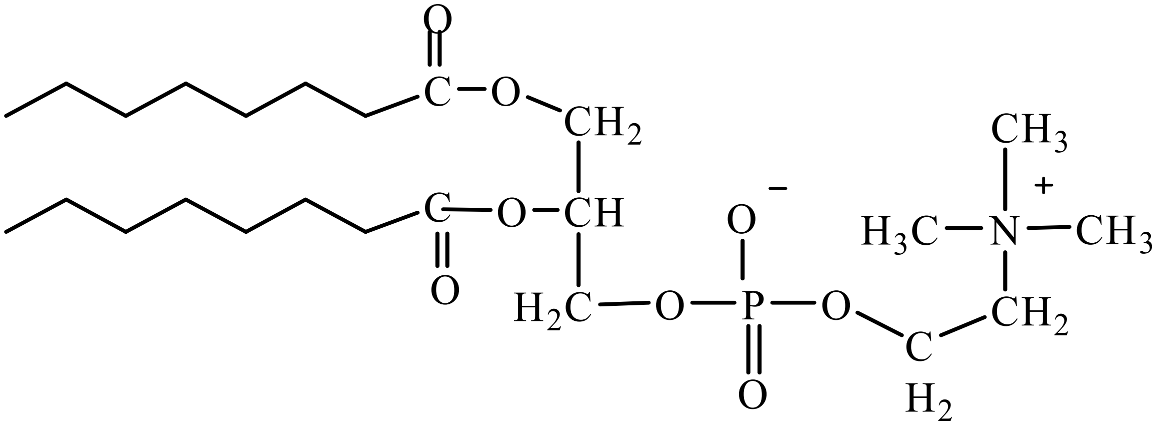
Lipid (DMPC):



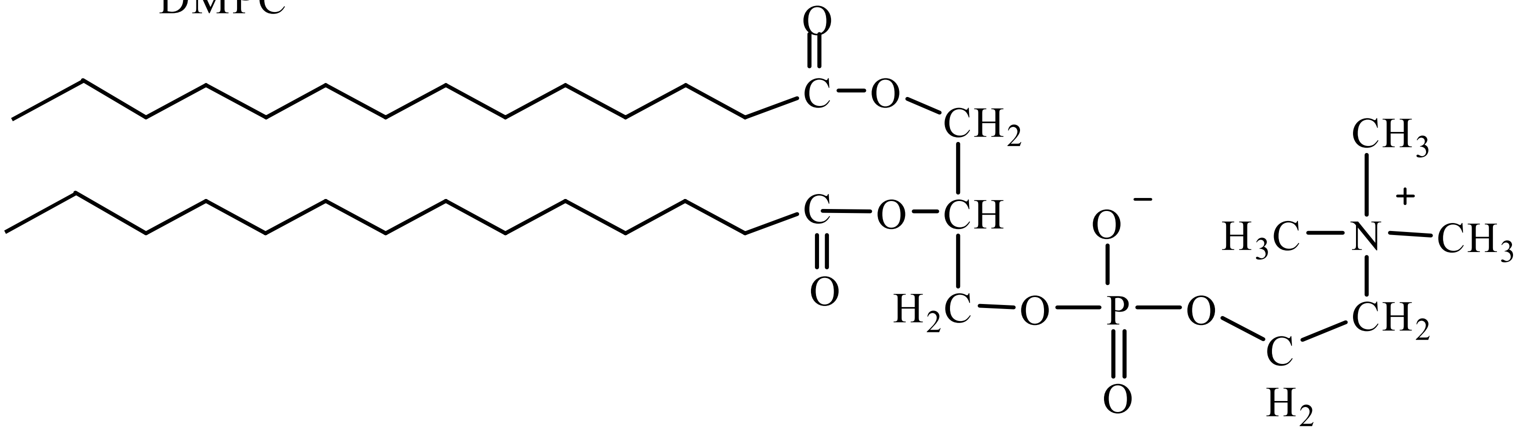
All Atom Model
118 atoms

Coarse-Grained Model
10 sites

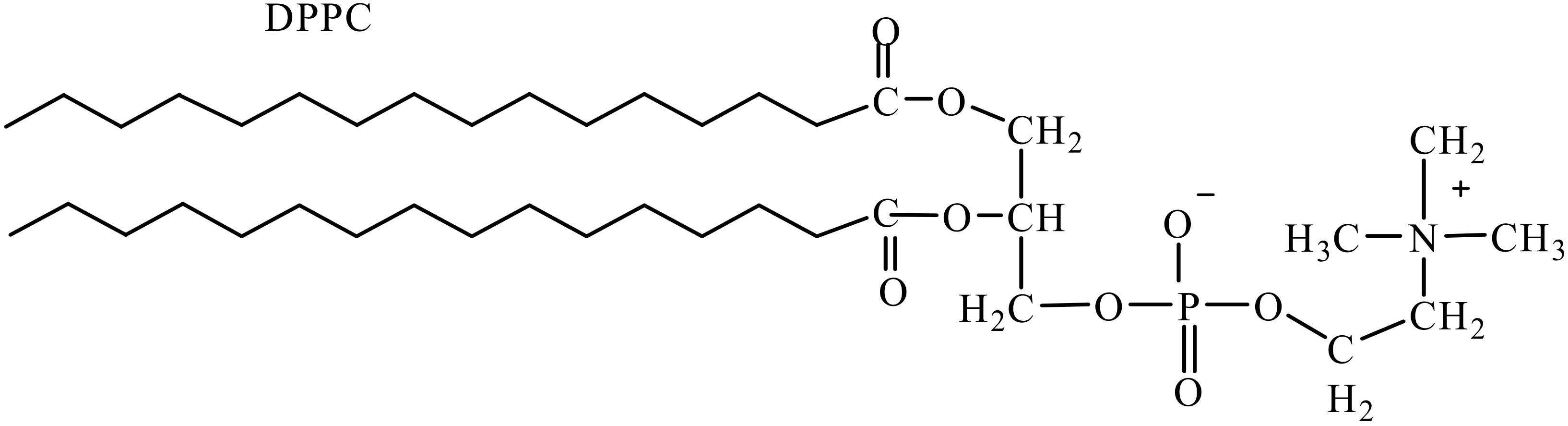
DCPC



DMPC



DPPC



Interaction Potential :

$$u=\sum_{i,j} 4\epsilon_{ij}\left[\left(\frac{r_{ij}}{\sigma_{ij}}\right)^{-12}-\left(\frac{r_{ij}}{\sigma_{ij}}\right)^{-6}\right]+q_iq_j/r_{ij}$$

Parameter Set					
i, j	**Qo**	**Qa**	**Na**	**C**	**P4**
Qo	IV	II	III	IX	O
Qa	II	I	III	IX	O
Na	III	III	III	VI	III
C	IX	IX	VI	IV	VIII
P4	O	O	III	VIII	I

O, I, II… etc are paremeter sets provided by Marrink et al
d- donor, a-acceptor, da-both, o-none, 1~5-degree of polarity
P-polar, N-nonpolar, C-apolar, Q-charged

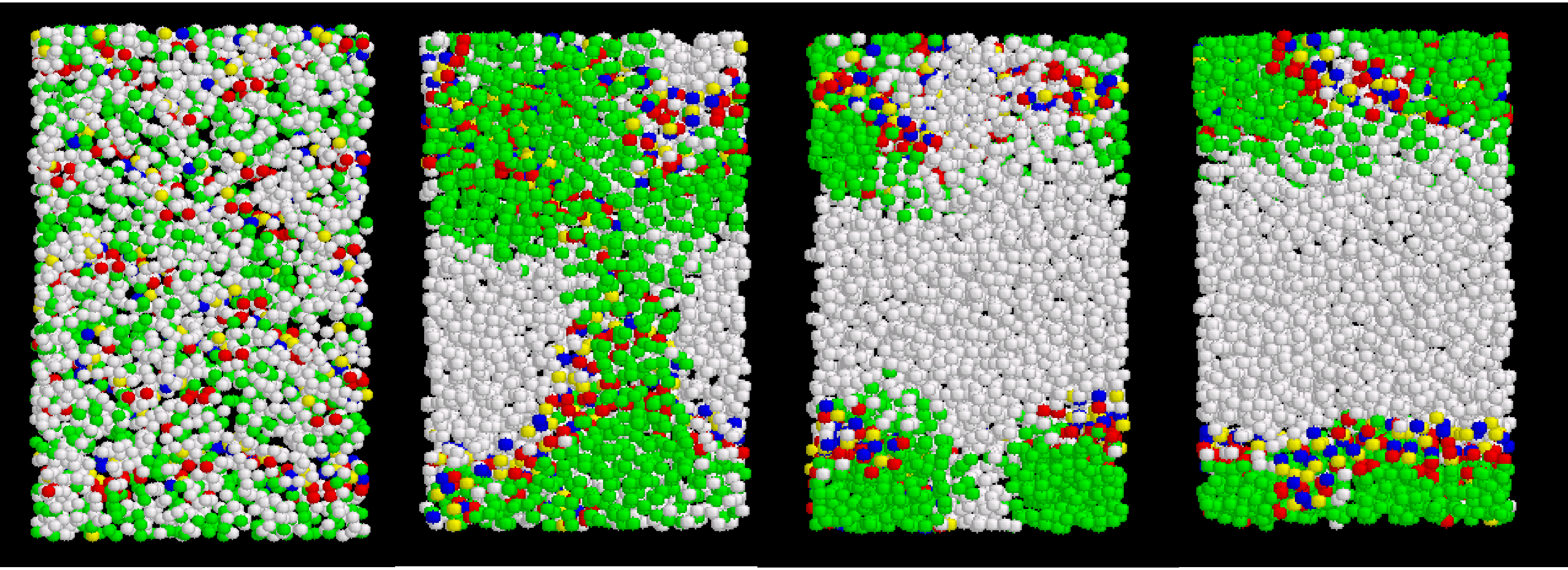
bond stretch: $V_{\text{bond}}(R)=\frac{1}{2}K_{\text{bond}}(R-R_{\text{bond}})^2$

angle bend: $V_{\text{angle}}(\theta)=\frac{1}{2}K_{\text{angle}}\{\cos(\theta)-\cos(\theta_0)\}^2$

Validation tests for coarse grained DPPC

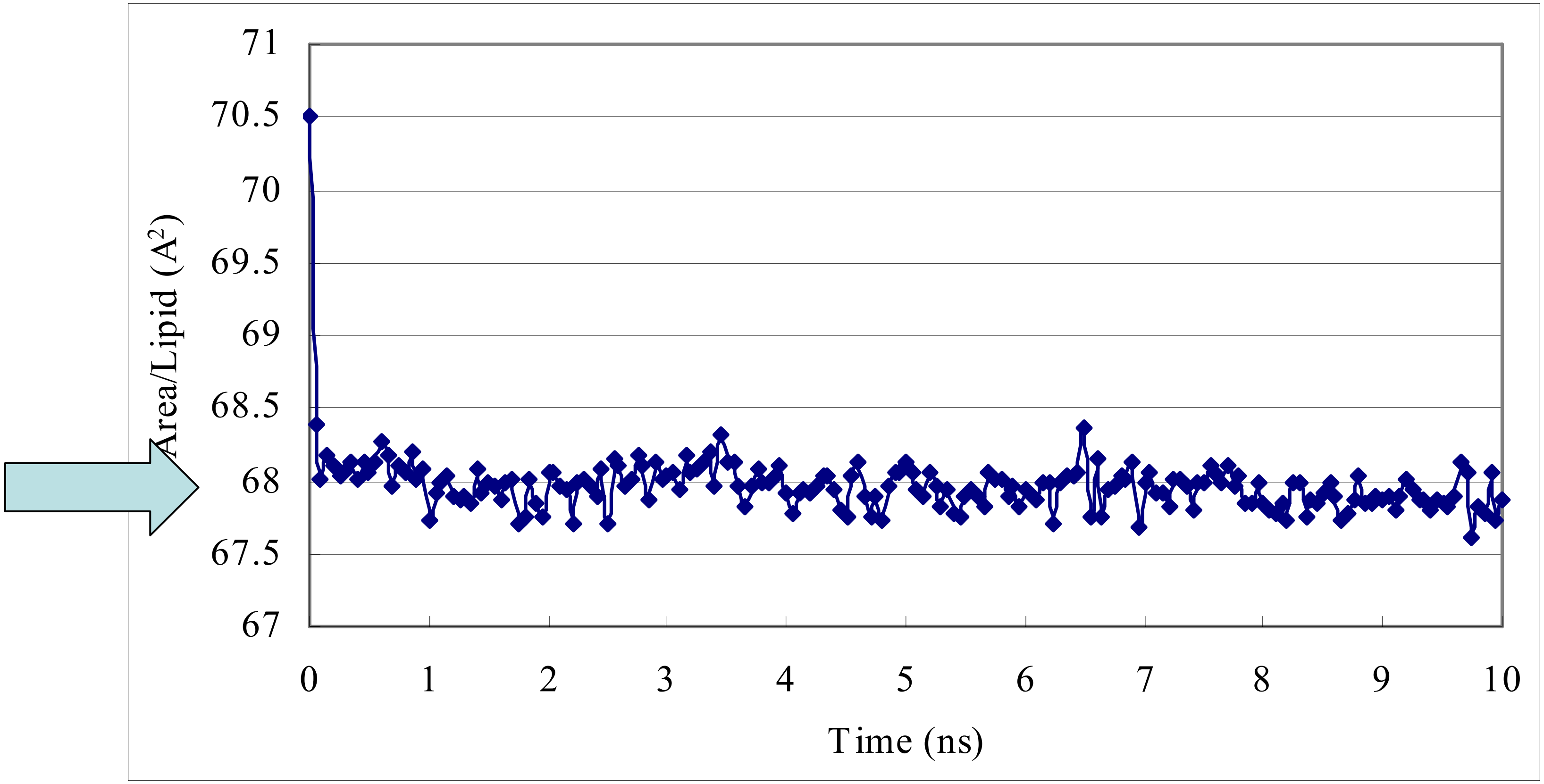
- self assembly to a stable bilayer
- area per lipid for the self-assembled bilayer
- thickness of the bilayer (phosphate-to-phosphate distance across)
- density profile along the normal to bilayer
- order parameter of segments of the tail

Self-Assembly occurs at 323K :



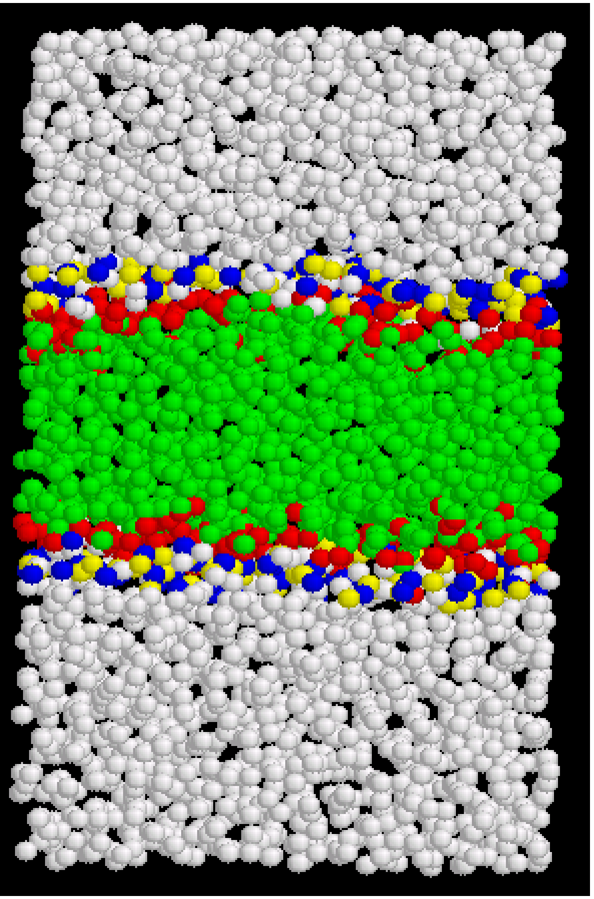
t = 0 ns t = 1 ns t = 10 ns t = 100 ns

area per lipid at 323 K

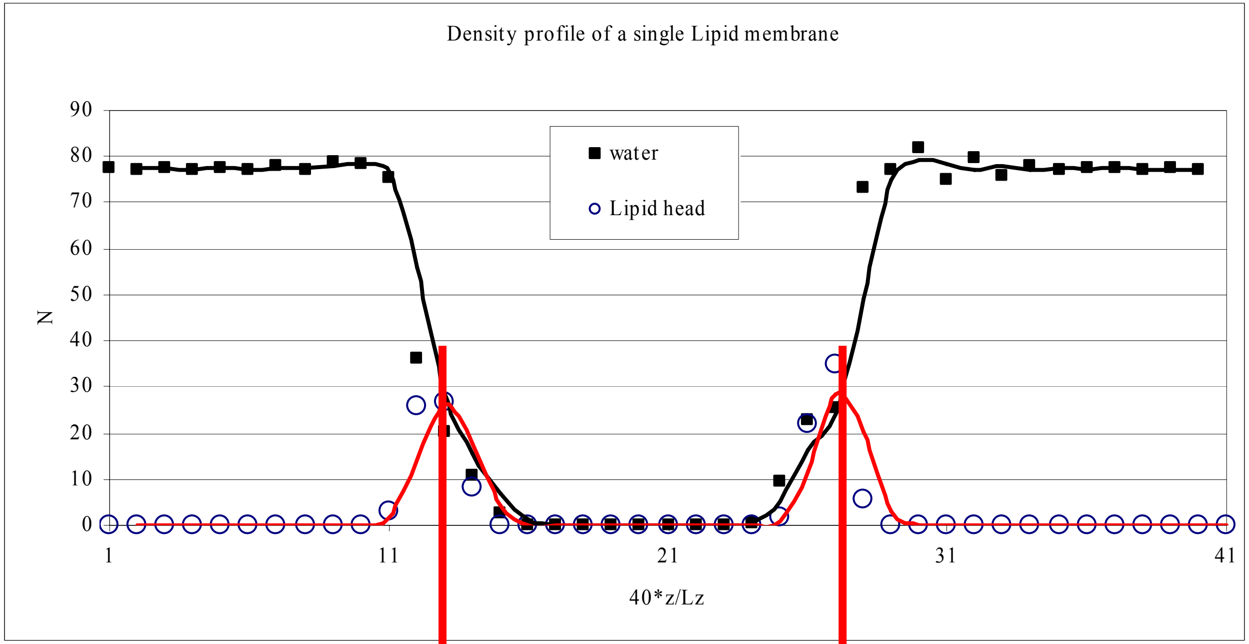


We find 68 Å², in good agreement with experimental values which are in the range 57-71 Å²

Lipid Bilayer in water at 323K: 128DPPC+2000H_2O



Density profile at equilibrium



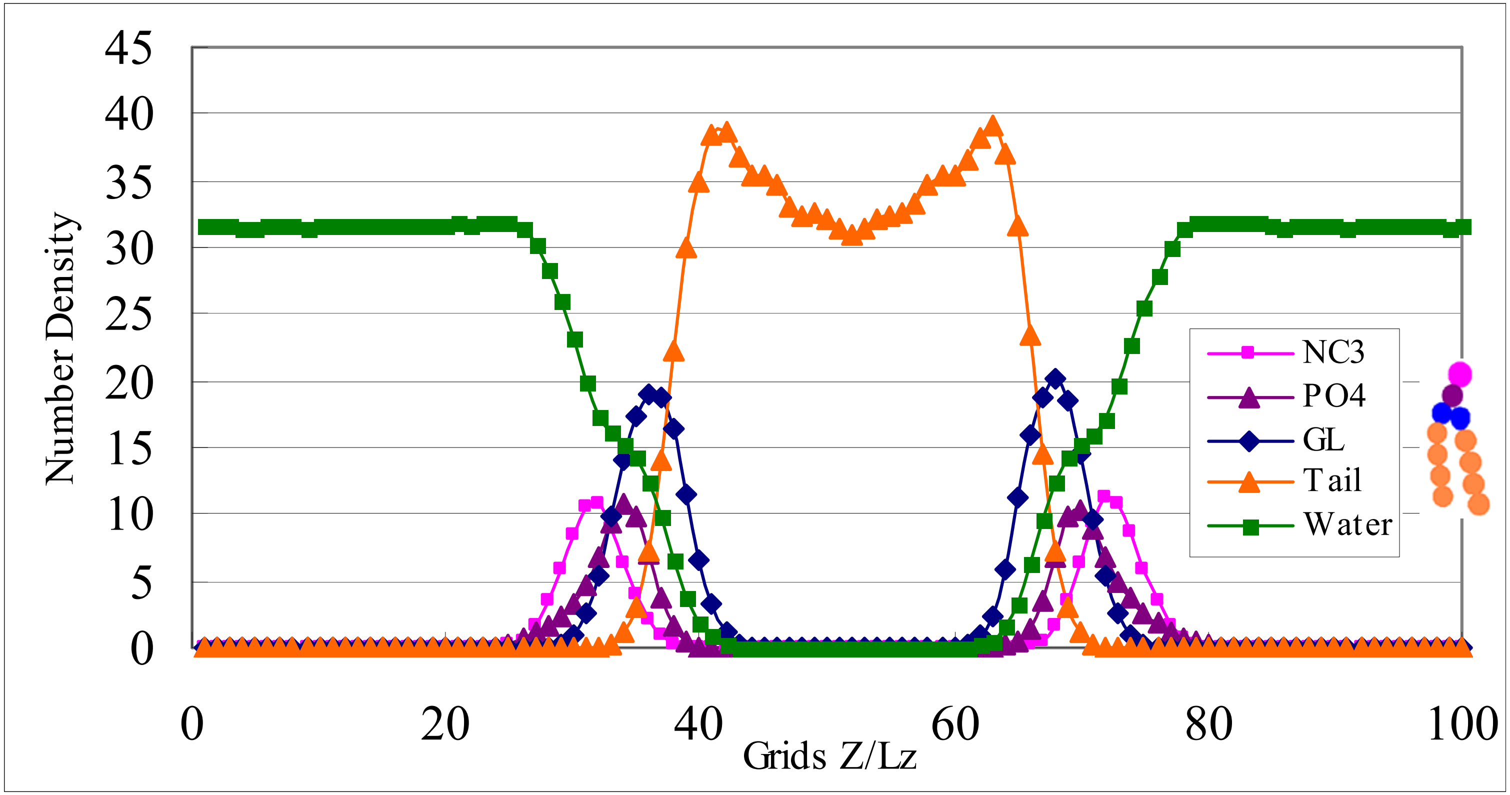
THICKNESS OF THE BILAYER

3.75 nm

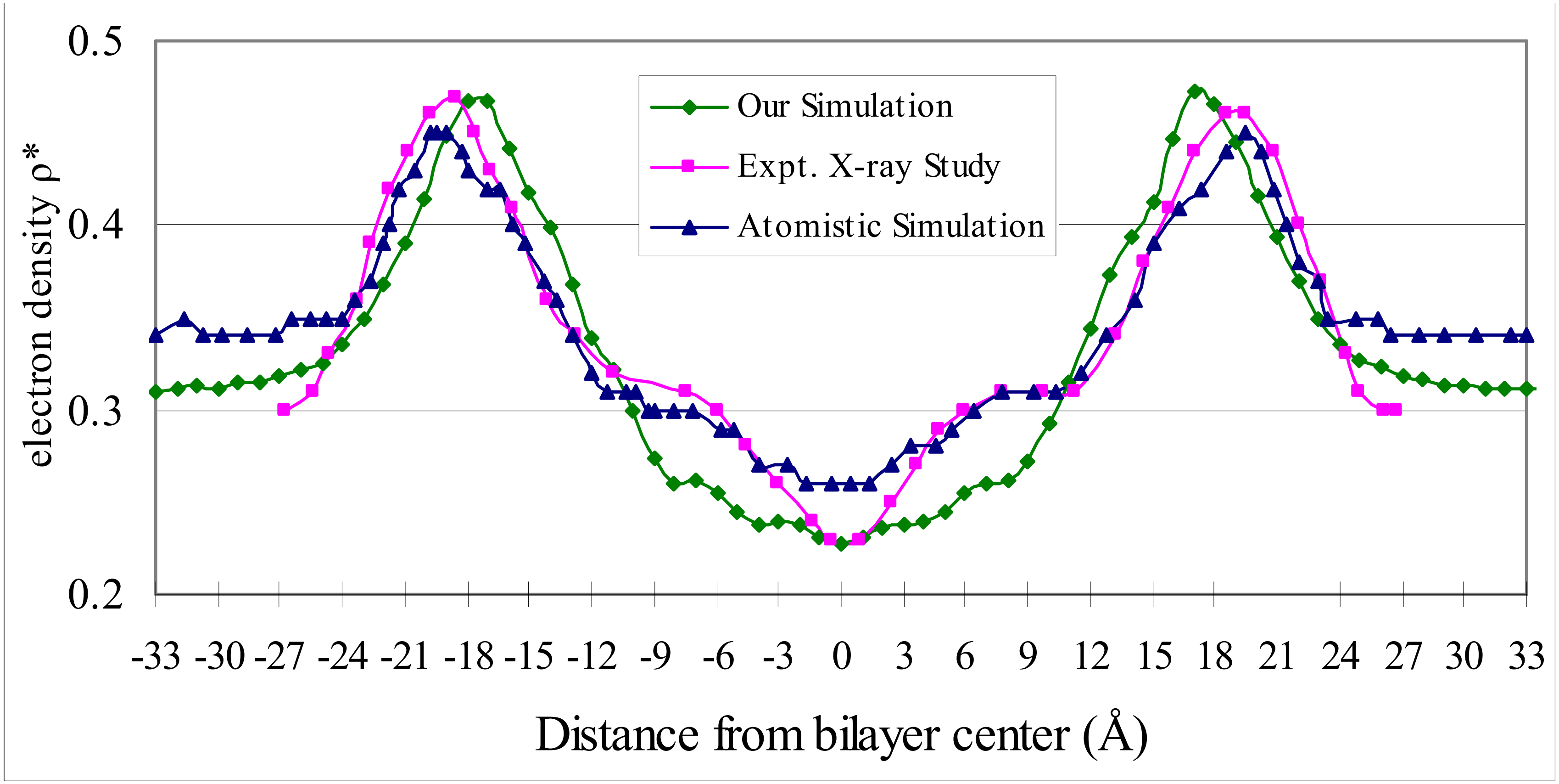
Expt. 3.85 nm

Ref. Marrink et al, J.Phys.Chem B 2004, 108, 750-760

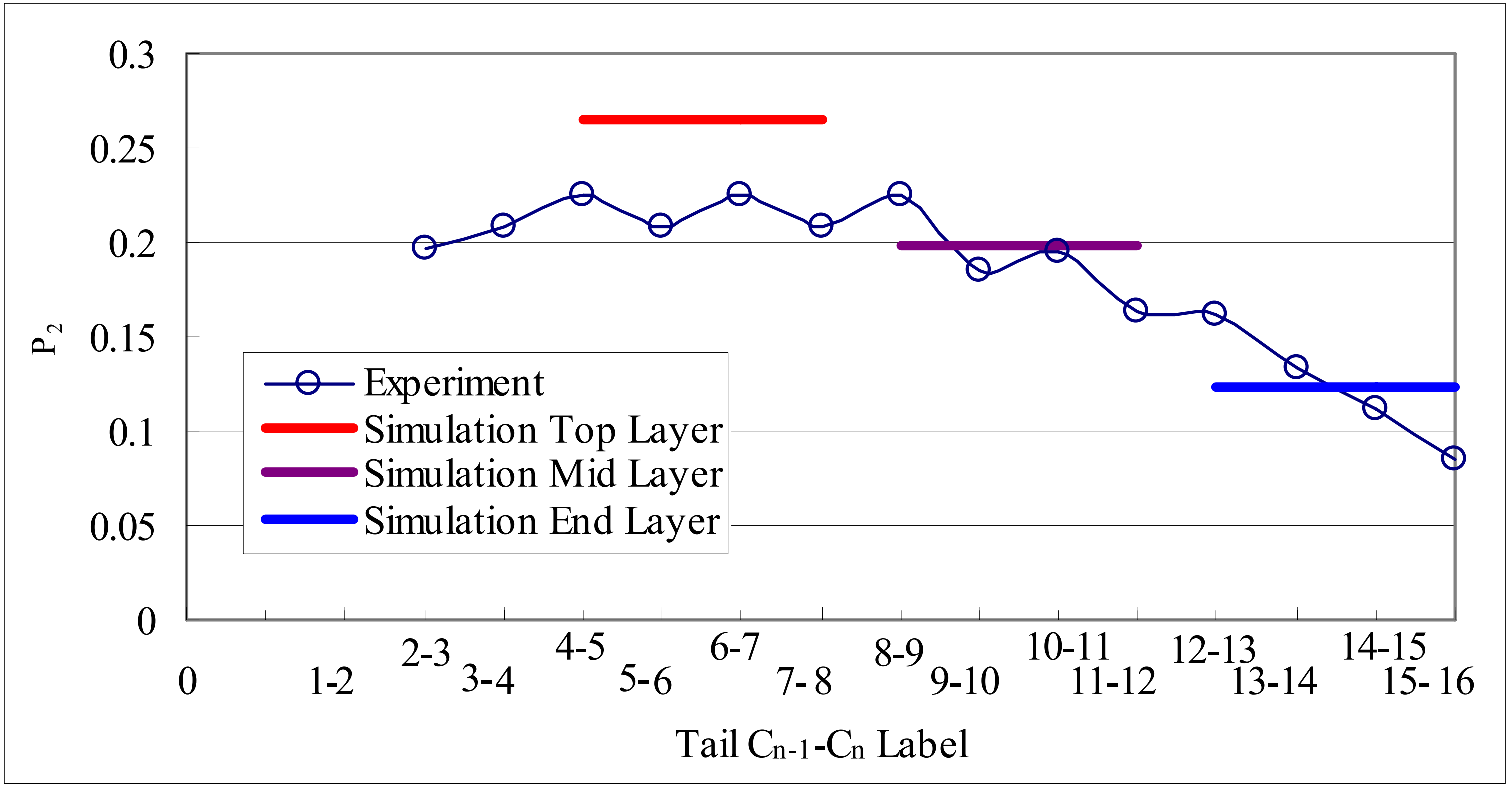
density profile along the normal

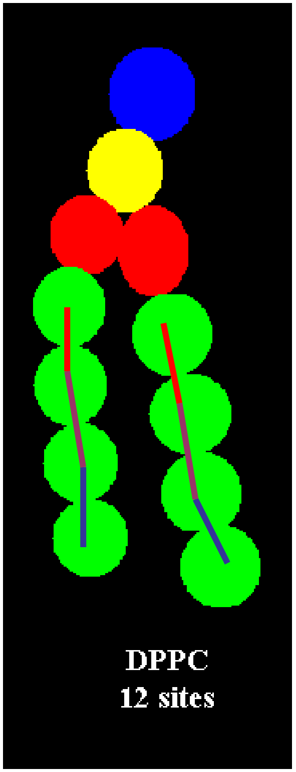


Comparison of simulated electron density with X-ray data and with atomistic simulations



order parameter of segments of the tail

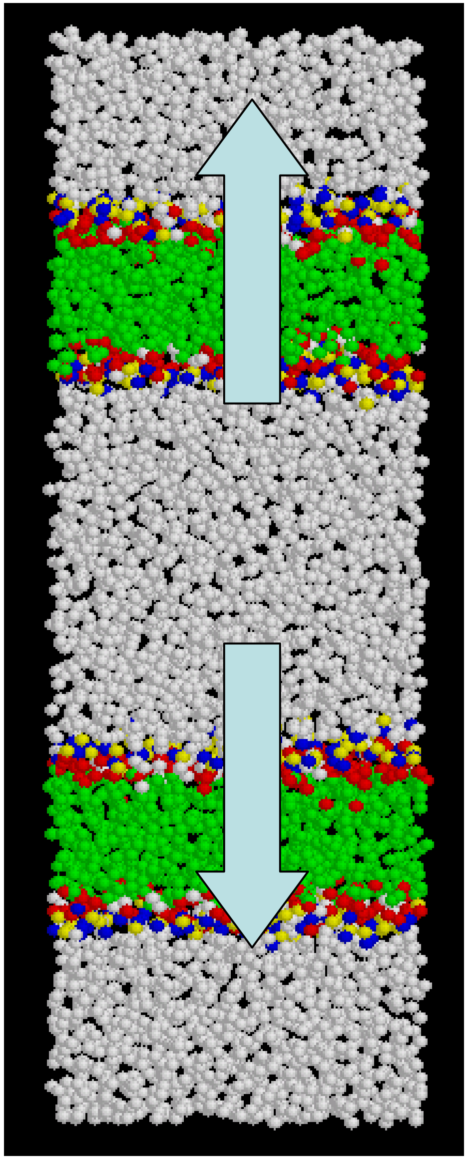


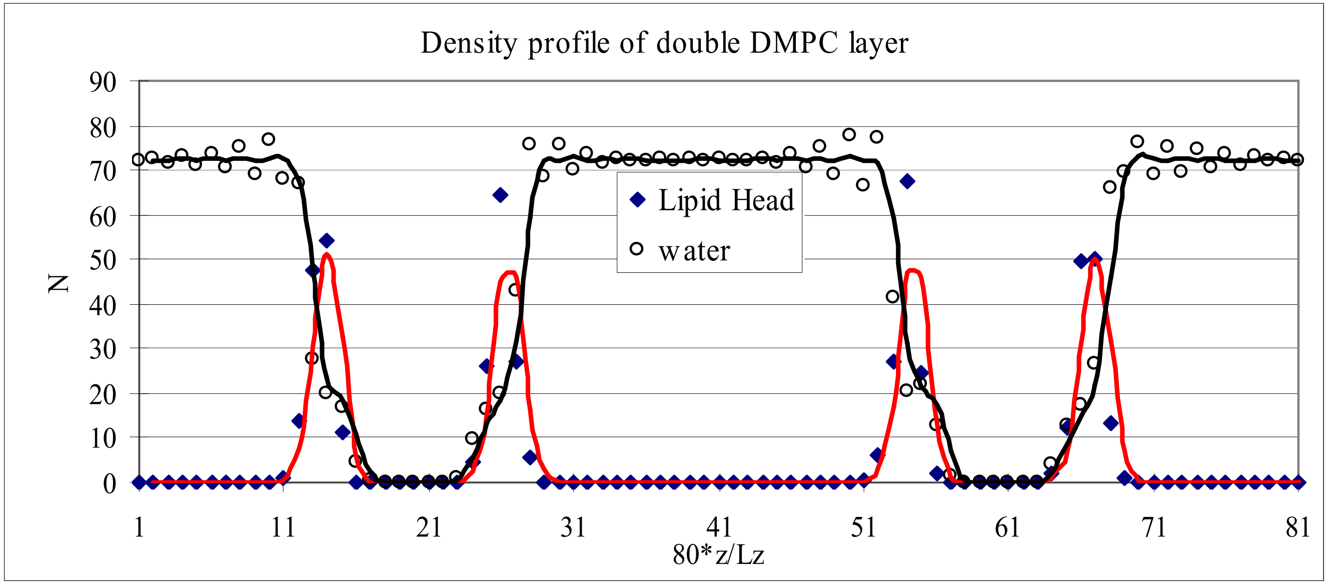


Experimental values are order parameters for C_{n-1}-C_n bonds, derived from measured C-D bond order parameters from deuterium NMR measurements in DPPC.

transport of gases through the lipid bilayer model membrane

Simulation box for transport studies





Xe placed initially in the center compartment
will diffuse through the membrane to the outside compartments

Gas parameters

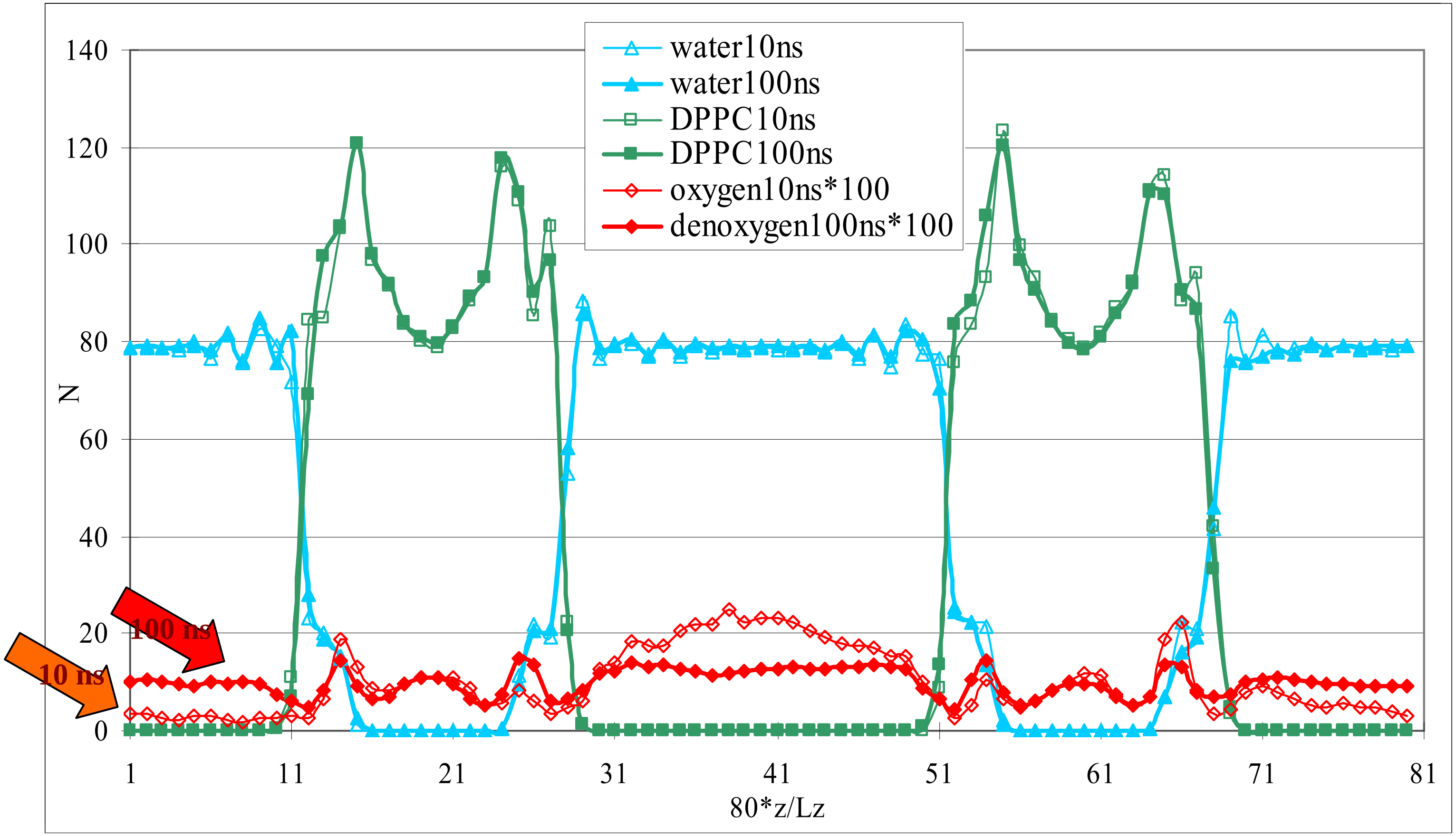
- Oxygen : 1-site (central LJ)
- Xenon :

	σ(Å)	ε(k)
O_2	3.36	120
Xe	3.96	228

- Carbon Dioxide: 3-site Harris Model

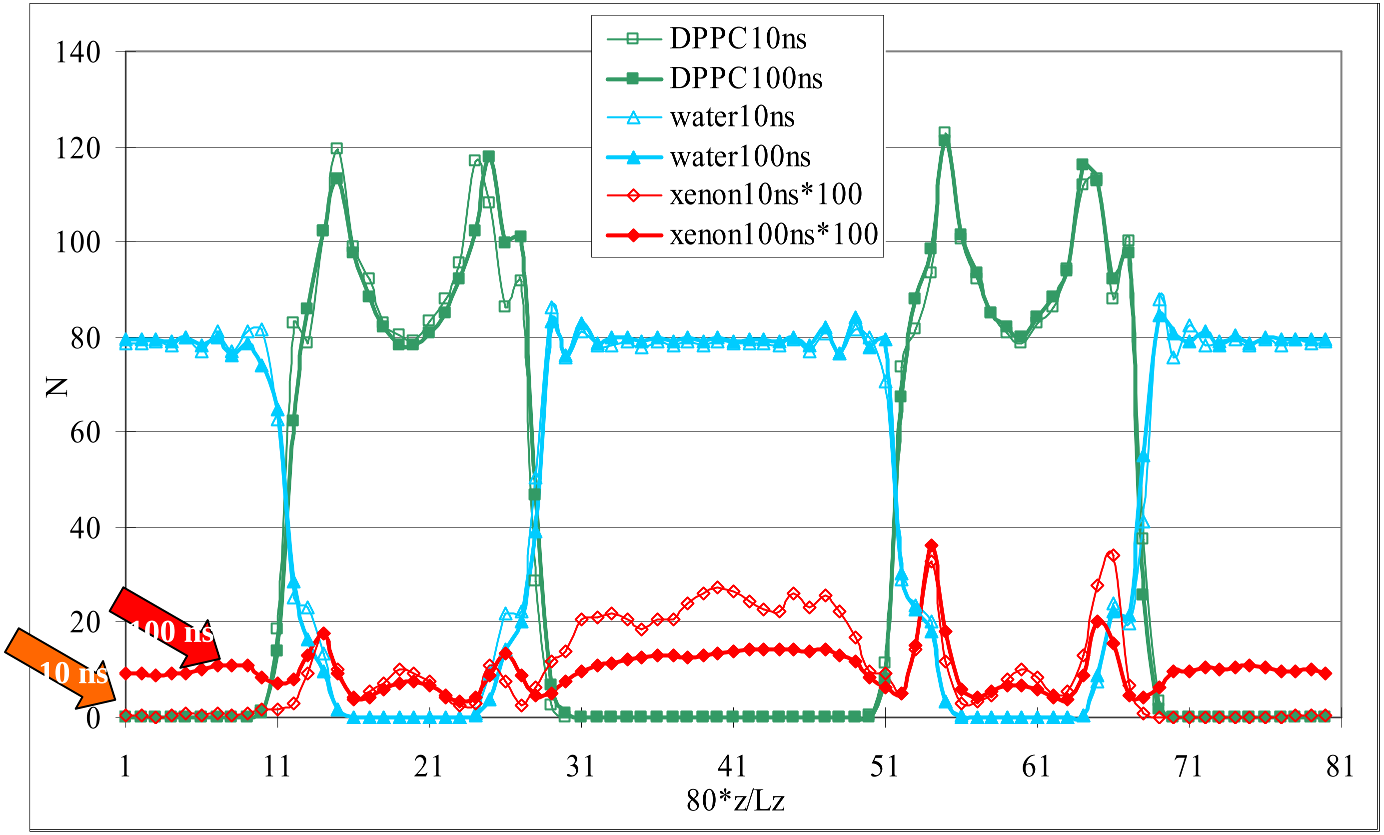
EPM2 Model (EPM rescaled to reproduce critical properties)					
	σ(Å)	ε(k)	Q (e)	lc-o	1.149 Å
C	2.757	28.129	+0.6512	k_θ	1236 KJ/mol /rad^2
O	3.033	80.507	-0.3256		

Gas permeation: **DPPC + O_2**, 323K



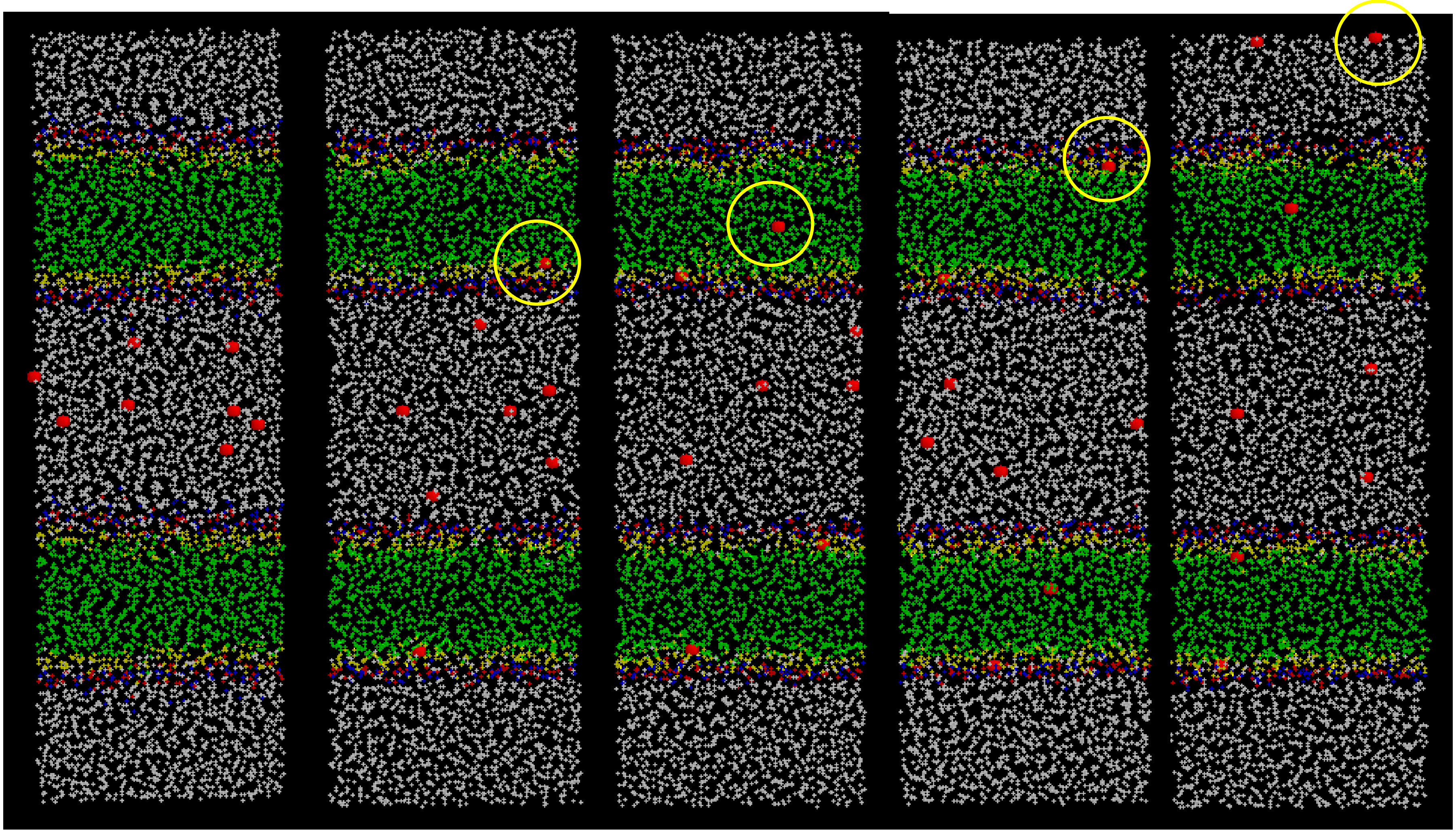
Empty symbol--density profile at 10 ns
Filled symbol --density profile at 100 ns

Gas permeation: **DPPC + Xe**, 323K



Empty symbol--density profile at 10 ns
Filled symbol --density profile at 100 ns

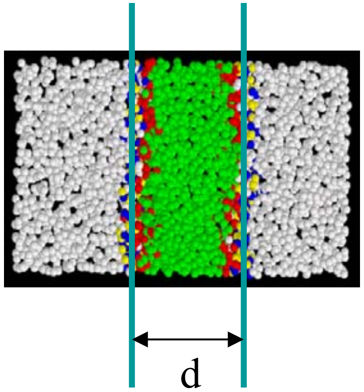
Observation of Xe atom permeation through DPPC lipid membrane



Investigation of permeation theory:

Model I: Inhomogeneous Solubility-Diffusion mechanism: 3 steps

- Step 1. Solute molecules are **partitioned** into the lipid membrane
- Step 2. Solute molecules **diffuse** in the membrane
- Step 3. Solute molecules **dissolved to the other water side** of the membrane



define permeation coefficient P:

$$\frac{1}{P} = \int_{z_1}^{z_2} R(Z)\mathrm{d}Z = \int_{z_1}^{z_2} \frac{\exp\{\Delta G(Z)/k_{\mathrm{B}}T\}}{D_z(Z)}\mathrm{d}Z.$$

such that $P \approx kD/d$

k = partition coefficient of solute between H_2O and membrane

D = diffusion coefficient of solute in membrane

d = membrane thickness

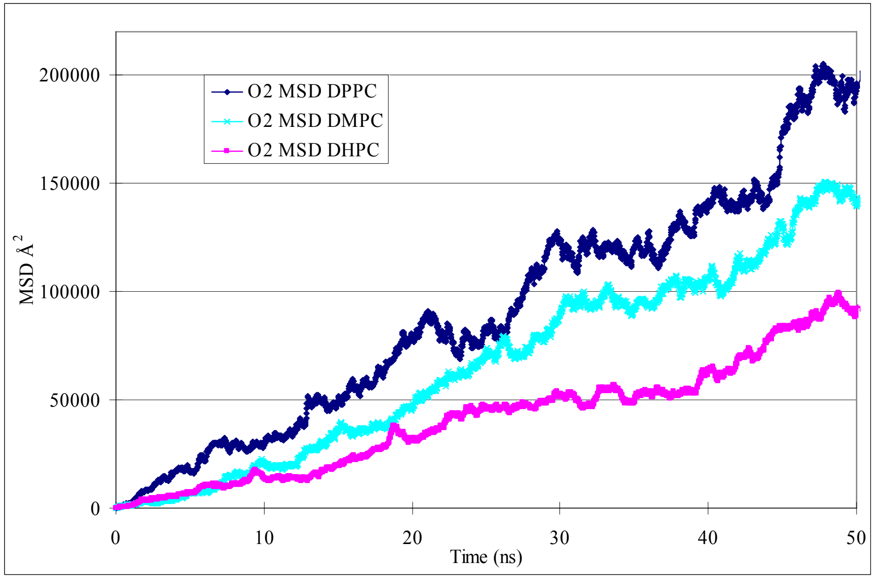
(Marrink & Berendsen)

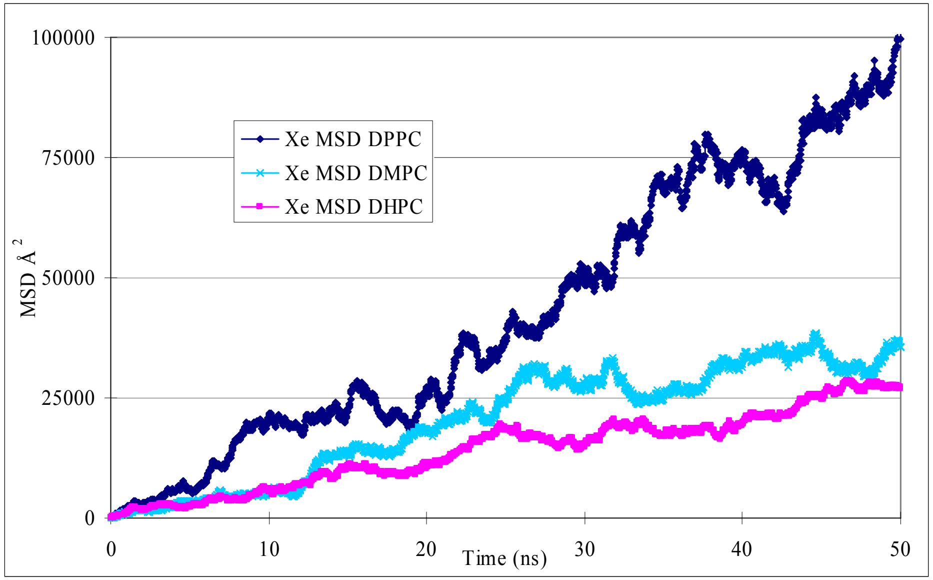
Diffusion coefficient usually obtained from MSD from MD simulations

- However, for this complex system, MSD is averaged over <u>both</u> the aqueous and membrane regions

Oxygen/Xe + DPPC/DMPC/DHPC MSD: 323K

MSD (Time averaged over both water + membrane)





$$D = \frac{1}{2 \times 3} \lim_{t \to \infty} \frac{\langle |r(t) - r(0)|^2 \rangle}{t}$$

Effective Diffusion Coefficient $D/10^{-5}$ cm^2/s			
	O_2	Xe	CO_2
DPPC+Water	**6.06**	**2.92**	**N/A**
DMPC+Water	**4.59**	**1.34**	**N/A**
DHPC+Water	**2.78**	**0.92**	**N/A**
AA model[1], POPE+Water	**2.14**		**1.8**
Expt[2], Bulk Water	**3**		**2.93**

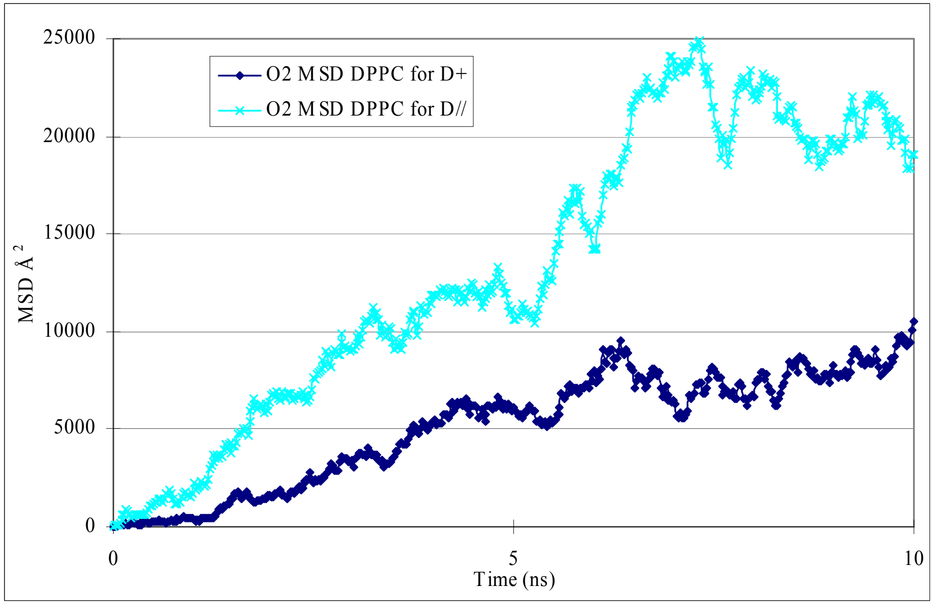
[1] Y.Wang et al, J.Structural Biology157(2007)534-544 [2] Tamimi et al J.Chem.Eng.Data 1994, 39,330-332

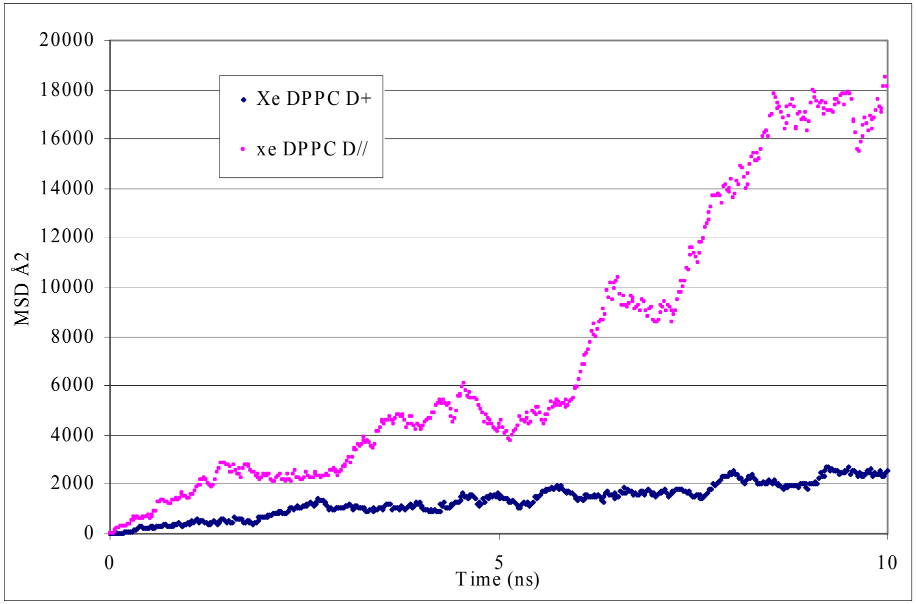
a more meaningful comparison: definition of permeability

$$P = \frac{D_\perp}{D_{//}}$$

$$D_\perp = D_z = \frac{1}{2\times 1}\lim_{t\to\infty}\frac{\langle |r(t)-r(0)|^2\rangle_z}{t}$$

$$D_{//} = D_{x+y} = \frac{1}{2\times 2}\lim_{t\to\infty}\frac{\langle |r(t)-r(0)|^2\rangle_{x+y}}{t}$$

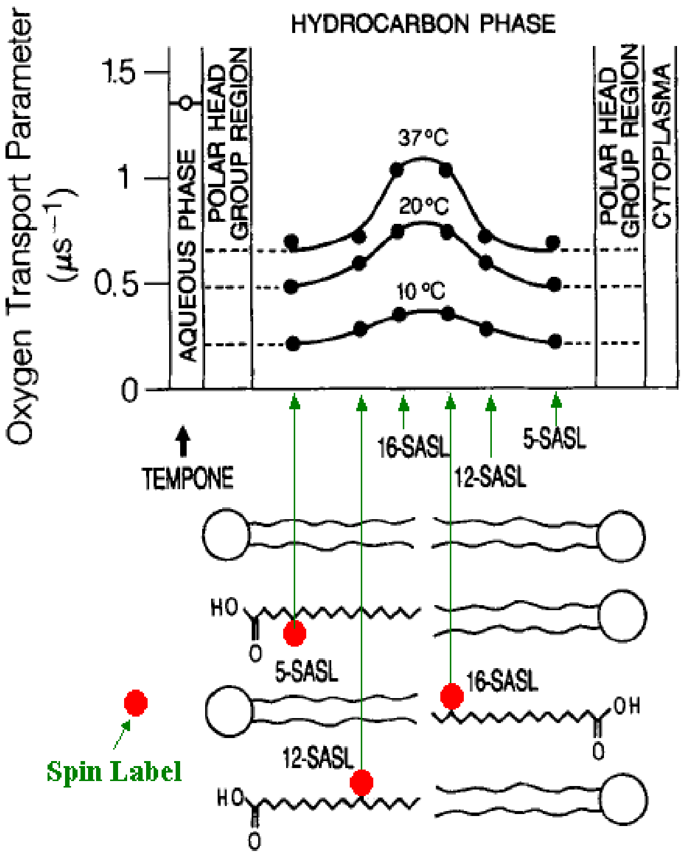




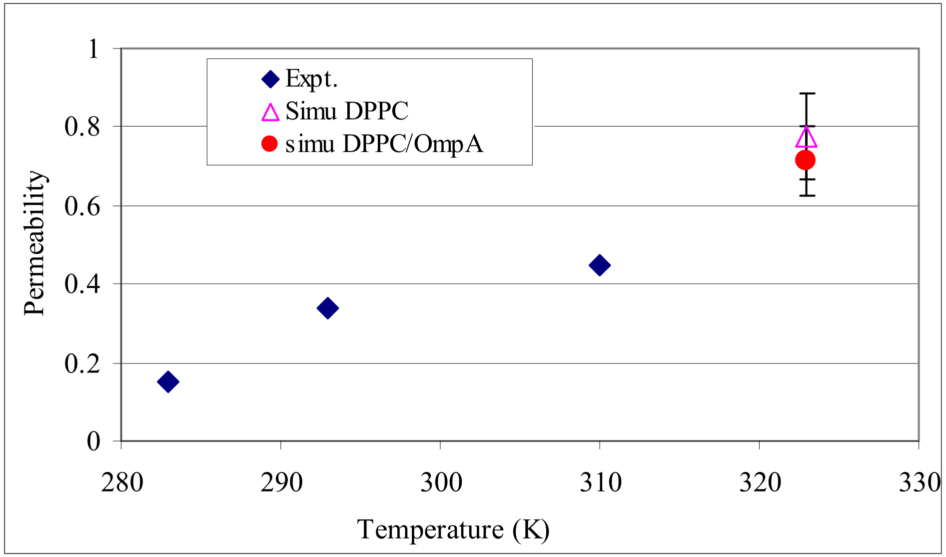
permeability for O_2 = 0.775

permeability for Xe = 0.334

Validation of permeability from simulations vs. experiment for O_2:



Ref: Witold Subczynski et al, J.Gen.Physiol Vol.100,69-87, 1992



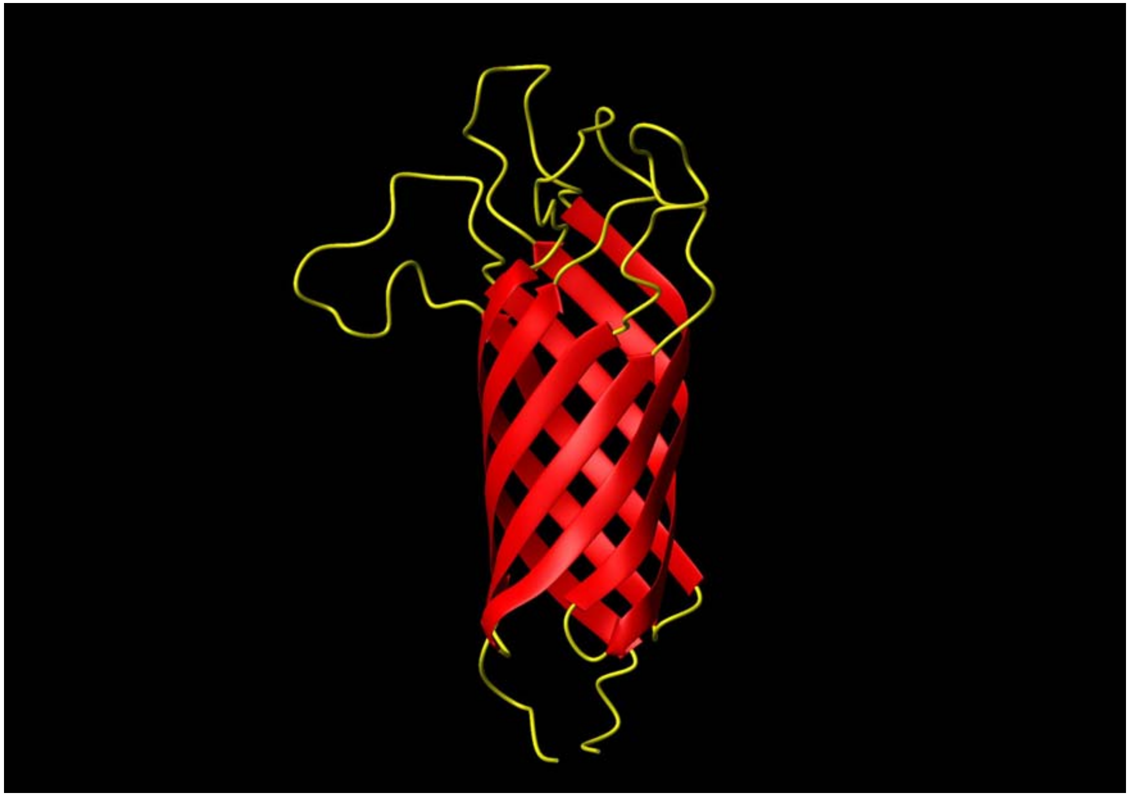
Our permeability for O_2 agrees well with the values obtained by EPR measurements

other proposed permeation model

- **Model II:** Transient pore model (Jansen & Blume): transport of water across a membrane largely occurs through transient pores in the lipid bilayer.
- Thermal fluctuations lead to the spontaneous formation of transient pores across the membrane, which are nothing more than fluctuating defects in membranes.

Transport through a membrane with embedded outer membrane protein

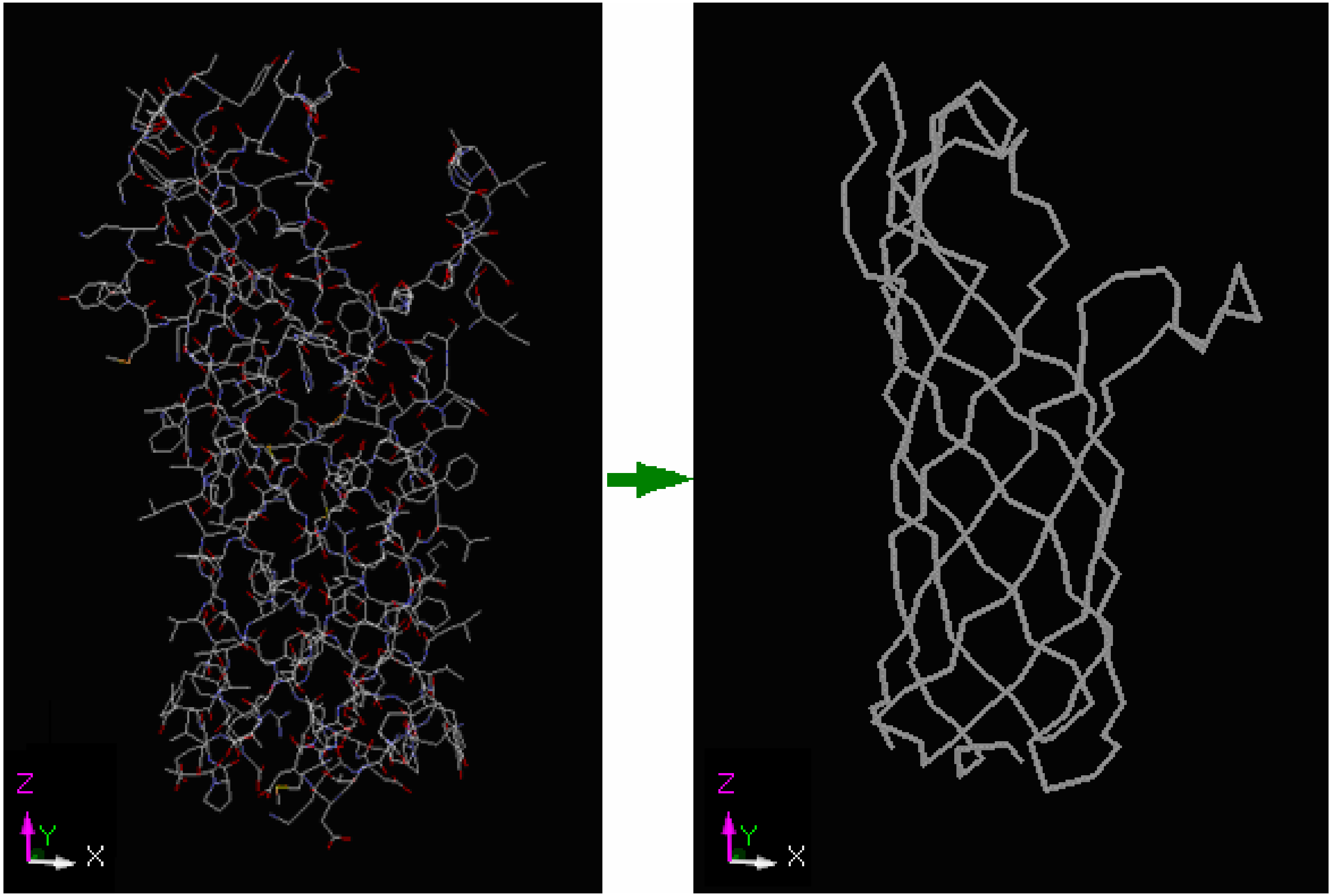
Outer membrane protein A (OmpA)



Arora et al. Nature Structural Biology **8**, 334 - 338 (2001)

- OmpA is a relatively simple bacterial outer membrane protein (from E coli) which has an antiparallel beta barrel domain
- has a well characterized structure
- considered a slow porin

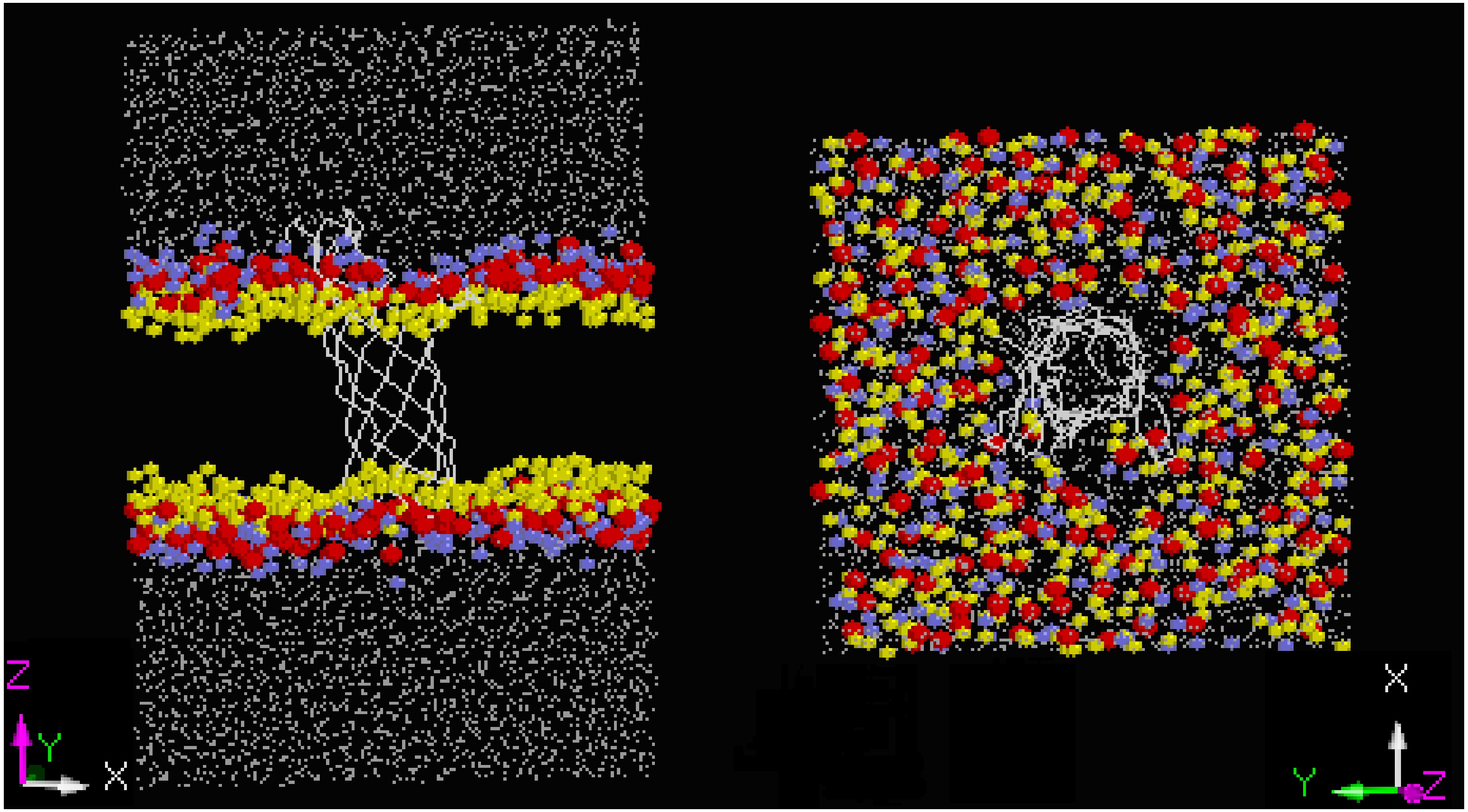
Coarse-grain mapping for OmpA



atomistic **coarse-grain**

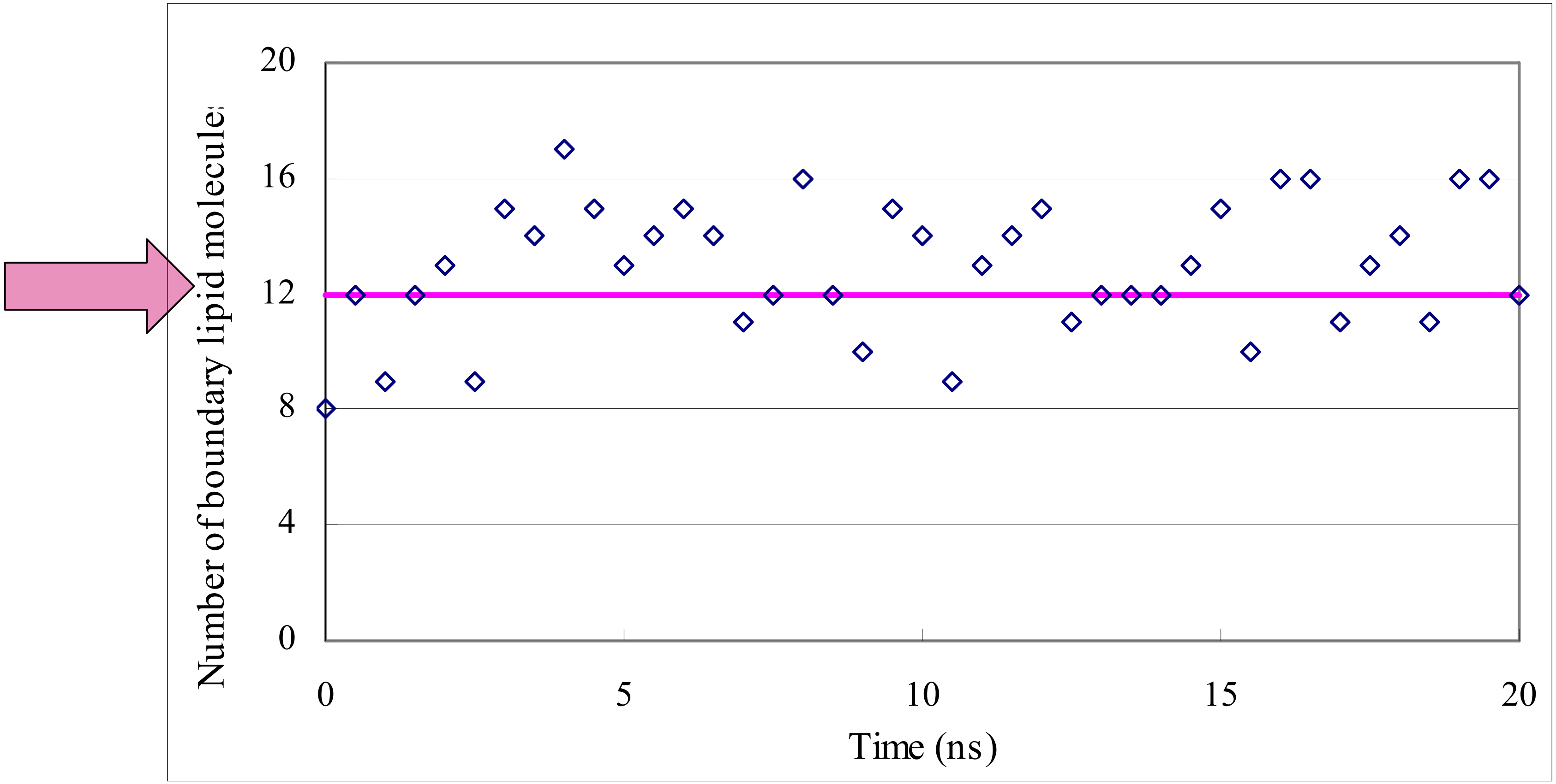
Use the MARTINI force field

Simulation system for permeability study: OmpA in DPPC membrane



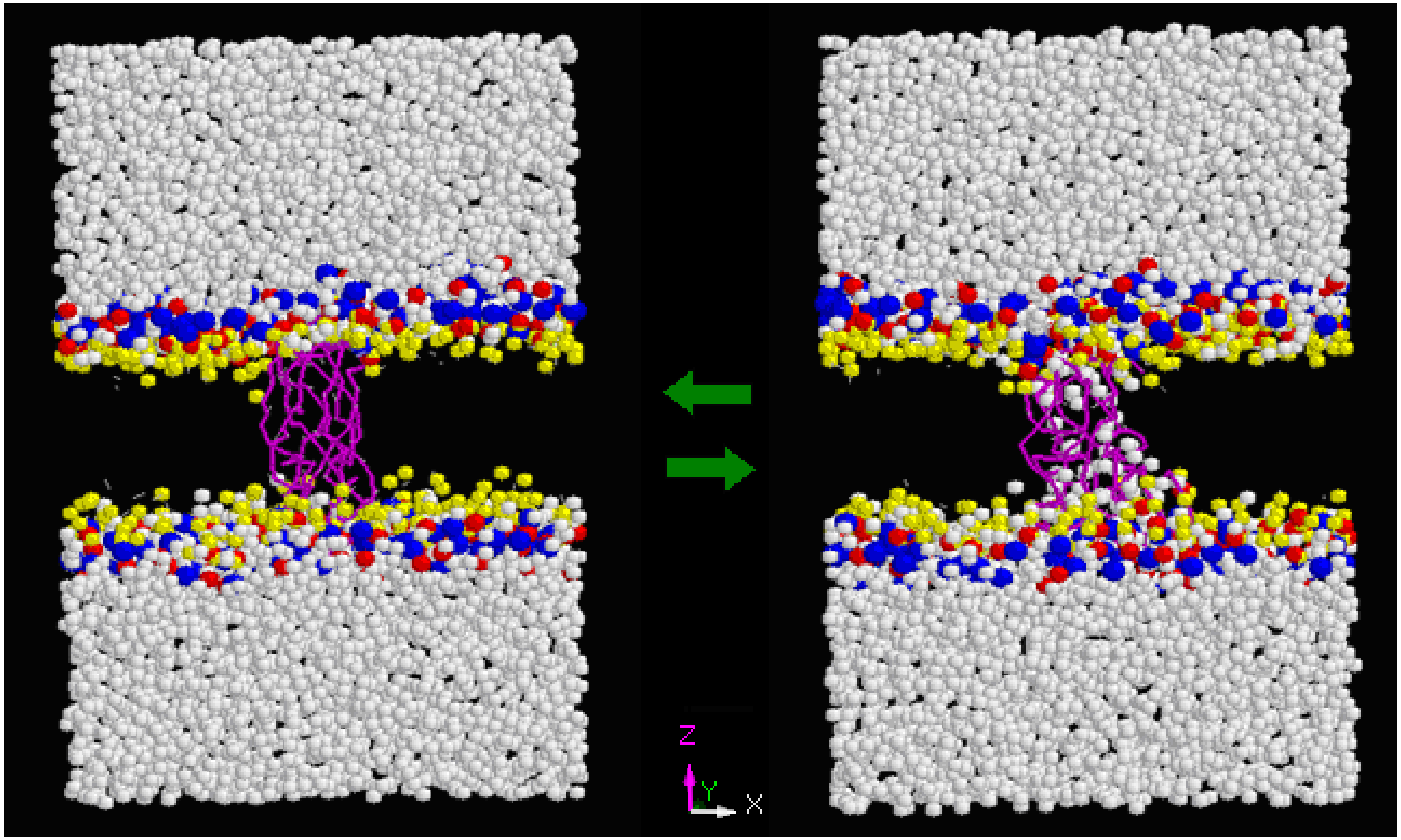
For DPPC, blue dots represent the choline group, red dots the phosphate group, yellow dots the glycol group

lipid protein interactions: average number of lipid molecules in contact with OmpA



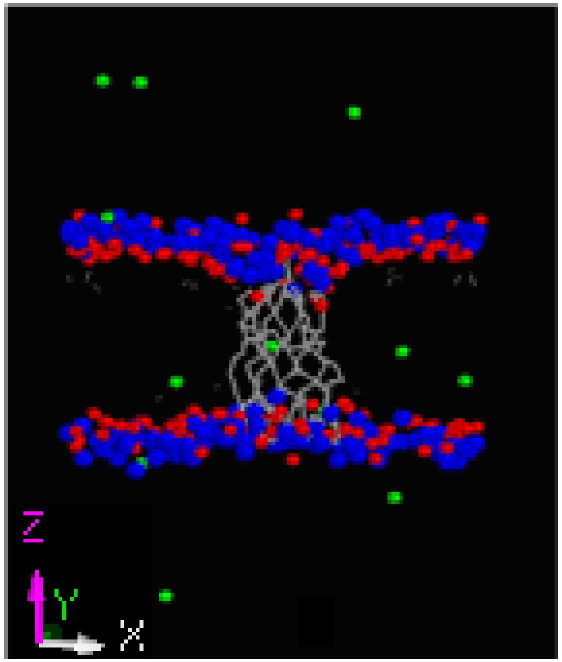
compare with experimental value of 11 found for OmpA in DMPglycerol

water channel formed by OmpA

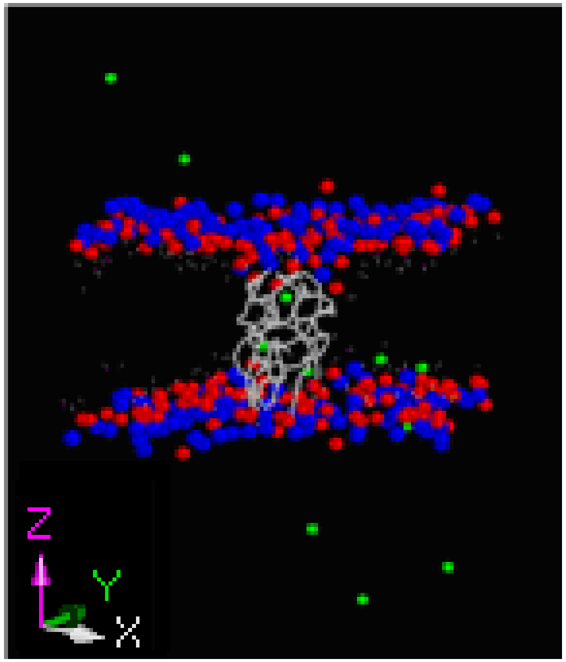


closed **open**

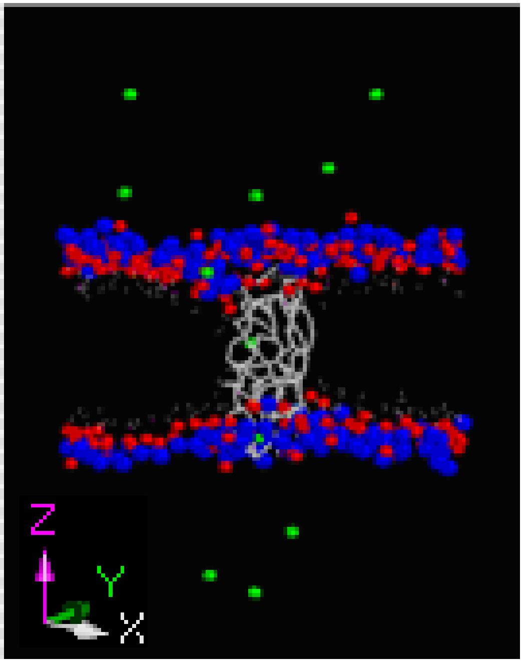
Snapshots of gas molecules in the simulation system



Xe

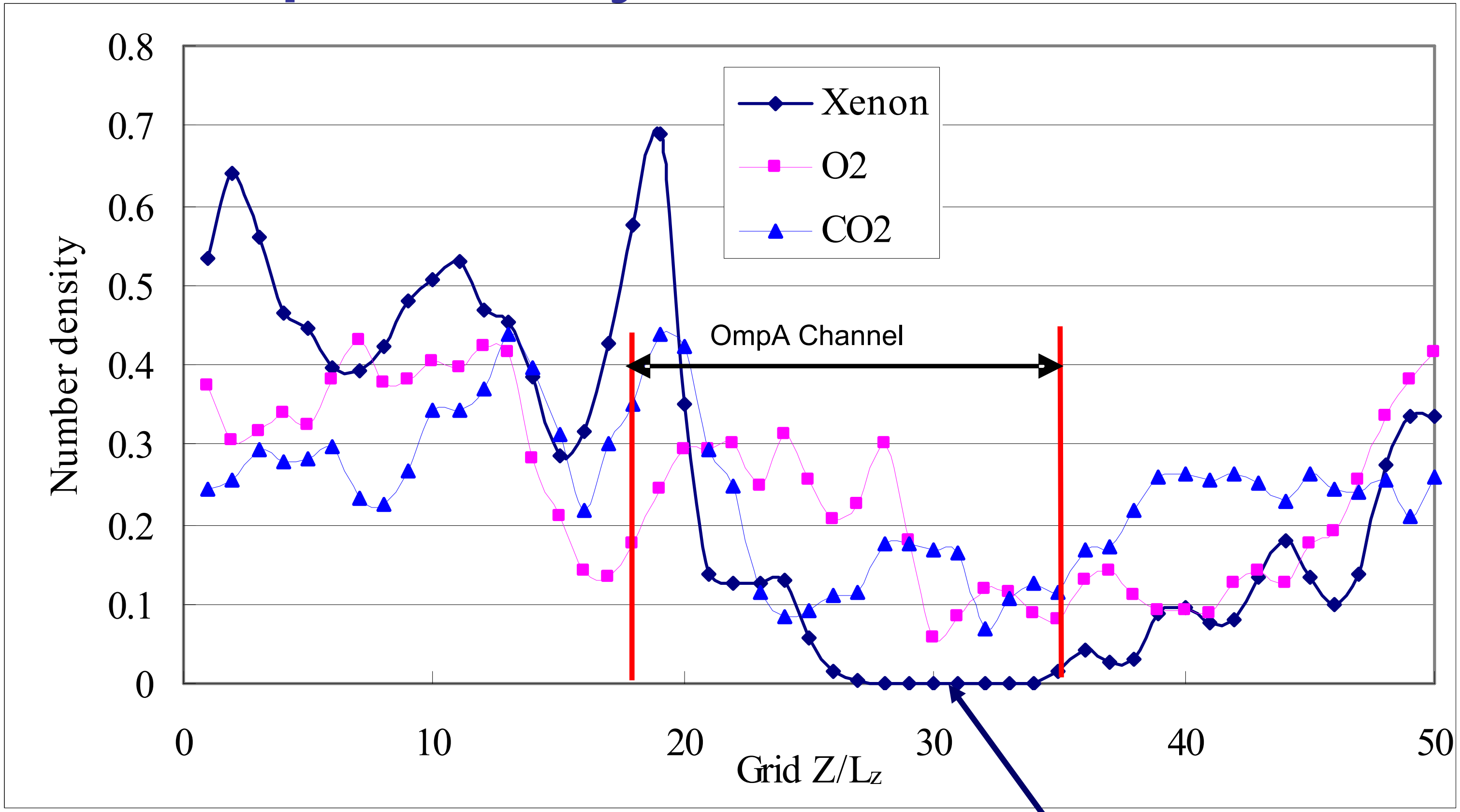


O_2



CO_2

density profiles of Xe, O_2 and CO_2 molecules along the direction normal to the lipid bilayer surface



Xe does not permeate through the OmpA channel

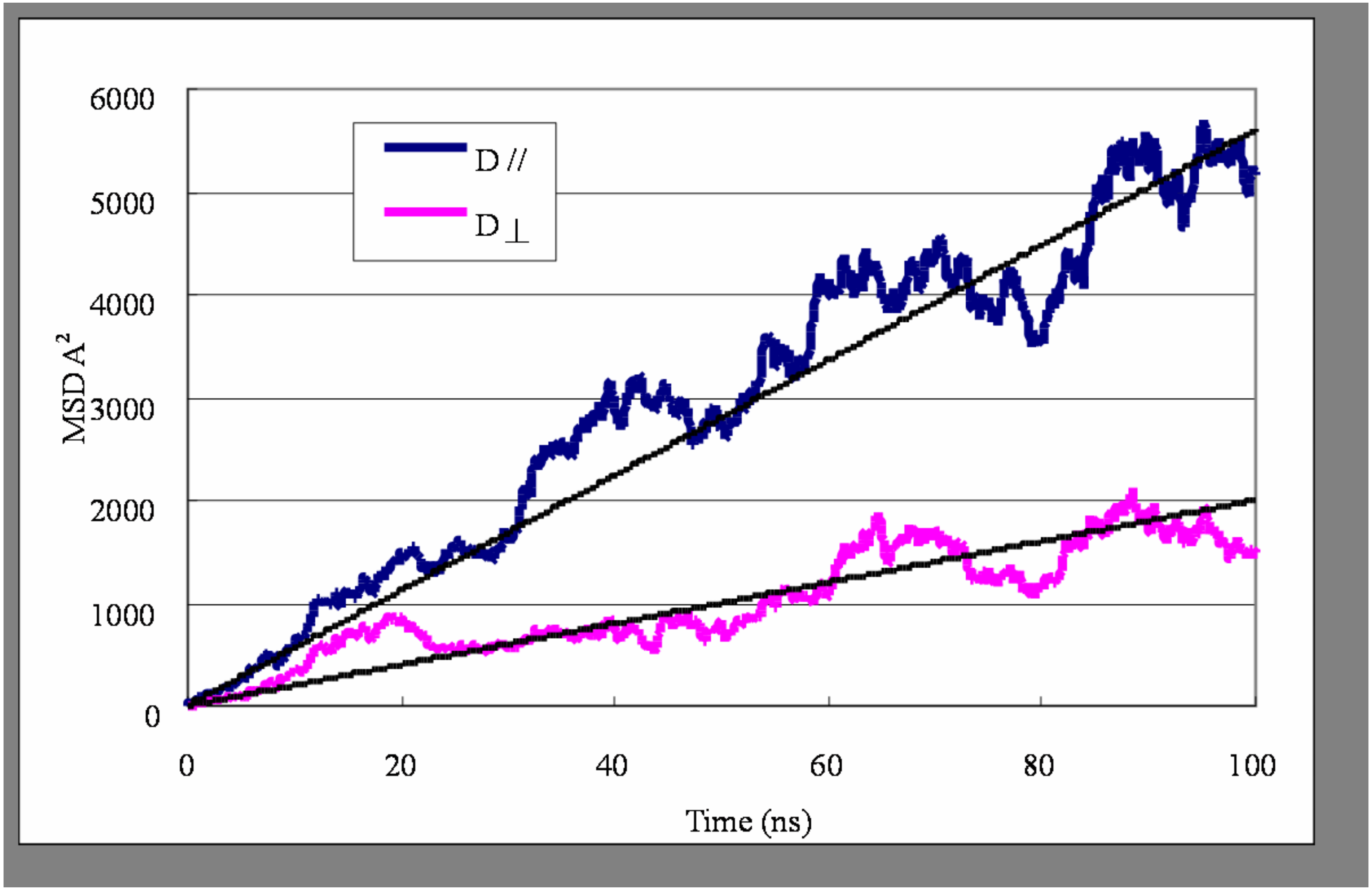
Xe permeates through lipid bilayer, not through OmpA

Gas	Permeation through OmpA (%)	Permeation through DPPC lipid bilayer (%)
Xe	0.0	100.0
CO_2	53.8	46.2
O_2	62.5	37.5

from counting permeation events

Xe does not permeate through the OmpA channel

mean square displacements for permeability calculation



Permeability of gas molecules through membrane = $\mathbf{D}_{\perp} / \mathbf{D}_{\parallel}$

Gas	Permeability through DPPC	Permeability through DPPC with OmpA
Xe	0.334 ± 0.06	0.548 ± 0.04*
CO_2	0.464 ± 0.07	0.65 ± 0.10
O_2	0.775 ± 0.11	0.714 ± 0.09

*lipid bilayer with embedded OmpA has a more permeable structure than lipid bilayer without OmpA.

Summary

- Studied the transport process of different small molecules through a lipid membrane.
- Compared diffusion coefficients and permeability of different gases through different lipid membranes and through lipid membranes embedded with an outer membrane protein, OmpA.
- Compared our coarse-grain MD simulation results with atomistic simulations and with experiments.

CONCLUSIONS

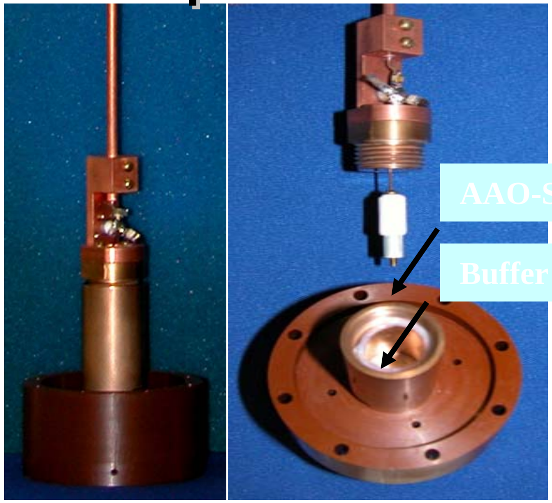
- The observed xenon permeability increase in the presence of OmpA is probably attributable to the change in lipid structure near the protein lipid interface. Since all xenon atoms permeate through the lipid bilayer, which depends on the dynamically formed defects at the water-lipid surface. The frequency of dynamically formed pores is affected by the protein OmpA embedded in the membrane.

CONCLUSIONS

- We validate our lipid membrane model with several tests: self-assembly, area per lipid, phosphate to phosphate distance across, density profile along the normal to the bilayer, order parameter of the tail segments.
- The MARTINI force field can be successfully used to simulate small molecule permeation in lipid bilayer membranes and in OmpA-embedded membranes.

Future:

- Study other gas molecules: N_2, CO, NO, to investigate the effect of different gas properties on permeability through lipid membranes
- Self assemble pore-formers into lipid bilayer model and observe transport in MD
- Combine with NMR experimental (Rex E. Gerald) measurements to explore transport processes



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Ion transport
Hongmei Liu

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Molecular transport across lipid bilayer model membrane
Huajun Yuan

DEPARTMENT OF ENERGY · UNITED STATES OF AMERICA