



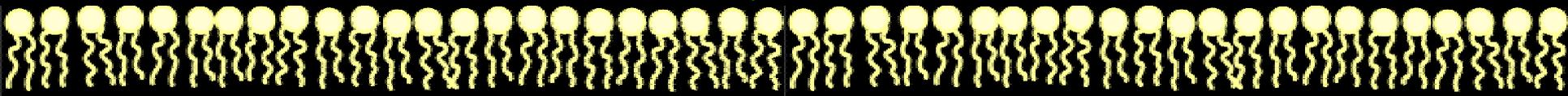
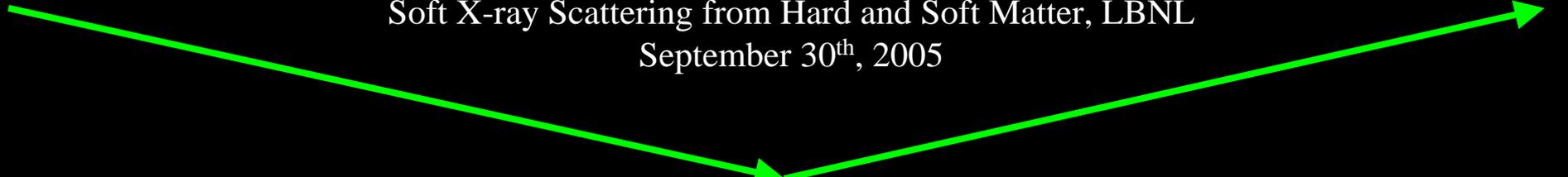
# Probing Lipid Membrane Structure by X-ray and Neutron Scattering

**Tonya Kuhl & Jarek Majewski**

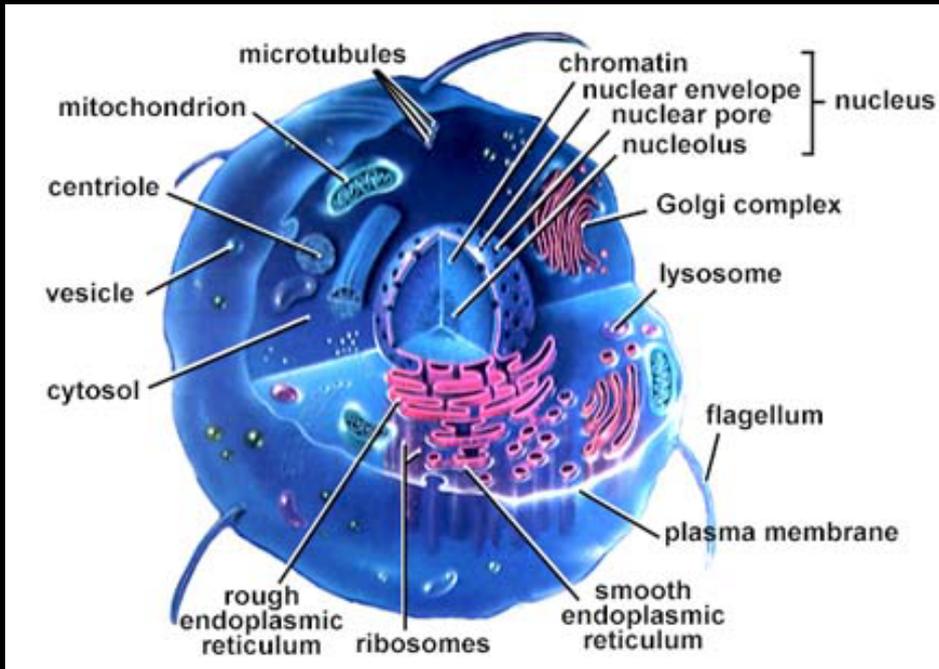
UC Davis/Los Alamos Neutron Scattering Center

[tlkuhl@ucdavis.edu](mailto:tlkuhl@ucdavis.edu)/[jarek@lanl.gov](mailto:jarek@lanl.gov)

Soft X-ray Scattering from Hard and Soft Matter, LBNL  
September 30<sup>th</sup>, 2005

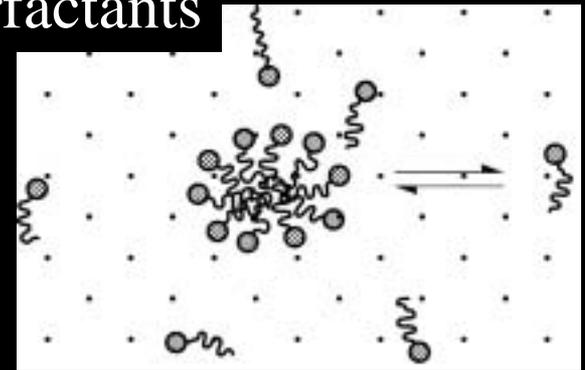


# Biophysics of the Cell Membrane

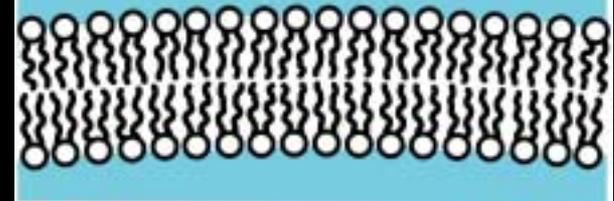


## Self-Assembly

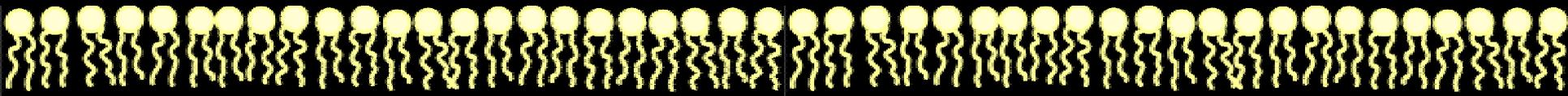
surfactants



## Lipid Bilayer

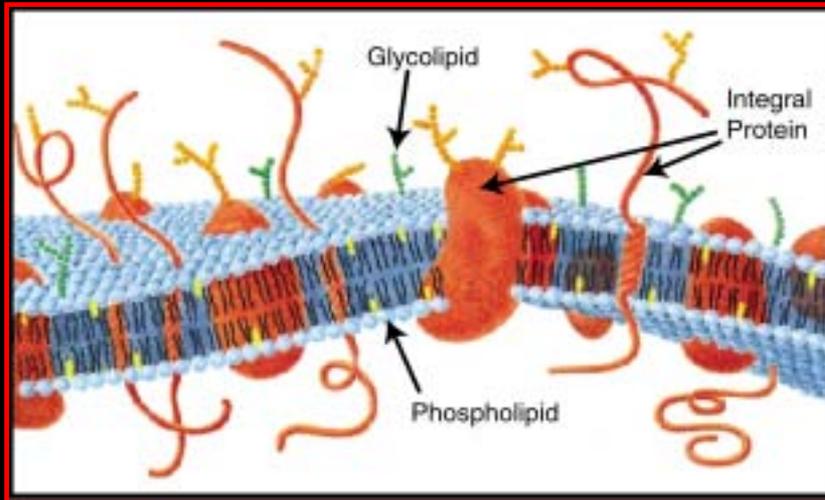


**Membranes are where the  
ACTION takes place!**

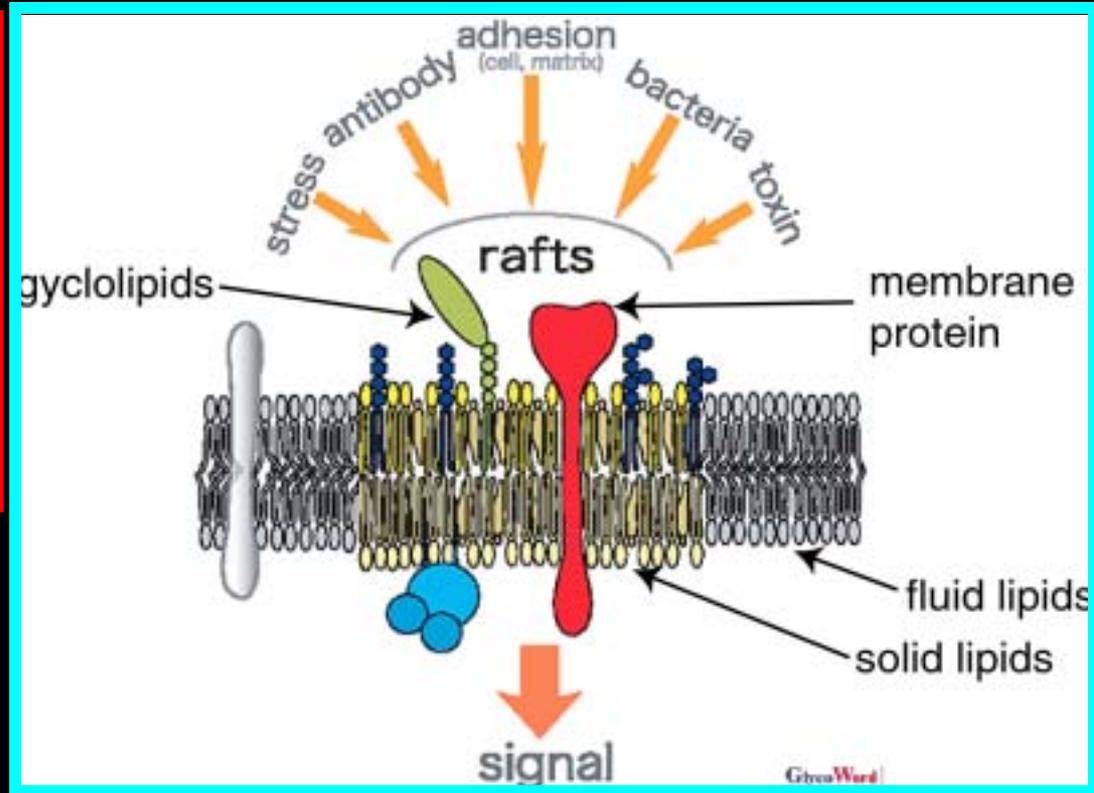


# Homogeneous vs Heterogeneous

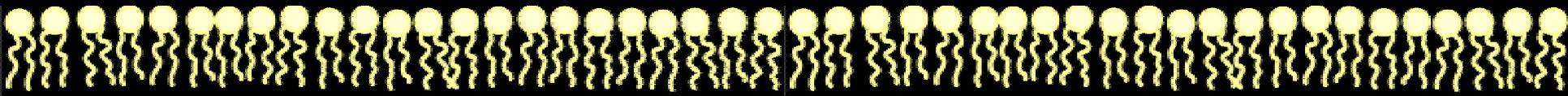
Fluid mosaic model vs. Lipid domains – “rafts” *Since 2000, over 2500 articles*



Cell membranes are no longer thought of as simply passive 2-D liquids.

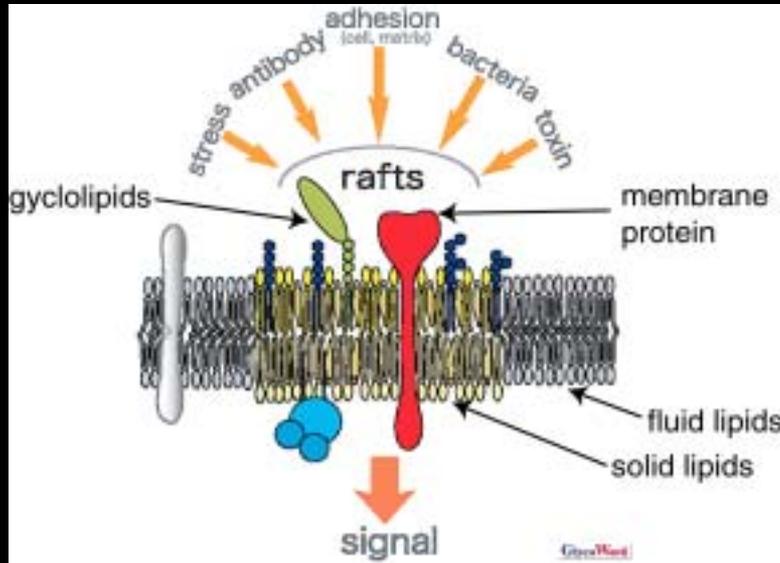


cell polarity, protein trafficking, signal transduction



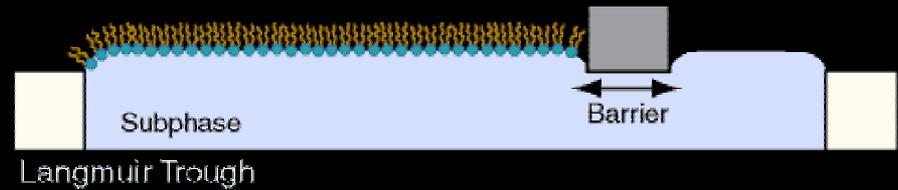
# Model Membranes

## Cellular Membranes

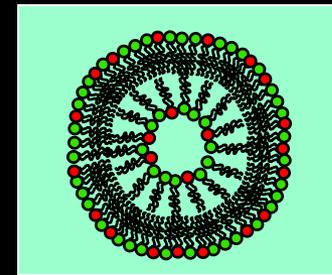


**80% of all Drugs interact with membrane proteins**  
**BIOSENSORS**

**monolayer** (water-air interface)

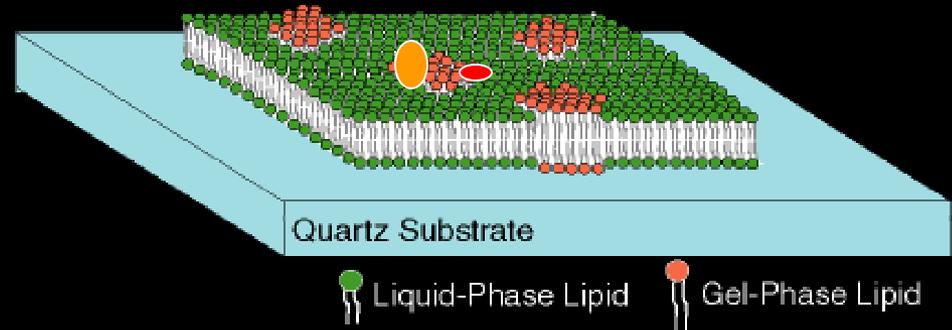


**“Model Membranes”**

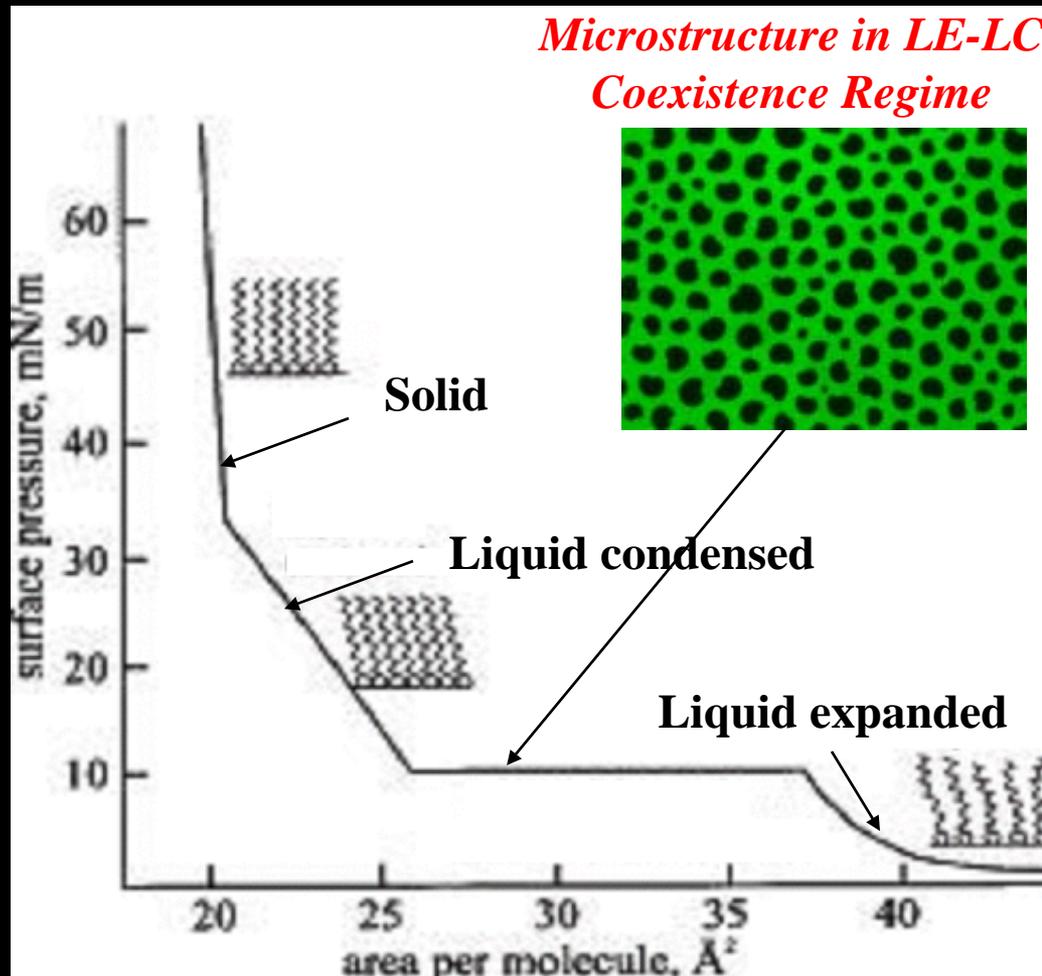


**vesicle**

**bilayer** (solid-water interface)

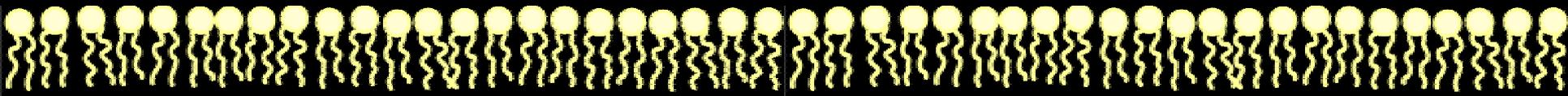


# Liquid Expanded – Liquid Condensed Phase Coexistence



Lateral heterogeneity also involves liquid-liquid immiscibility in the membrane plane.

V. M. Kaganer, Review of Modern Physics, Vol 71,779-819 1999



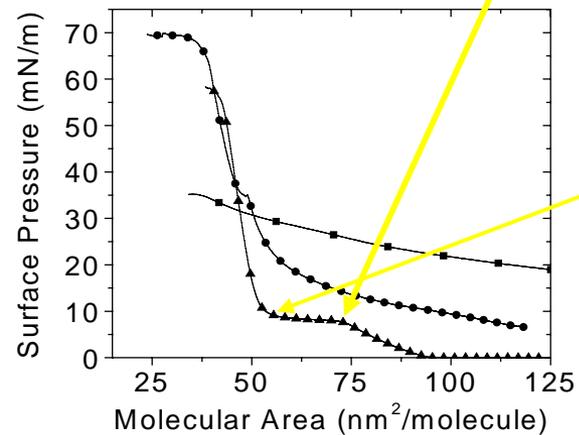
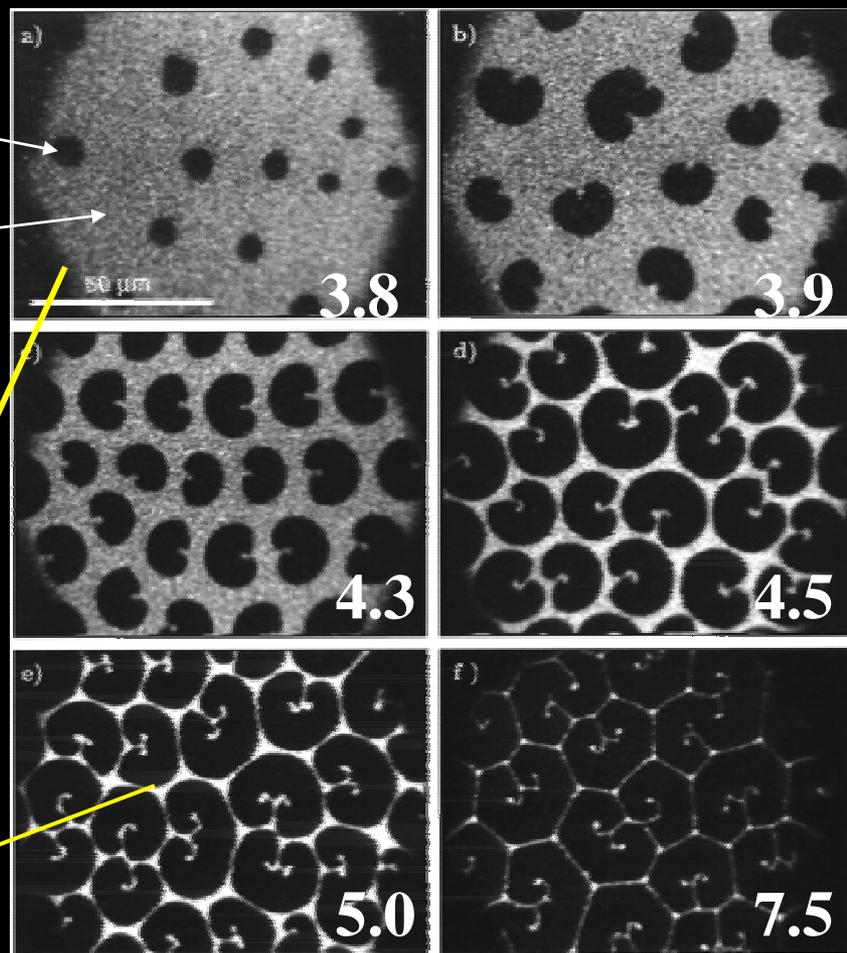
# Liquid Expanded – Liquid Condensed Phase Coexistence

## Domain Shape in LE-LC Coexistence Regime

Liquid condensed lipid

Liquid expanded lipid

Di-16-PC = DPPC

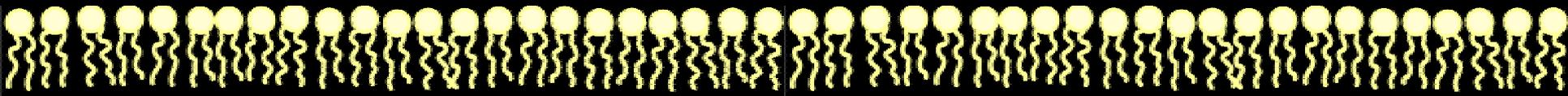
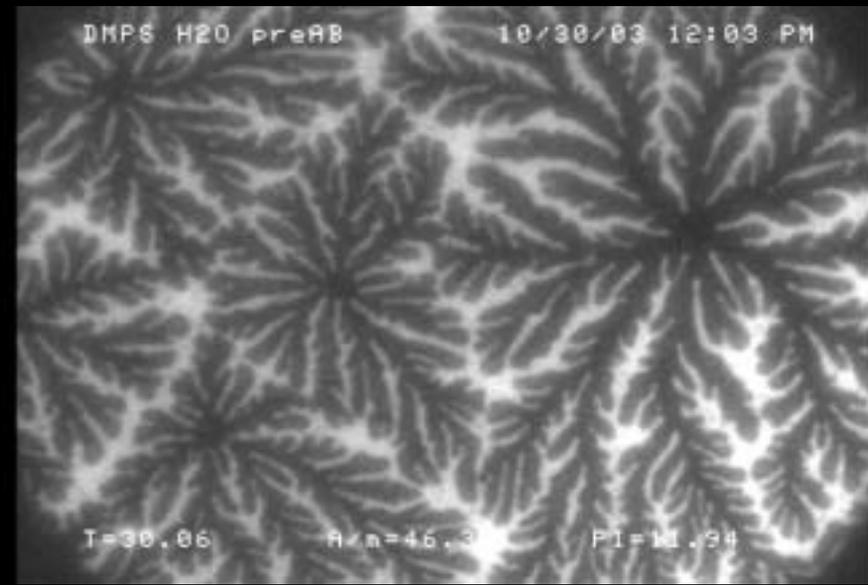


Cary W. McConlogue  
Langmuir 1999, 234-237



# Fluorescence Microscopy Image: DMPS lipids

Maximum resolution  $\sim 1 \mu\text{m}$

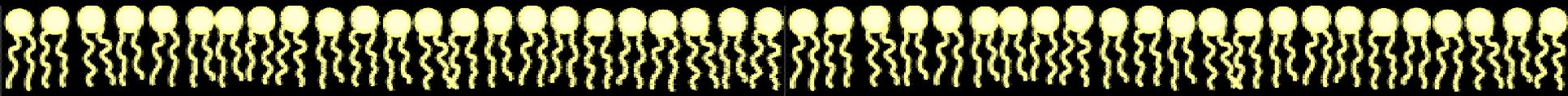


# Brewster Angle Microscopy Image

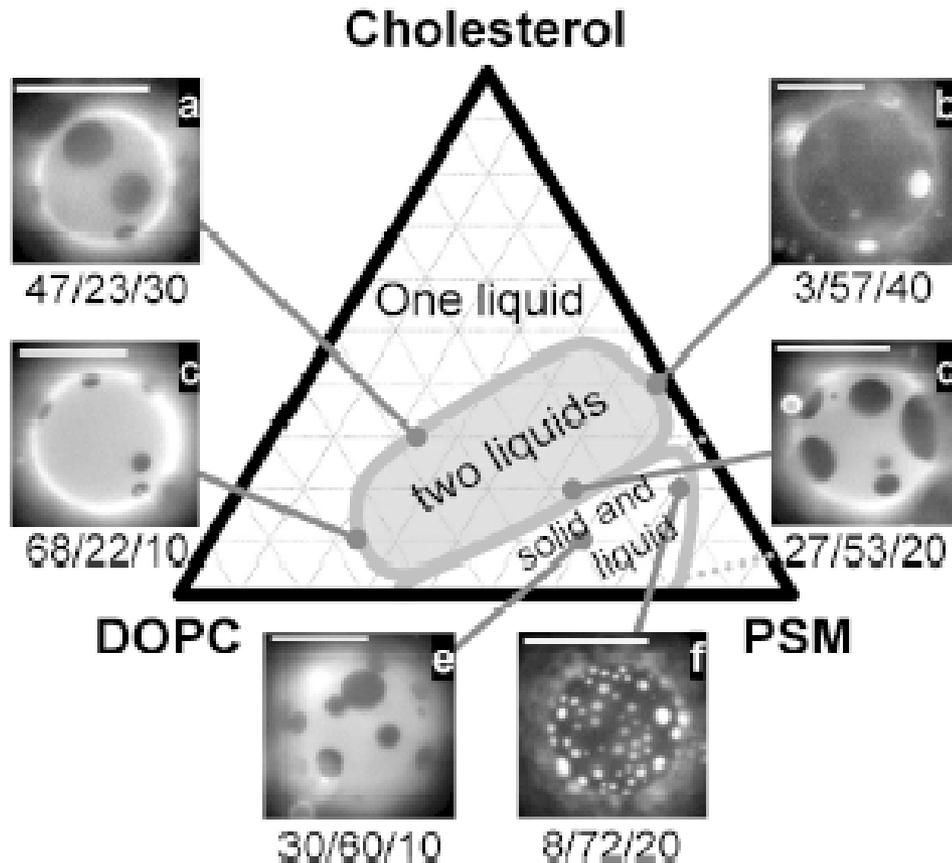
## DMPS lipids



Maximum resolution  $\sim 1 \mu\text{m}$



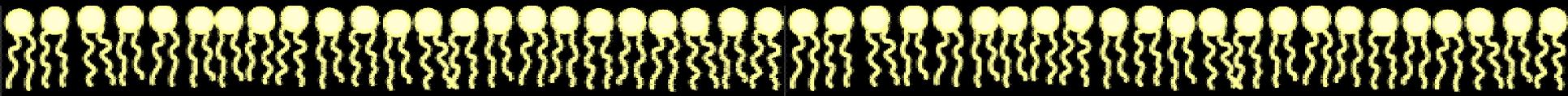
# Giant Vesicles and Liq-Liq Coexistence



DOPC/PSM/CHOL  
Scale bar 20 $\mu$ m

- Dark liquid phase **estimated** to be rich in PSM and CHOL
- Bright liquid phase **thought** to be rich in DOPC

Veatch and Keller, PRL 2005

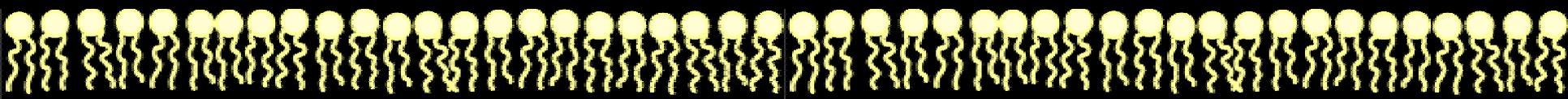
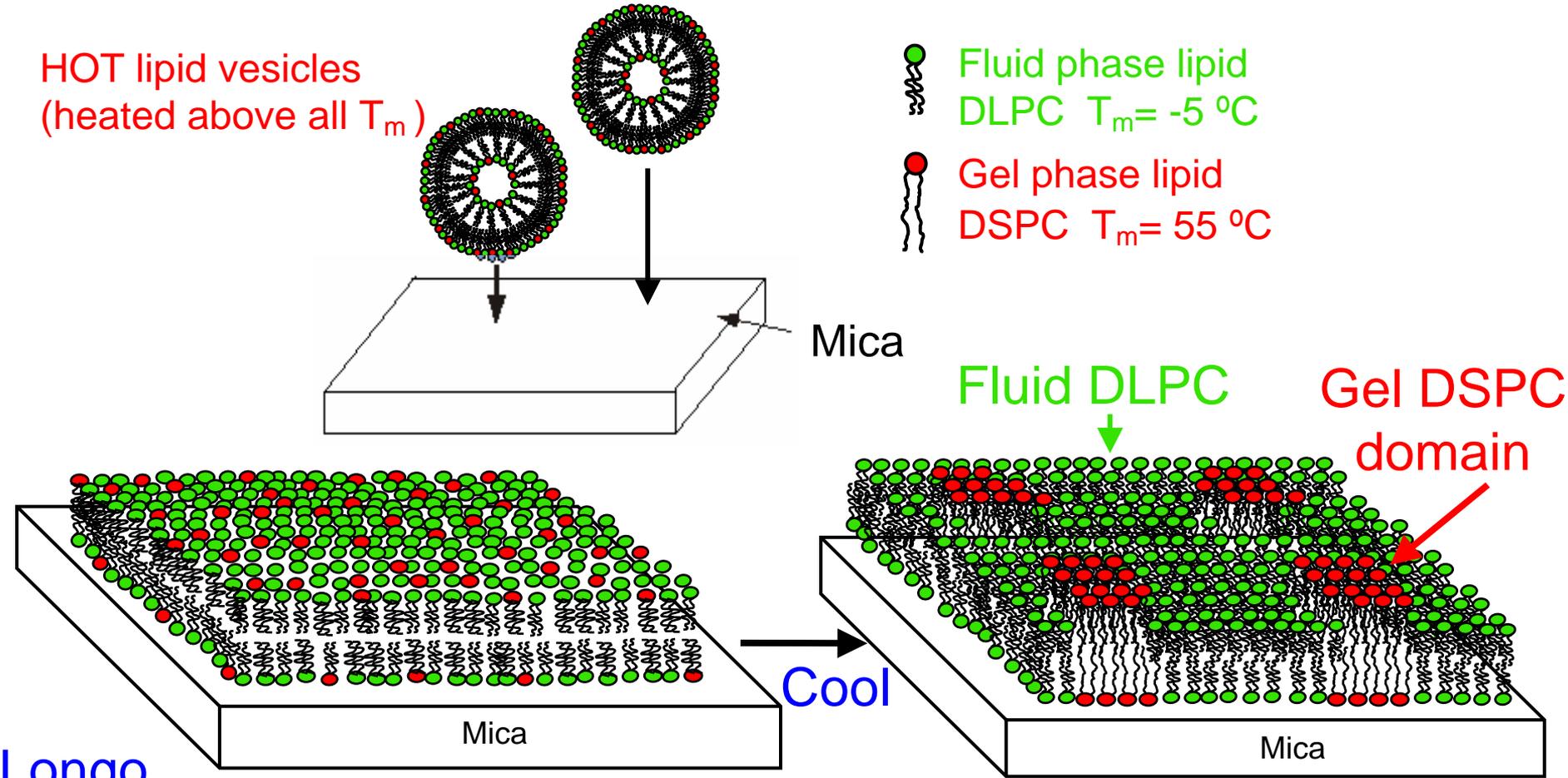


# Methods for Studying Lateral Domains

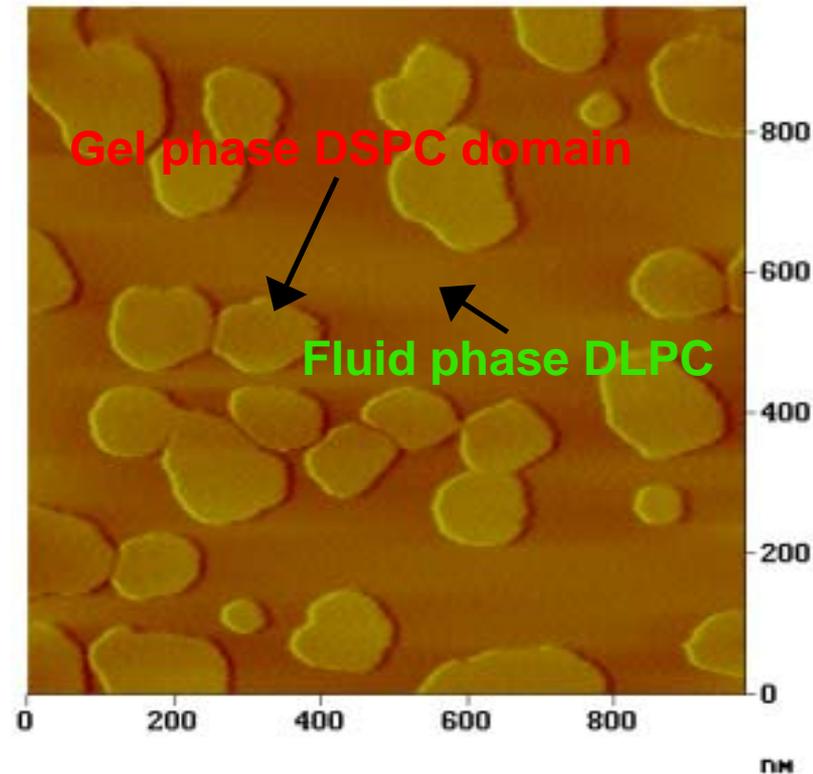
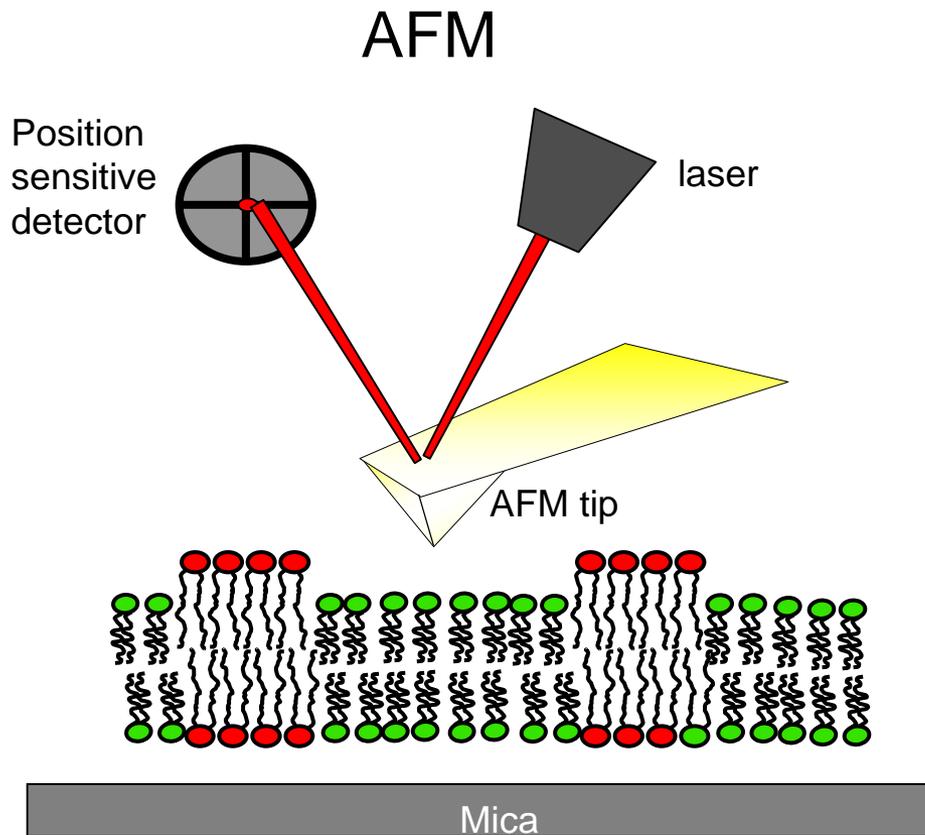
## Sample Prep - Quenched Vesicle Fusion

HOT lipid vesicles  
(heated above all  $T_m$ )

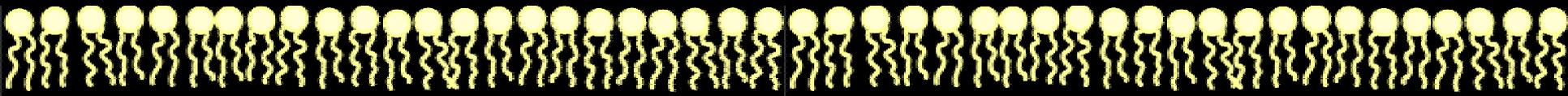
- Fluid phase lipid  
DLPC  $T_m = -5^\circ\text{C}$
- Gel phase lipid  
DSPC  $T_m = 55^\circ\text{C}$



# Characterizing Microstructure by AFM

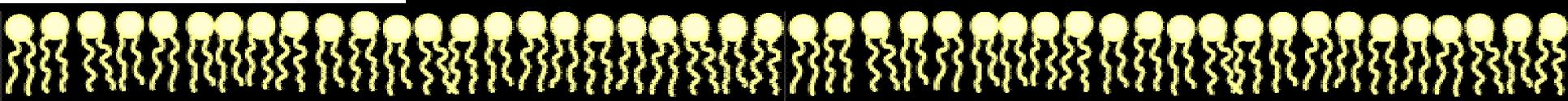
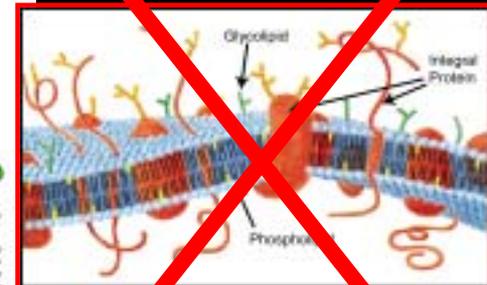
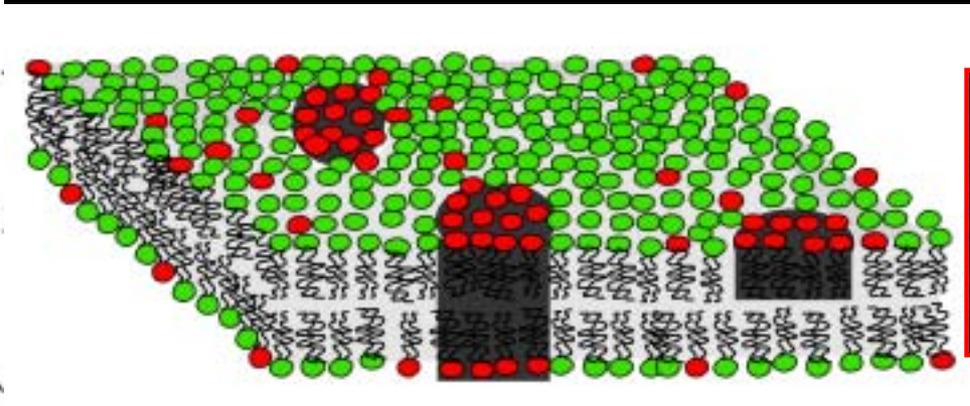
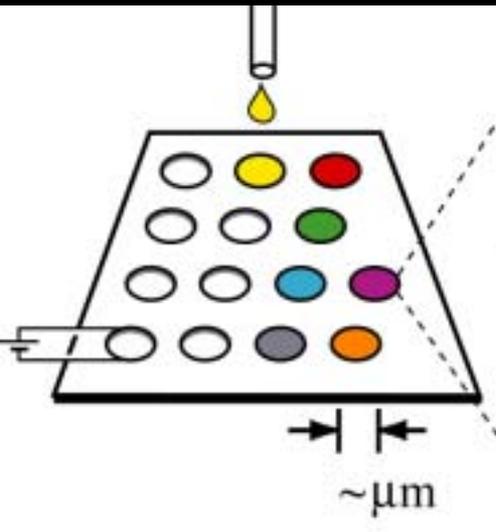
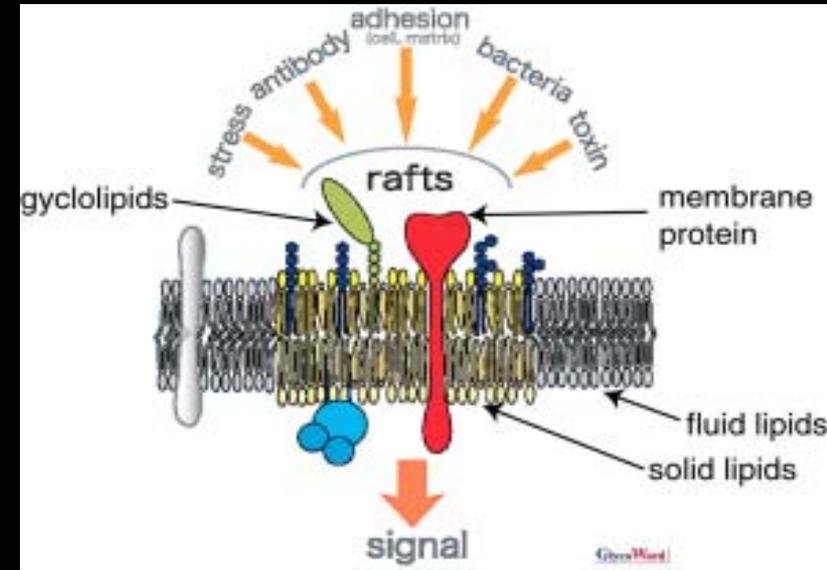


“Obstructed Diffusion in Phase-Separated Supported Lipid Bilayers, A Combined AFM and FRAP Approach”, *Biophysical Journal*, Ratto, T. V., Longo, M. L., 2002, 83:3380-3392



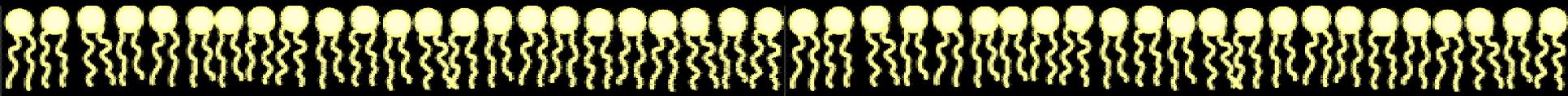
# Homogeneous vs Heterogeneous

- Better model systems
- How small are the lateral domains in real membranes
- Tools to characterize the domains *in-situ*

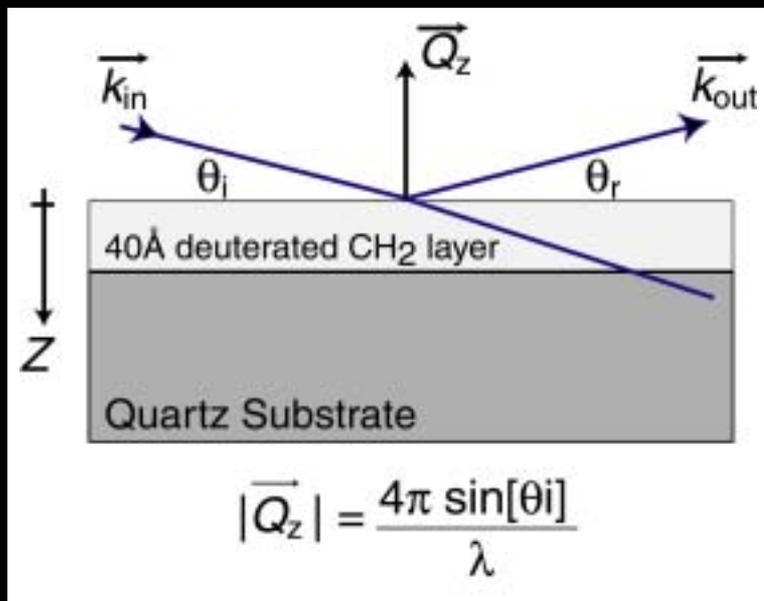


# Current Use of X-rays

*Monolayers and Supported Bilayers*



# Reflectivity [Neutrons and X-rays]

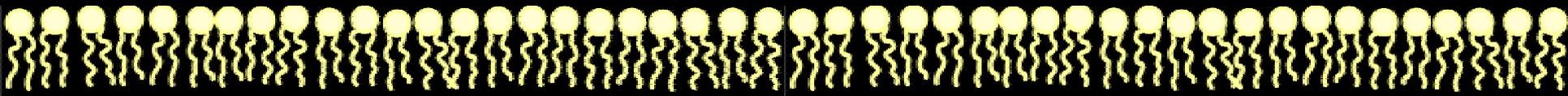
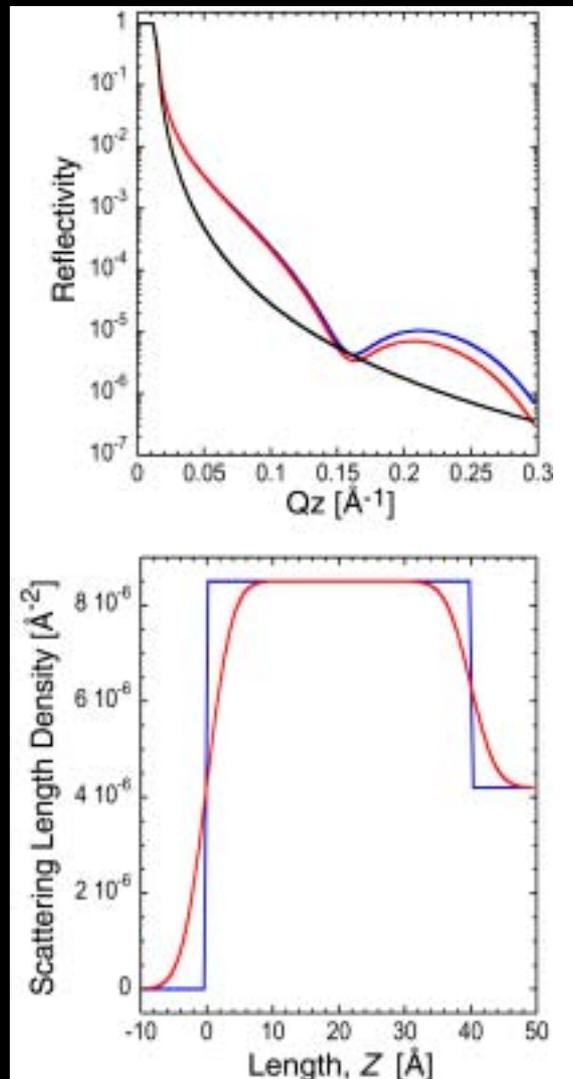


$\theta_i = \theta_r$   
**Specular  
Elastic**

## Measures:

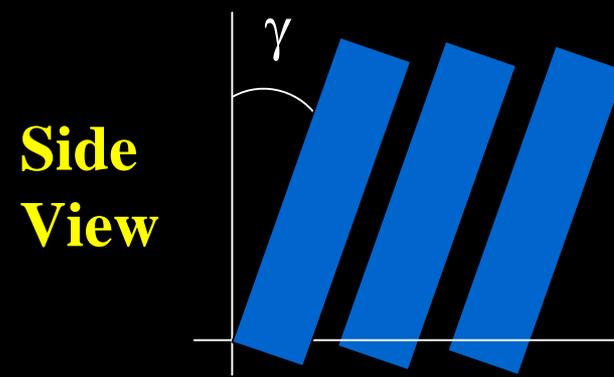
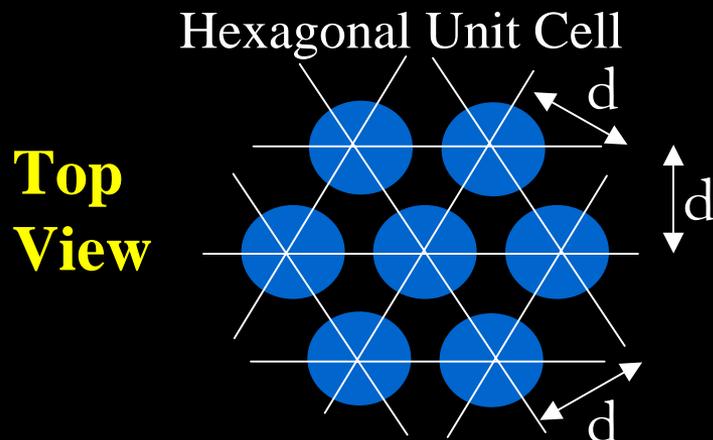
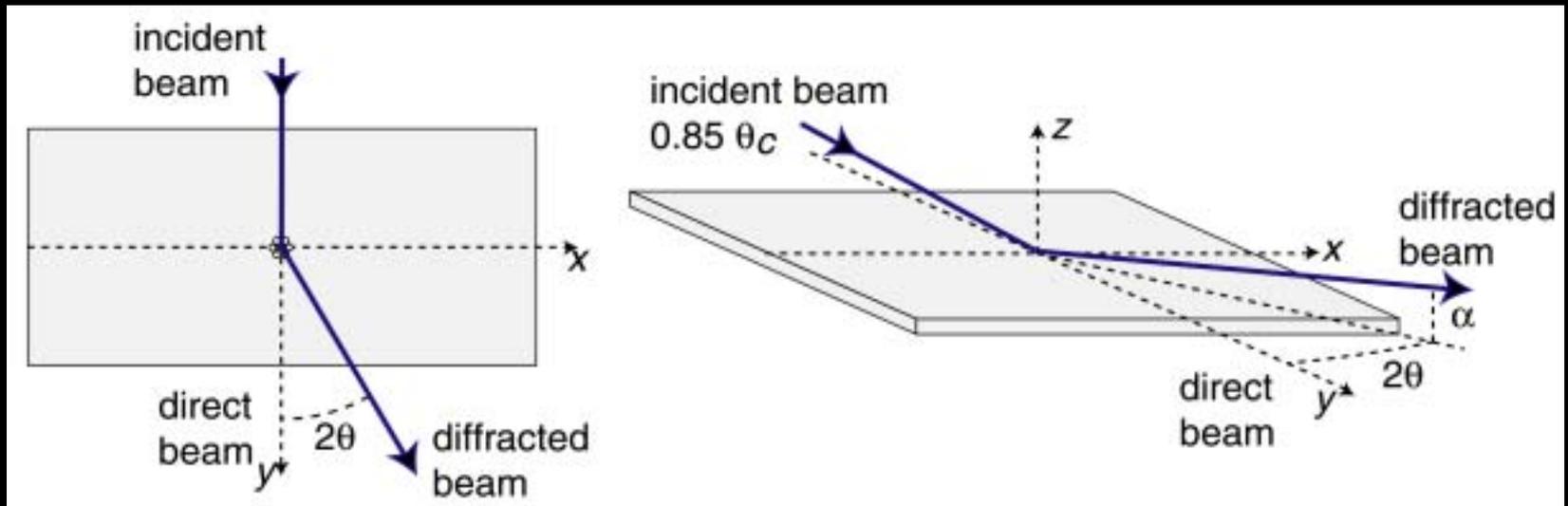
average density structure **normal** to the interface.

(layer thickness, density and roughness)



# X-ray Grazing Incidence Diffraction

## G I D

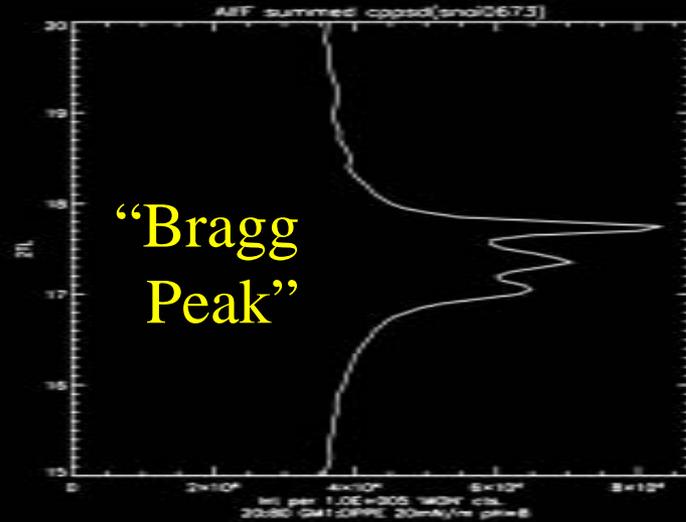


# Typical GIXD Data

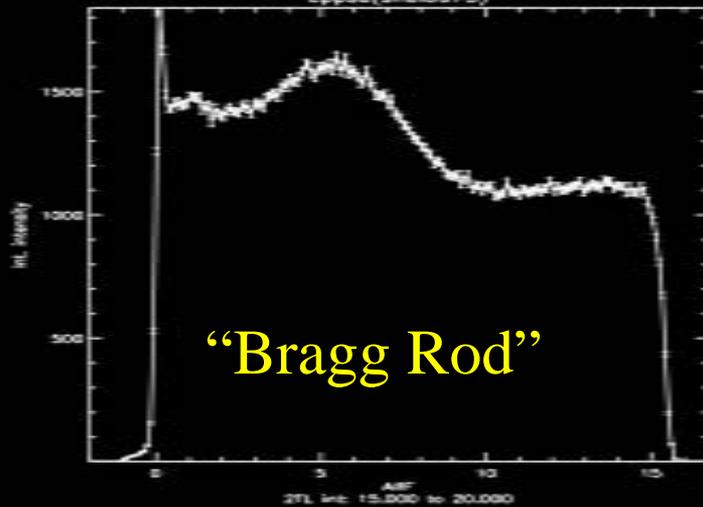
“Contour Plot”



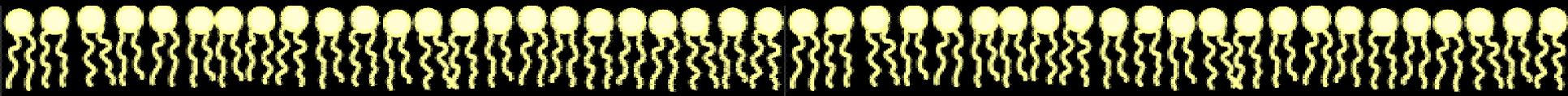
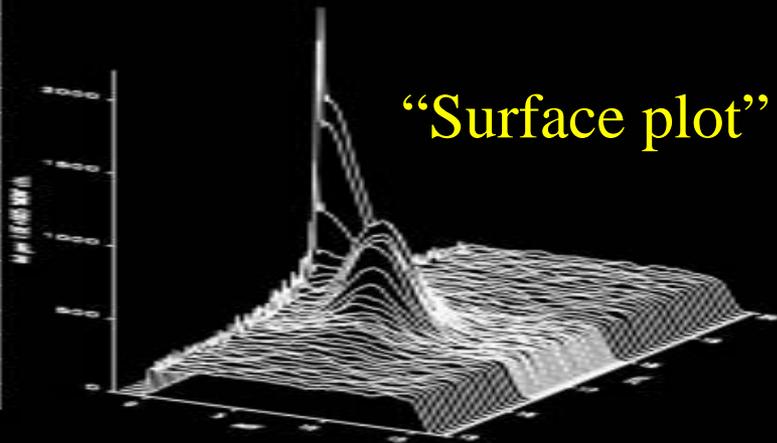
“Bragg Peak”



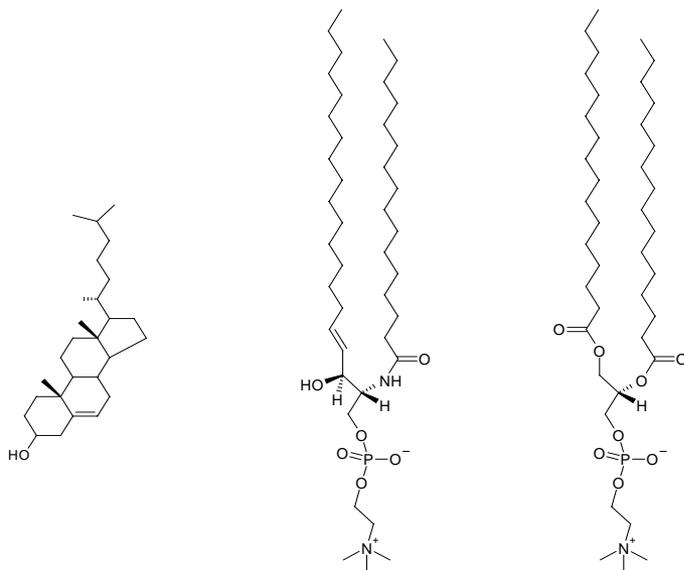
“Bragg Rod”



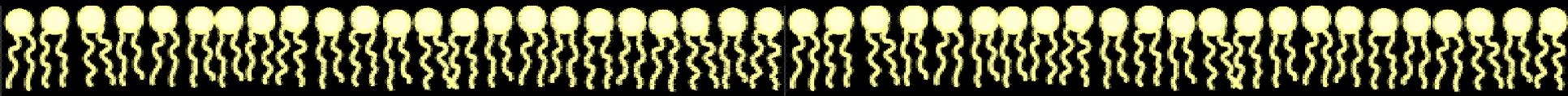
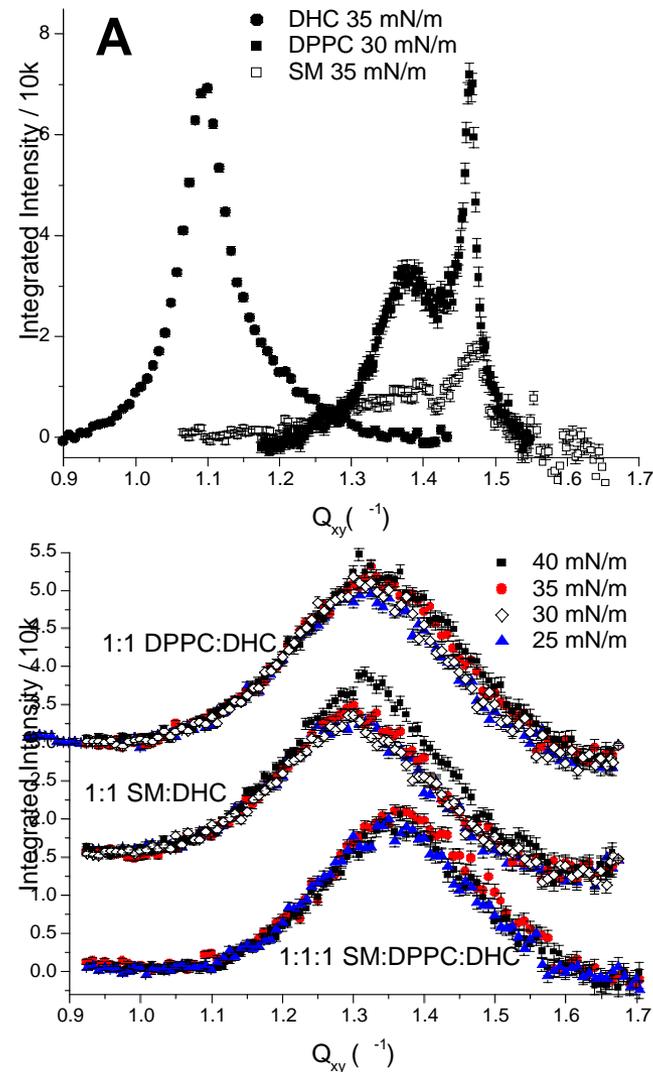
“Surface plot”



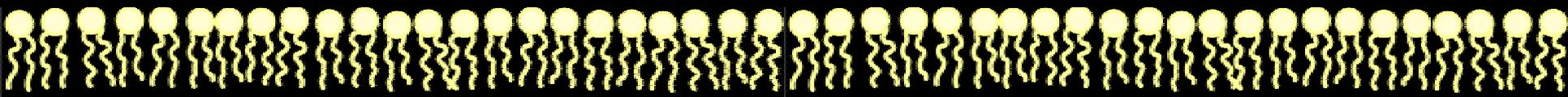
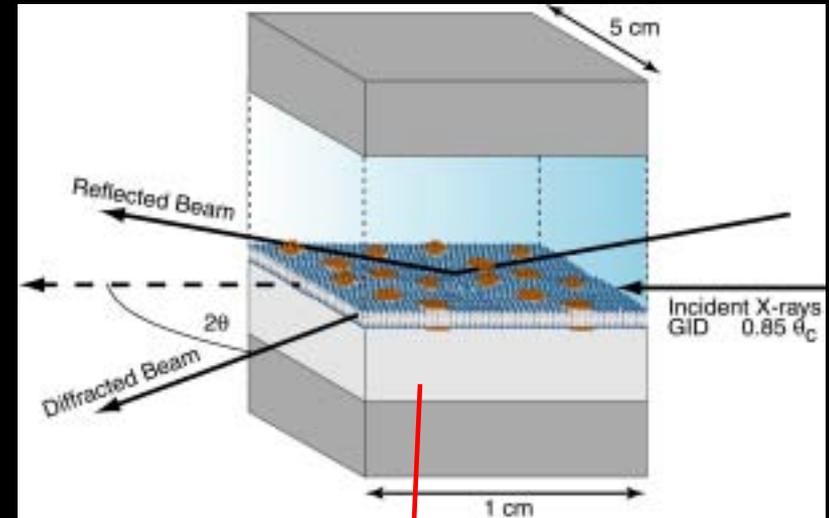
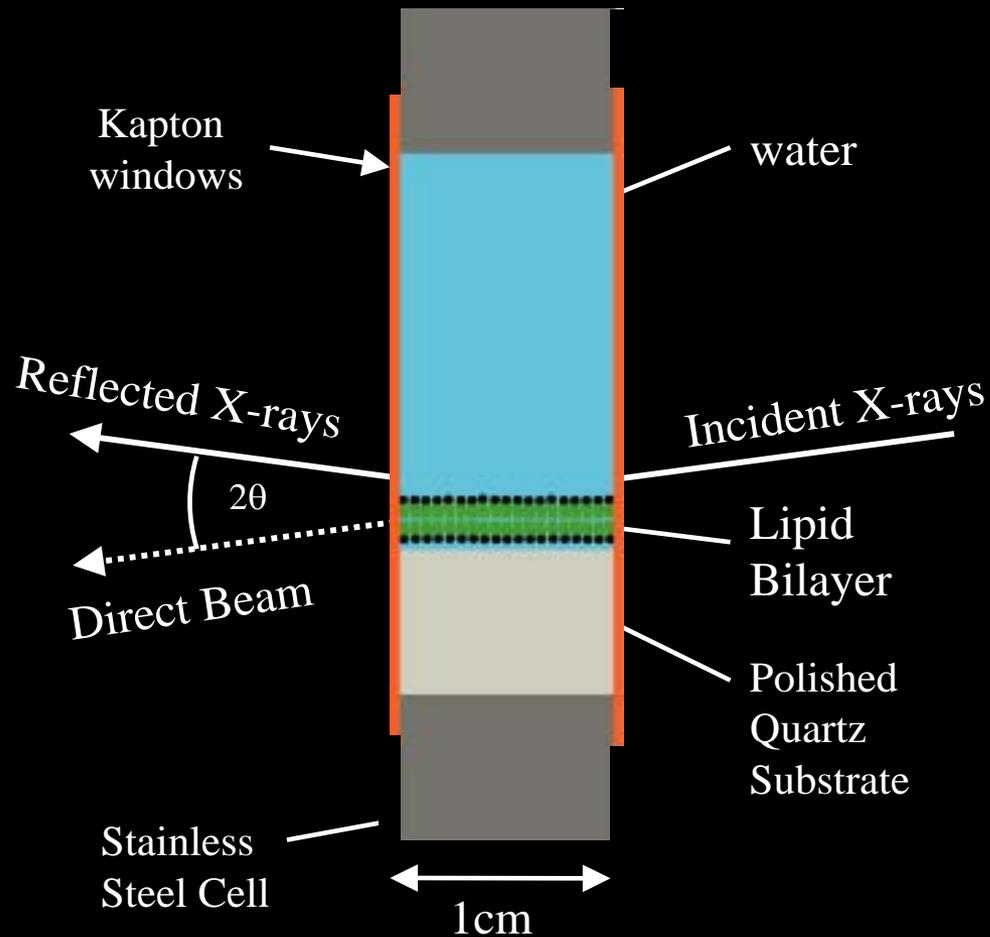
# Molecular Rafts: DPPC/SM/Cholesterol



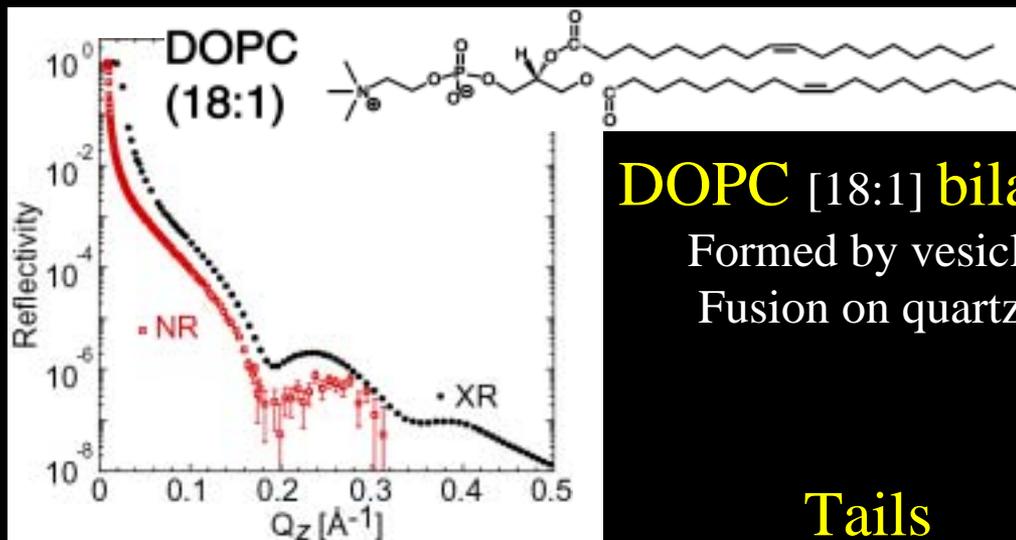
- coherence length  $\sim 22 \text{ \AA}$
- d-spacing  $\neq f$  (surface pressure)
- coherence length  $\neq f$  (surface pressure)
- amt of scattering entities  $\neq f$  (surface pressure)



# Solid-Liquid Interface



# Comparison to Neutron Reflectivity



## DOPC [18:1] bilayer

Formed by vesicle Fusion on quartz.

## Tails

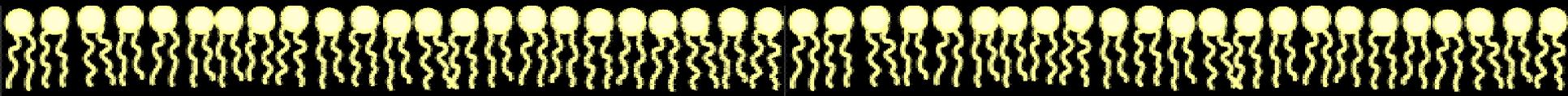
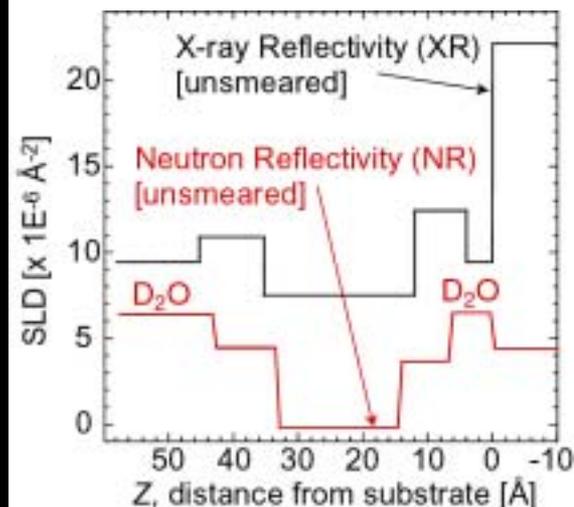
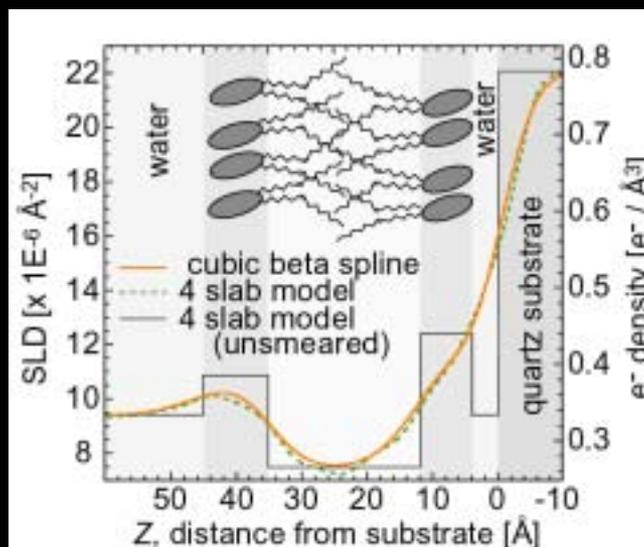
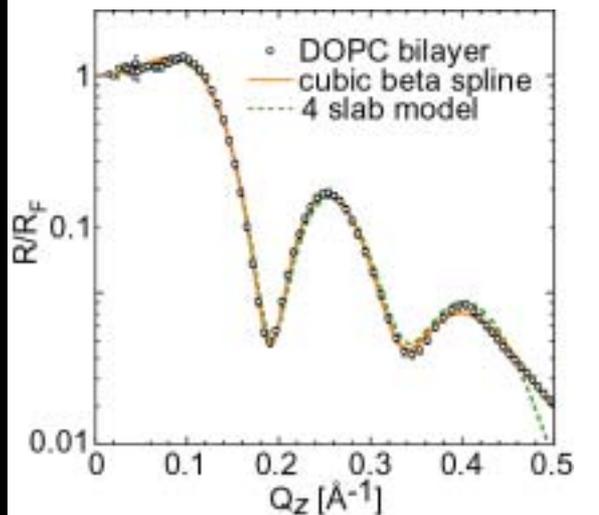
$23.2 \pm 0.5 \text{ \AA}$

Full extension

$23.4 \text{ \AA}$  per leaflet

## Water cushion

$4 \pm 0.5 \text{ \AA}$



# Summary

We can probe model-membrane structures either with:

1. micron resolution with optical microscopy  
(fluorescence or Brewster angle)
- or
2. ångstrom resolution through GIXD and reflectometry  
AFM (but only for solid phases)

*The range between is “terra incognita”!!!*

