NRC·CNRC

From Discovery to Innovation...

Lipid areas obtained from the simultaneous analysis of neutron and X-ray scattering

Norbert Kučerka, Mu-Ping Nieh and John Katsaras

Canadian Neutron Beam Centre
Chalk River, Ontario

NRC·CNRC

From Discovery to Innovation...

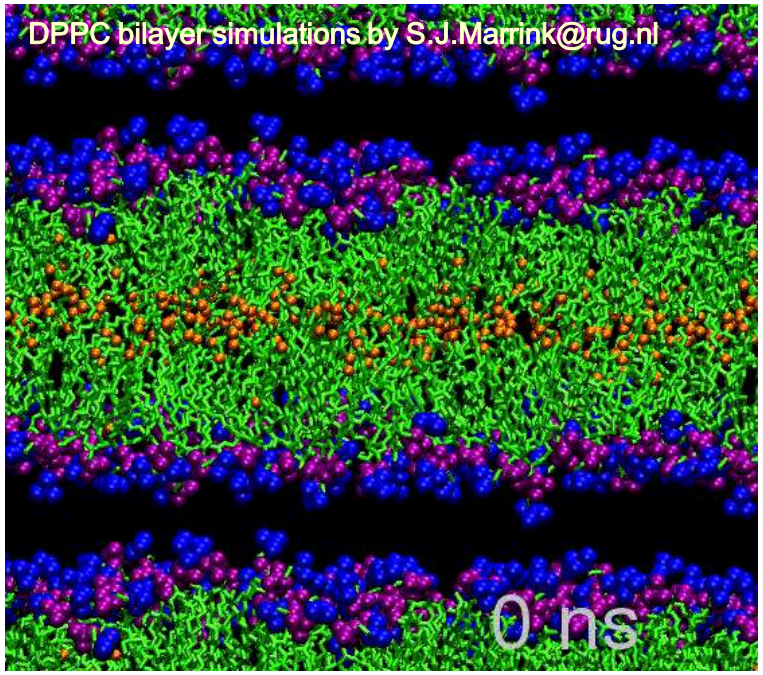
Outline

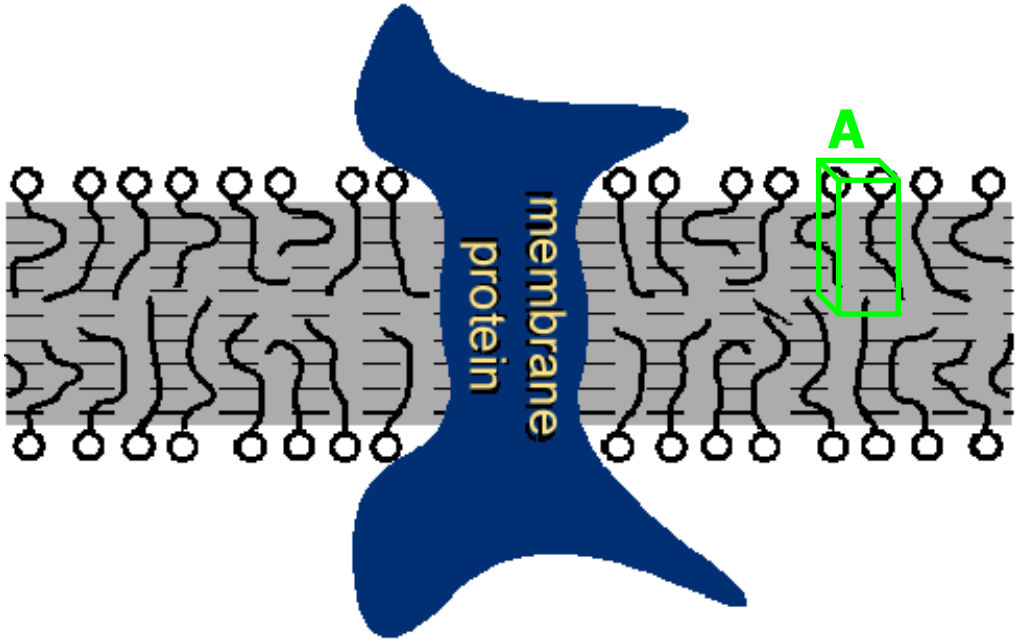
- Lipid Bilayers as Biomimetic Systems
- X-ray Scattering
- Neutron Scattering
- Reconciling X-ray and Neutron Scattering
- Lipid Areas for Various Lipids
- SIMulation to EXPeriment Comparison

NRC·CNRC
From Discovery to Innovation...

Biomimetic Systems

- lipid matrix is a 2D liquid, where lipids and proteins diffuse almost freely
- the complex structural dynamics of membranes involve a balance of forces
- lipid structural parameters determine the specific biomembrane functions
- knowledge of lateral lipid area is central to MD simulations





Quantitaive results

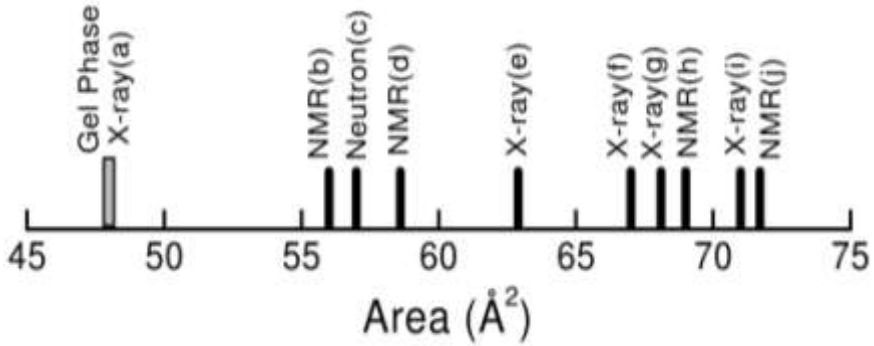
- MD simulations, due to non-perfect force fields, are carried out at a fixed area per lipid.

What value should be used?

- Despite their central role in membrane biophysics, values of lateral areas for lipid molecules had been very uncertain.

Literature Results for DPPC @ 50°C:

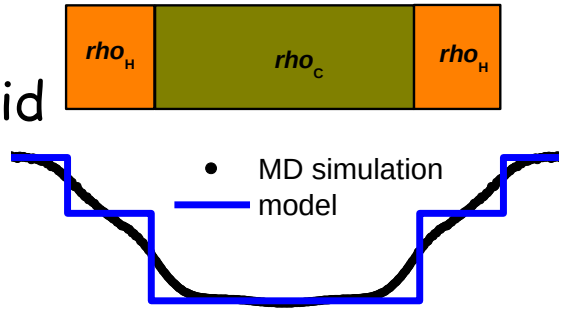
(Nagle and Tristram-Nagle, BBA 2000)

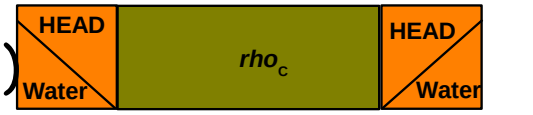


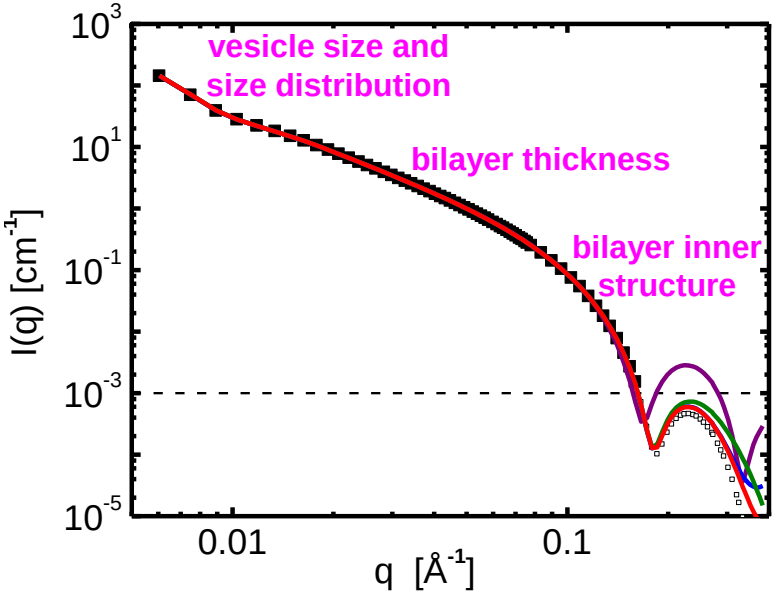
Chaos for theory/simulations

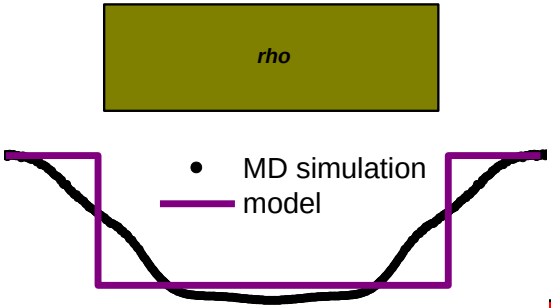
Neutron scattering simple/advanced models

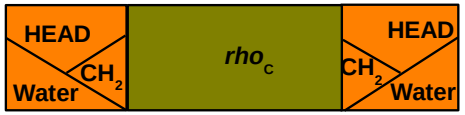
- The neutron scattering length density profiles of fluid bilayers in solution are inherently quite featureless (compared to X-ray scattering profiles)
- Nevertheless, the mid-*q* region provides high quality information, reflecting the large scattering contrast between the lipid bilayer (a lot of H) and solvent (D_2O)

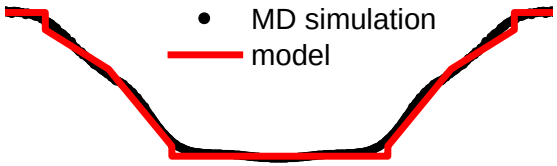






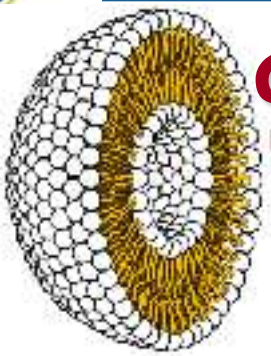


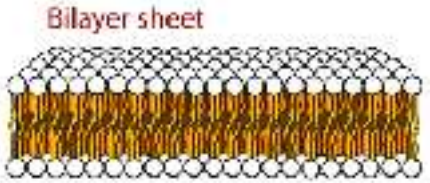




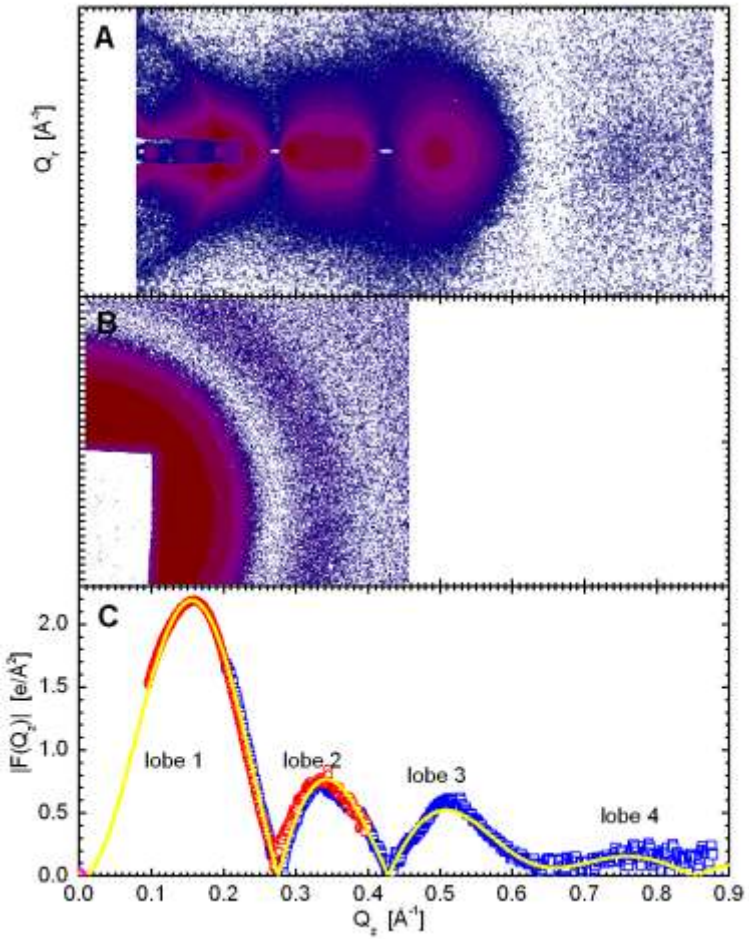
N. Kučerka, J.F. Nagle, S.E. Feller and P. Balgavý, Phys Rev E 69, 051903 (2004)

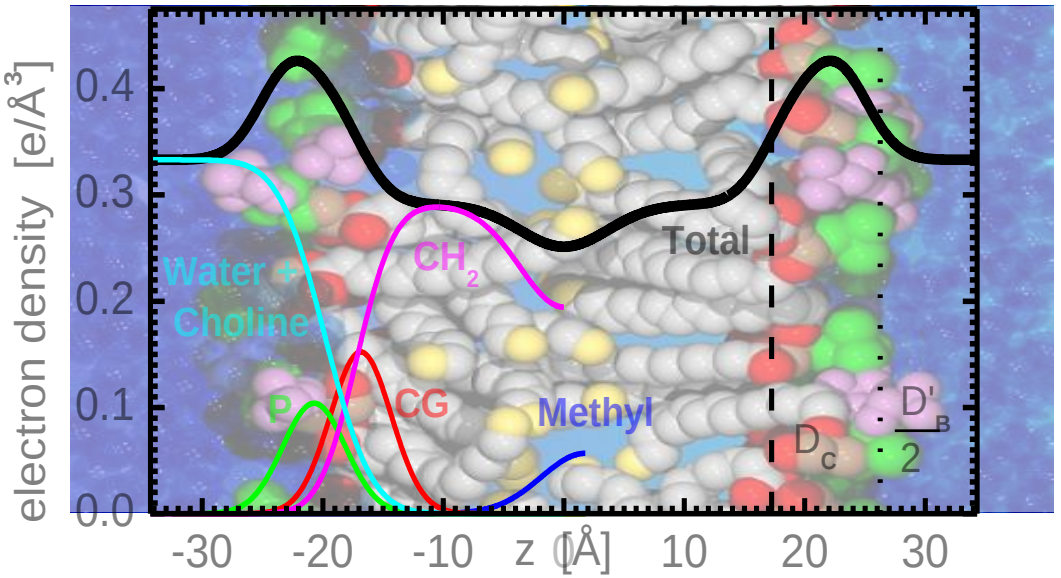
X-ray scattering advanced models





A combined global analysis approach takes advantage of the complementarity of ULVs and oriented samples, enhancing the spatial resolution of the bilayer structure.





DLPC, DMPC: N. Kučerka, Y. Liu, N. Chu, H. I. Petrache, S. Tristram-Nagle, and J.F. Nagle, Biophys. J (2005)
DOPC, POPC, DEPC: N. Kučerka, S. Tristram-Nagle, and J.F. Nagle, J Mem Biol (2005)
DPPC: N. Kučerka, S. Tristram-Nagle, and J.F. Nagle, Biophys J Lett (2006)

X-ray vs. Neutrons

Significant differences remain when comparing lipid areas determined from X-ray and neutron scattering experiments!

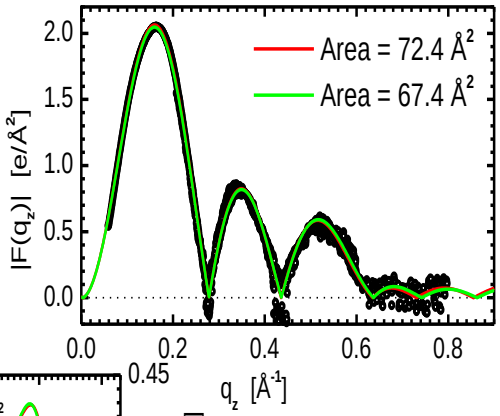
e.g., DOPC @ 30°C:

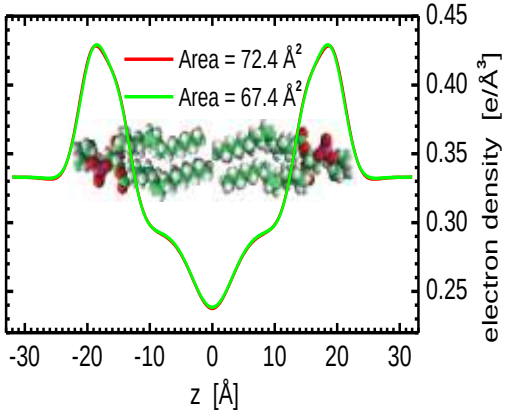
$A_{X\text{-}ray} \sim 72$ Å^2

$A_{neutrons} \sim 67$ Å^2

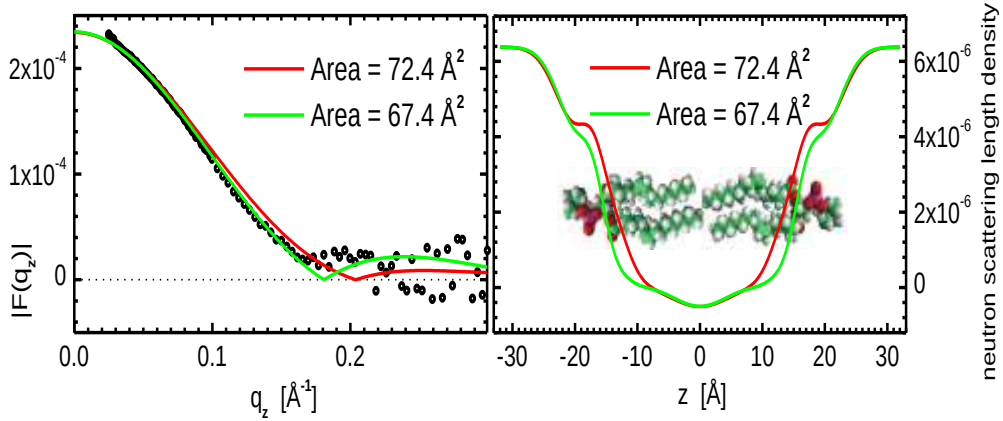
X-ray vs. Neutrons

- X-ray scattering is most sensitive to the electron dense headgroup peaks
- But not to the lipid area





- Neutron scattering, especially from fully protonated lipid in D_2O, is most sensitive to the bilayer thickness which directly relates to the area

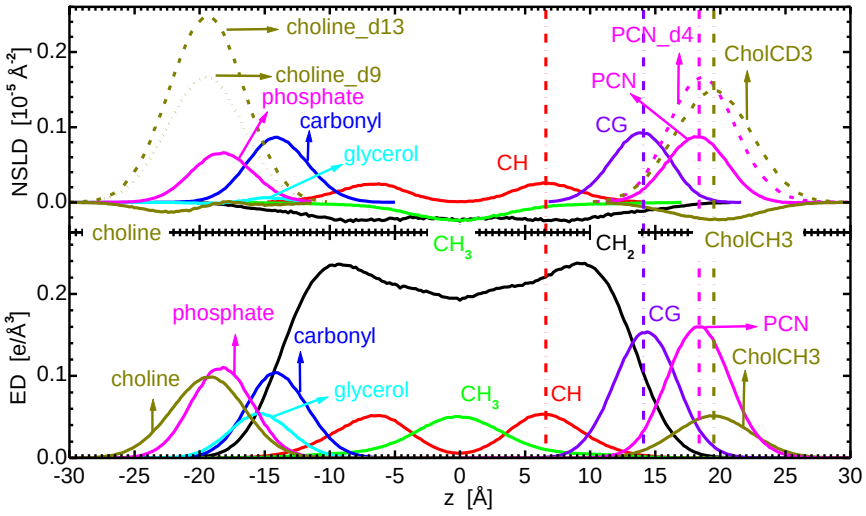


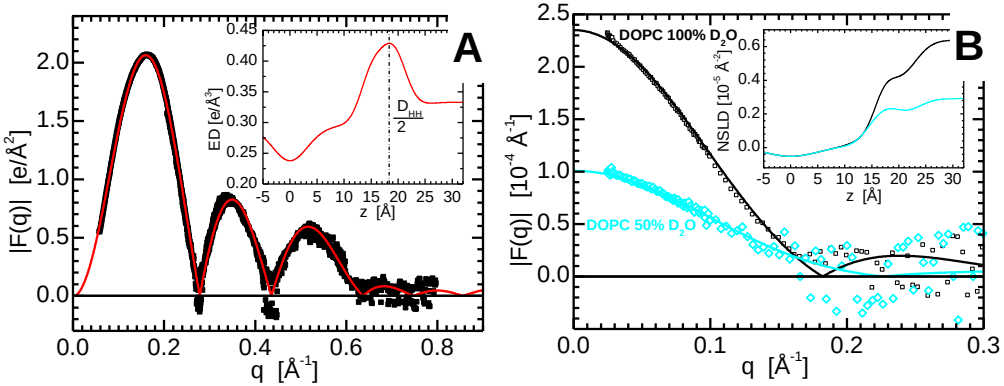
Simultaneous analysis of x-ray and neutron scattering data should allow for the inclusion of fine details, and better determination of the overall bilayer parameters.

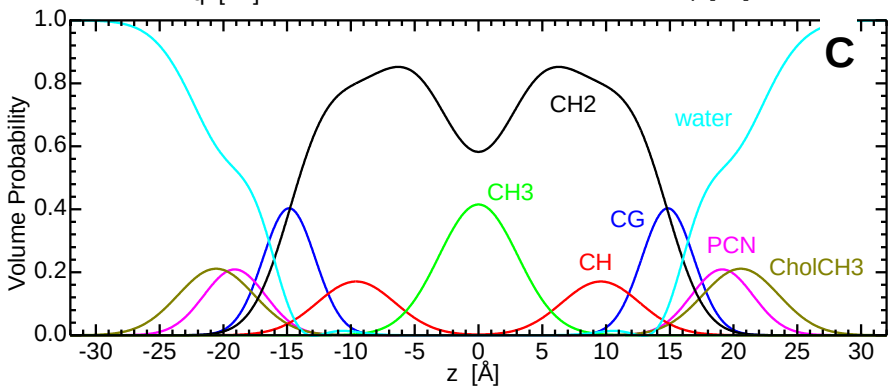
Universal model of Scattering Density Profile

- Each of the component groups has nearly the same functional form for all of the different contrast conditions
- Volume distributions satisfy a spatial conservation principle

The SDP model was fit (with only one set of parameters) simultaneously to the set of scattering data obtained at different contrast conditions (X-rays and neutron contrast variation).





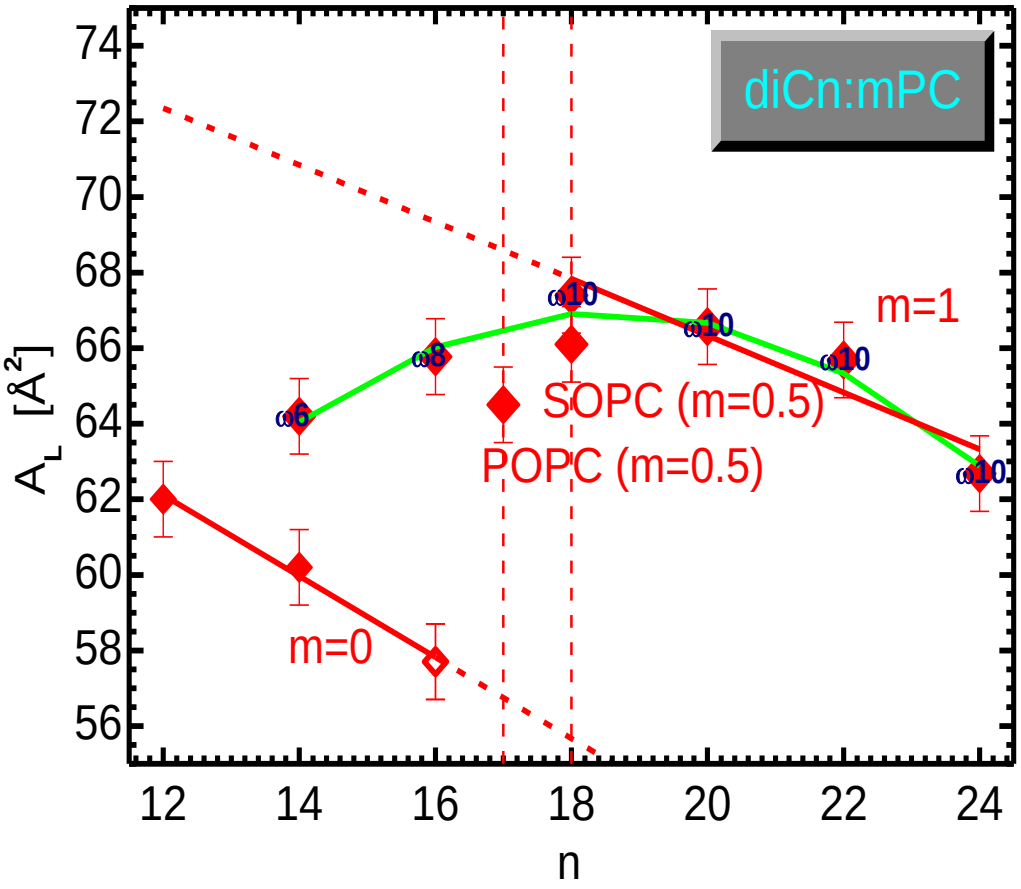


N.Kučerka, J.F.Nagle, J.N.Sachs, S.E.Feller, J.Pencer A.J.Jackson, and J.Katsaras, Biophys. J (2008)

Areas for Various Lipids

- chain length n (carbons)
- unsaturation m

(0 - fully saturated, 1 – di-monounsaturated, 0.5 – mixed chains)



1.) Lipid area increases with the increasing chain length:
-linearly in a case of saturated chain lipids and
-non-linearly in the case of lipids with di-monounsaturated chains when the double bond is fixed relative to the bilayer centre

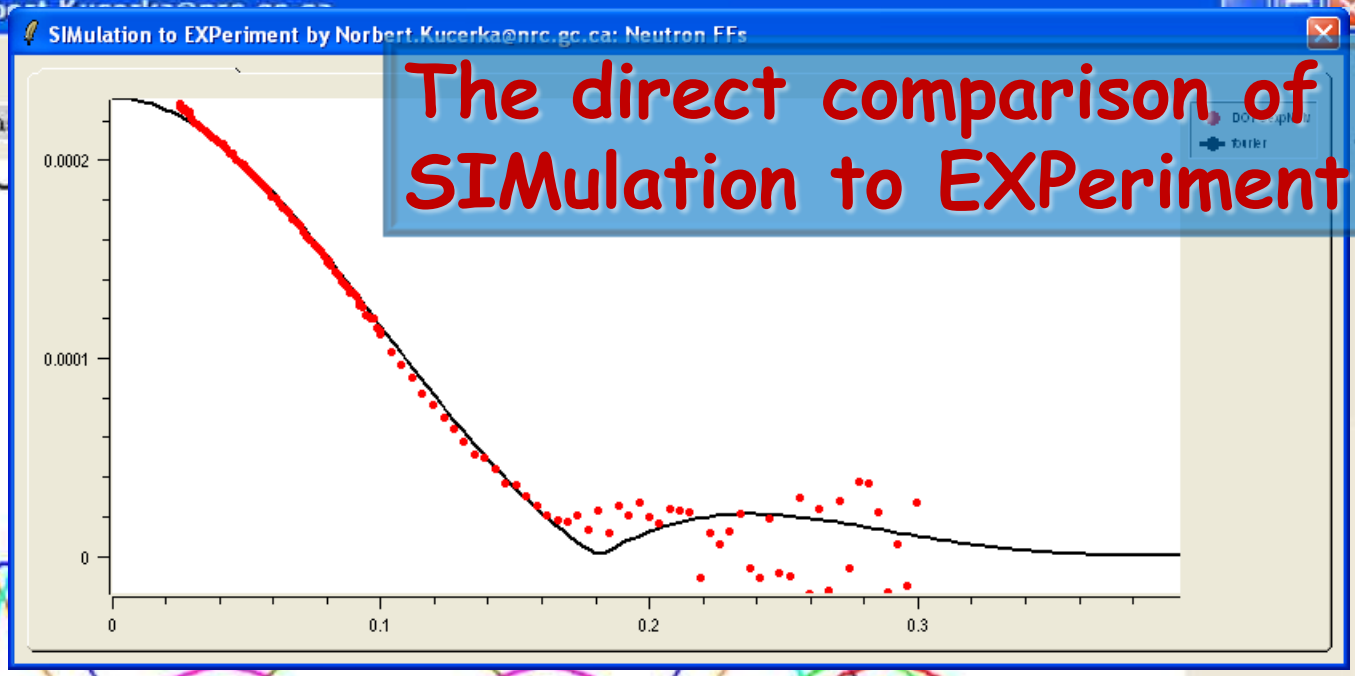
2.) Lipid area increases with the introduction of first double bond much more significantly than with the addition of double bond to the second chain

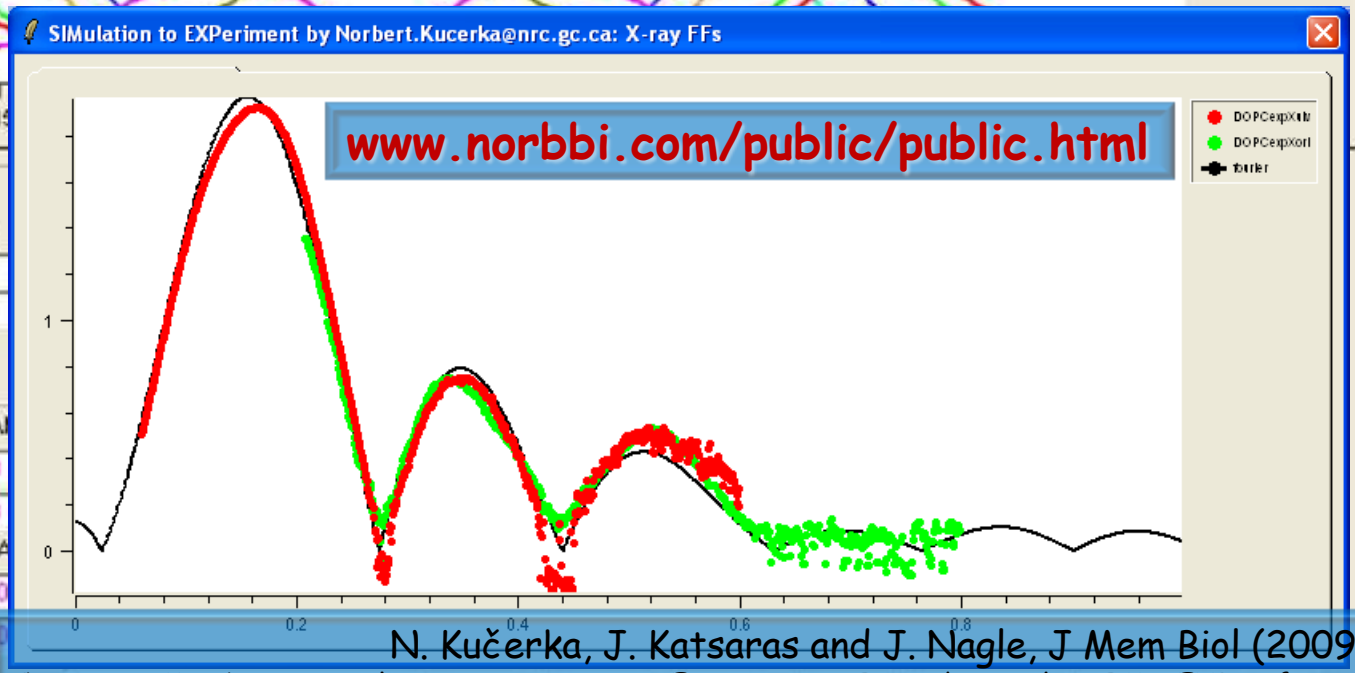
N. Kučerka, S. Tristram-Nagle, and J.F. Nagle, J Mem Biol (2005)
N.Kučerka, J. Gallová, D. Uhríková, P. Balgavý, M. Bulacu, S.-J. Marrink, and J.Katsaras, Biophys. J (2009)

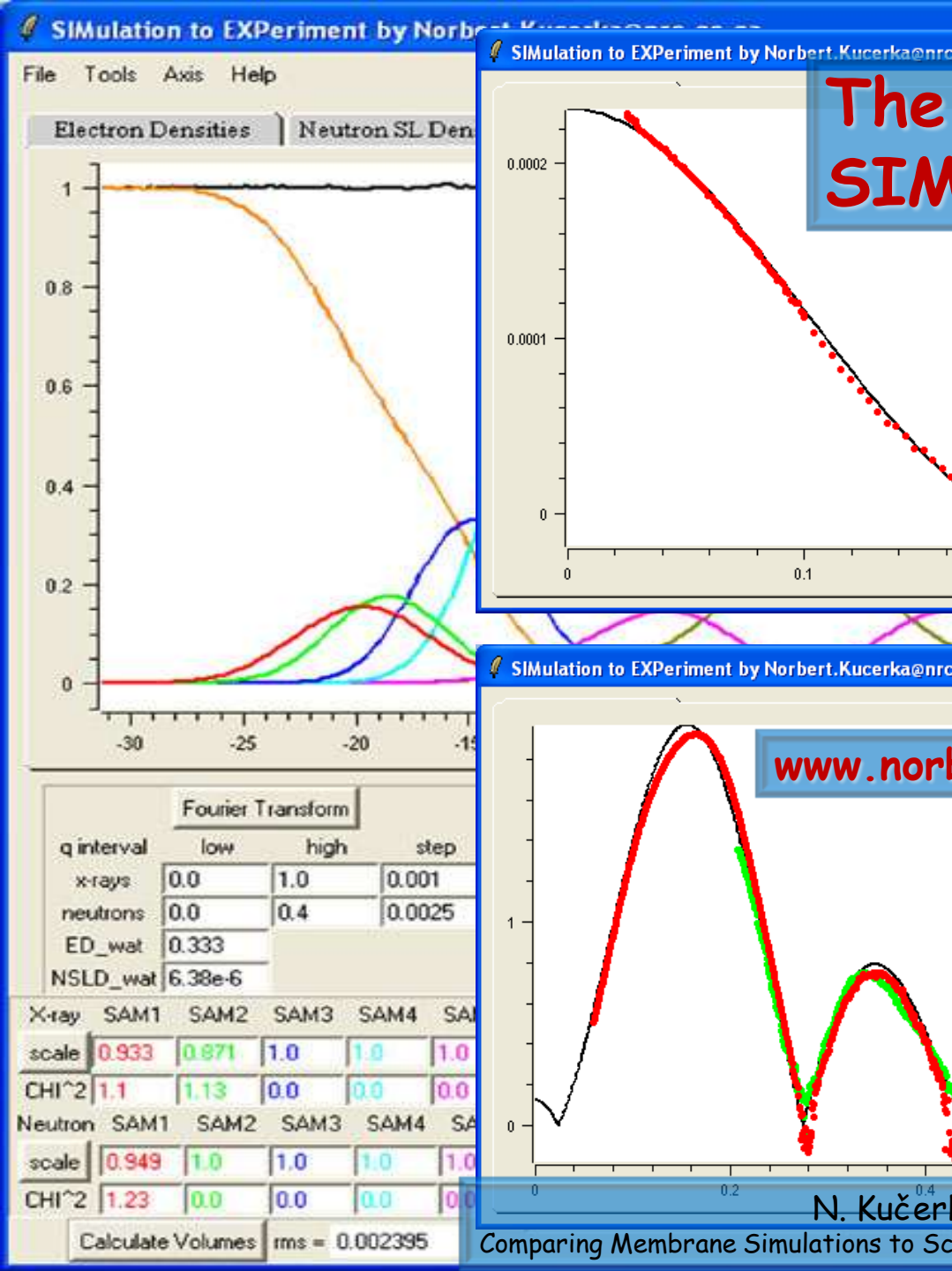
Concluding remarks

- Neutron scattering techniques are proving more and more their importance in the fields of structural biology and biophysics.
- Recent development has reconciled the differences between small-angle X-ray and Neutron scattering experimental results.
- MD simulations complement experimental results and experimental results provide guides for MD simulations.

The direct comparison of SIMulation to EXPeriment







N. Kučerka, J. Katsaras and J. Nagle, J Mem Biol (2009)

Comparing Membrane Simulations to Scattering Experiments: Introducing the SIMtoEXP Software

NRC·CNRC

From Discovery to Innovation...

Acknowledgement and Collaborators

- SDP model development
  - John Nagle (CMU, Pittsburgh, PA)
- Small-Angle Neutron Scattering
  - Andrew Jackson (NIST, Giathersburg, MD)
  - William Heller (ORNL, Oak Ridge, TN)
- Low-Angle X-ray Scattering
  - Arthur Woll (CHESS, Ithaca, NY)
  - Stephanie Tristram-Nagle (CMU, Pittsburgh, PA)
- Molecular Dynamics simulations
  - Jonathan Sachs (U of Minnesota, Minneapolis, MN)
  - Scott Feller (Wabash College, Crawfordsville, IN)
  - Siewert-Jan Marrink (U of Groningen, Netherlands)