School of Food Science and Nutrition
FACULTY OF MATHEMATICS AND PHYSICAL SCIENCES
UNIVERSITY OF LEEDS

New Insights on the Interaction of Flavonoids with Biomimetic Membranes

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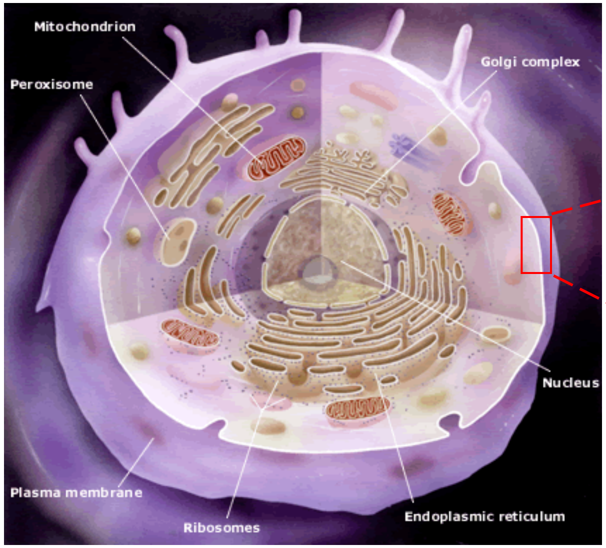
Neutrons and Food Conference, University of Lund, Sweden
02. June 2016

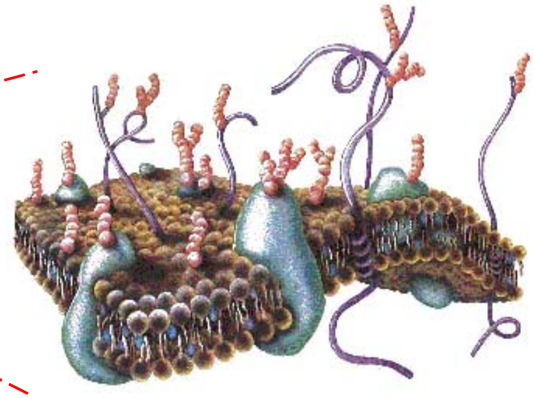
Overview

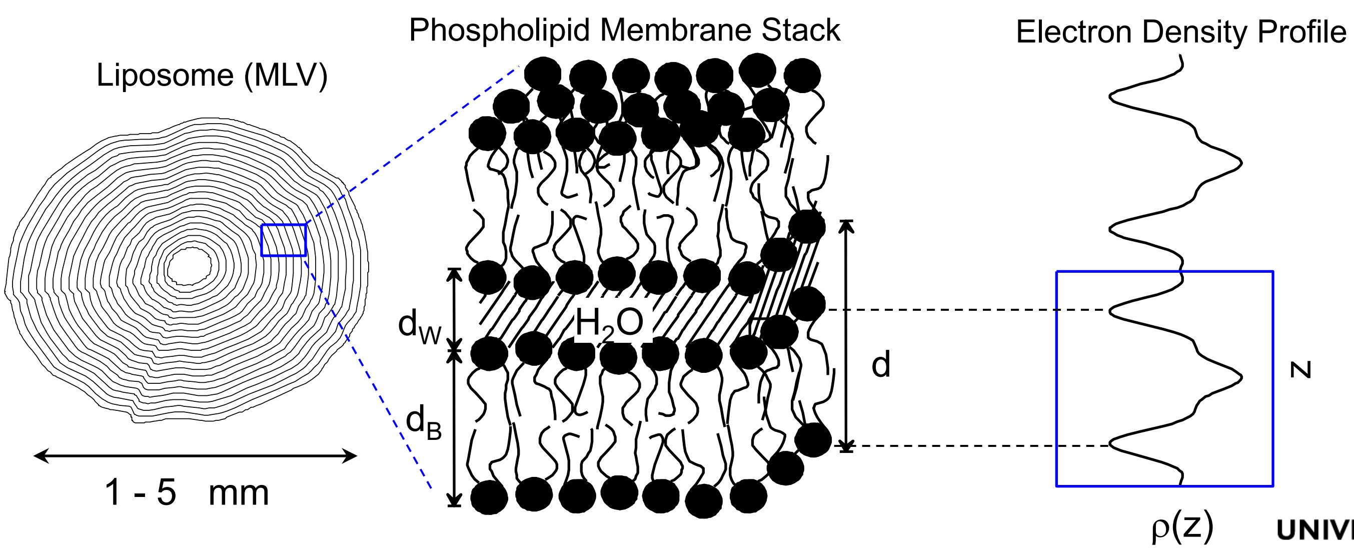
- Playing with “Poor” Diffraction Data
- Global Analysis of Lipid Bilayer Structure
- Flavonoids’ Interaction with Biomimetic Membranes
- Flavonoids’ Protective Properties in Mitochondrial Stress
- Outlook: Combining Structural & Simulation Data

From Cells to Vesicles



Life bases on Biomembranes

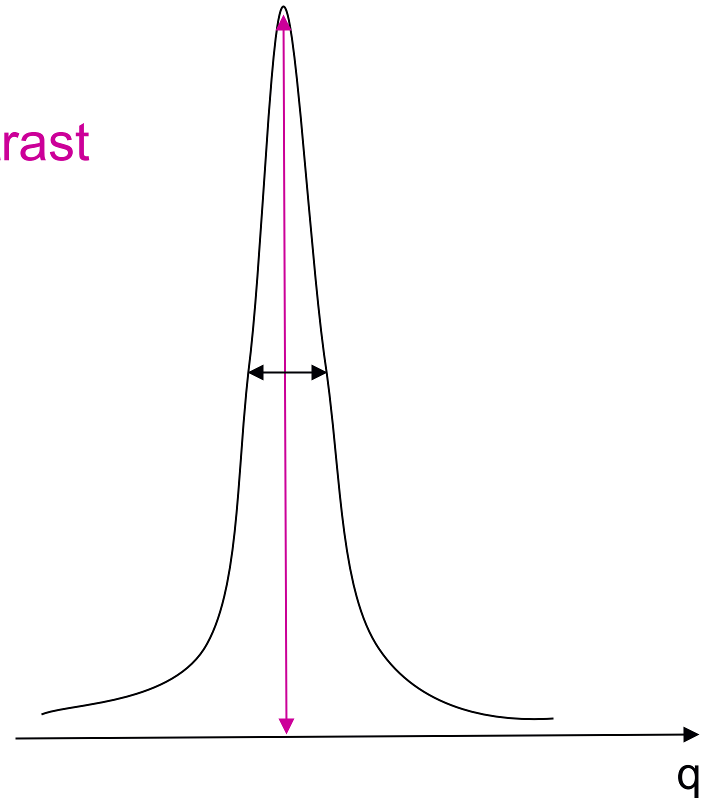




The Information Given in Diffraction Peak

- Position: *d*-spacing
- Width: crystallite size
- Height: electron density contrast
- Shape: disorder type

$$FWHM = \frac{0.89 \cdot \lambda}{L} \approx \frac{\lambda}{N \cdot d}$$



The Information Given in Diffraction Peak

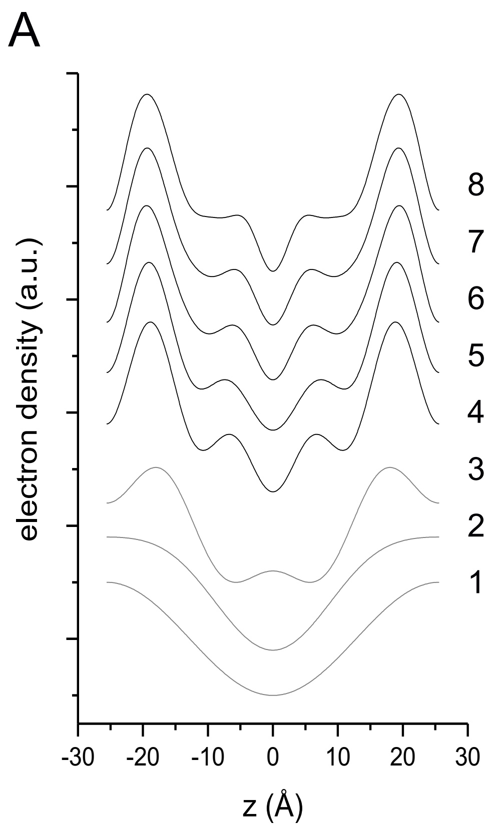
$$\tilde{\rho}(z) = \sum_{h=1}^{h\max} \alpha_h F_h \cos\left(\frac{2\pi z}{d}\right)$$

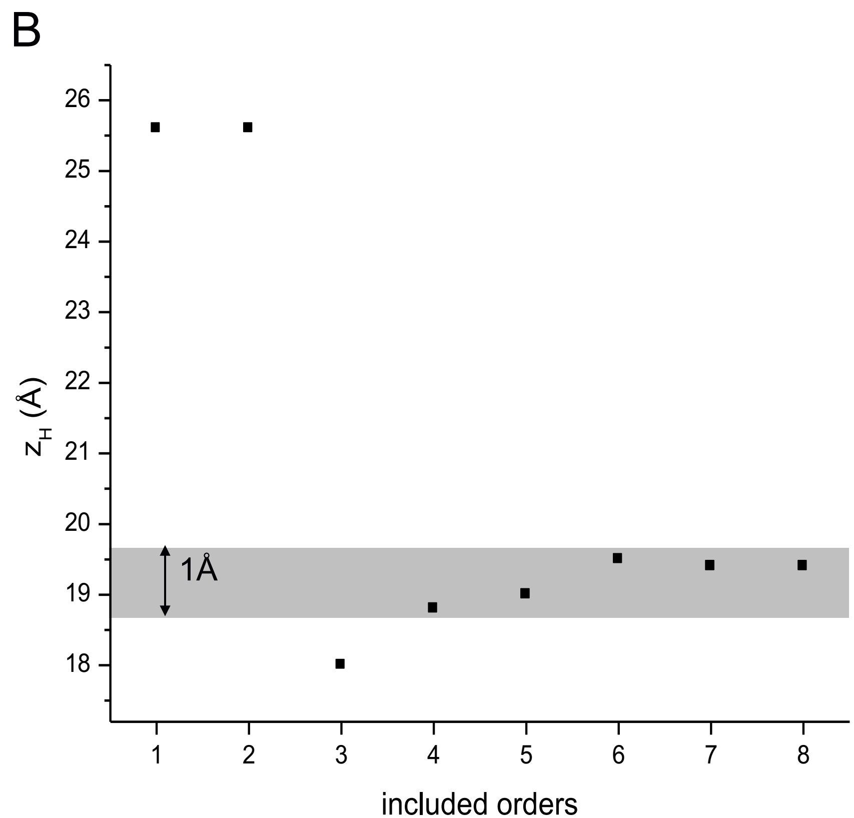
What do I need?

1.) 4 diffraction peaks

2.) The correct phase combination: - - + -

Why are Four Diffraction Orders Enough?





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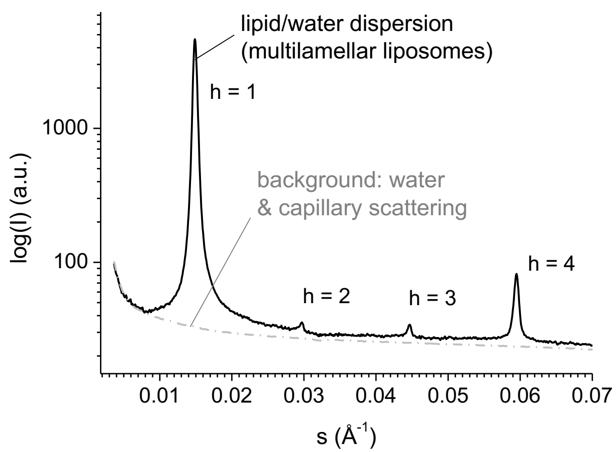
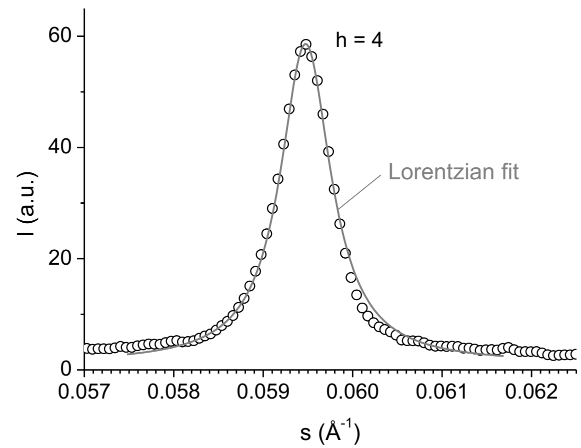
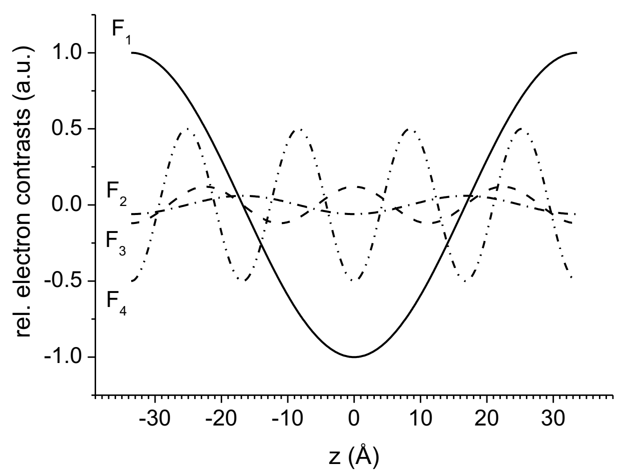
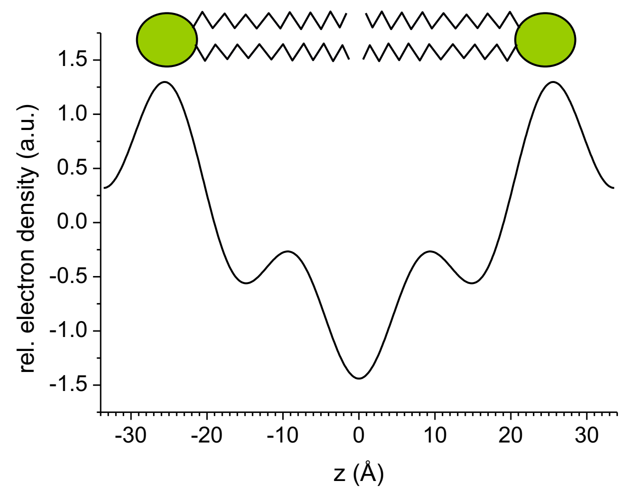
Electron Density Profile Determintion

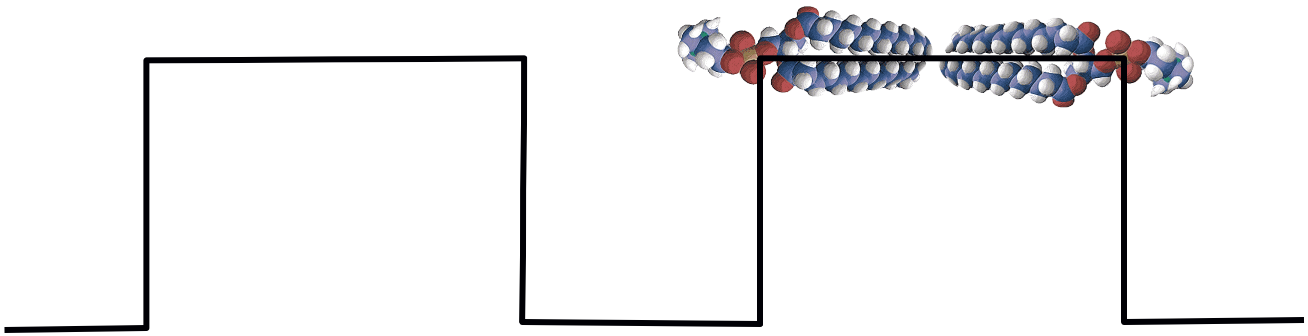








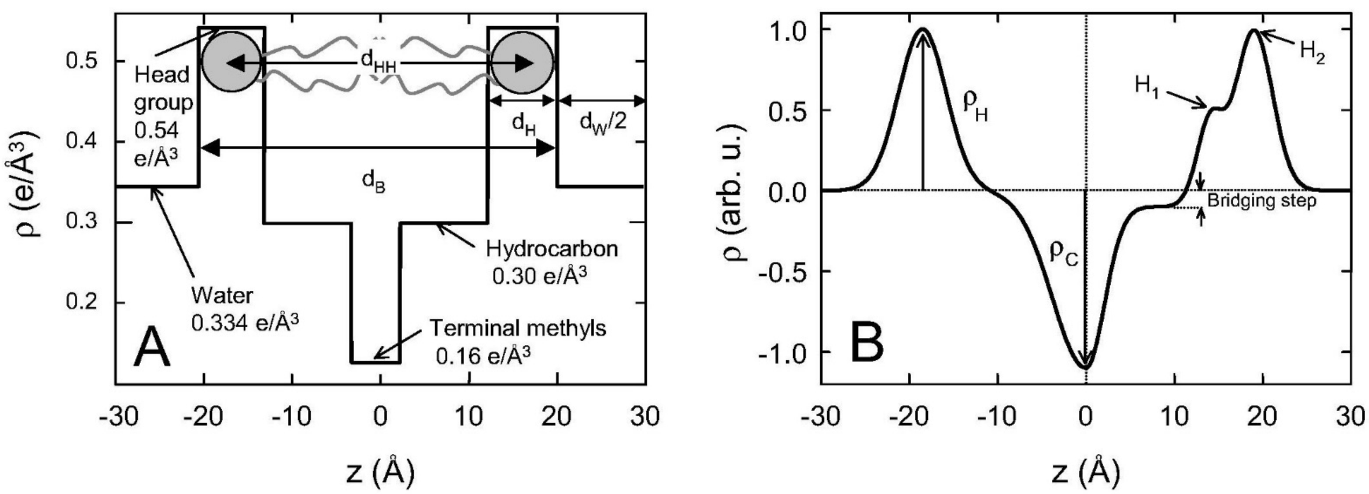
Bilayer Models: Vittorio Luzzati's Idea



$d_{B\_Luzzati} = \Phi_L\, d$

$\sim d_{HH}$

Modelling Bilayers: Strips and Gaussians



$$|F(q)|^2 = 2\pi[2\sigma_H \exp(-\sigma_H^2 q^2/2) cos(q z_H) - \sigma_C \rho_R \exp(-\sigma_C^2 q^2/2)]^2$$

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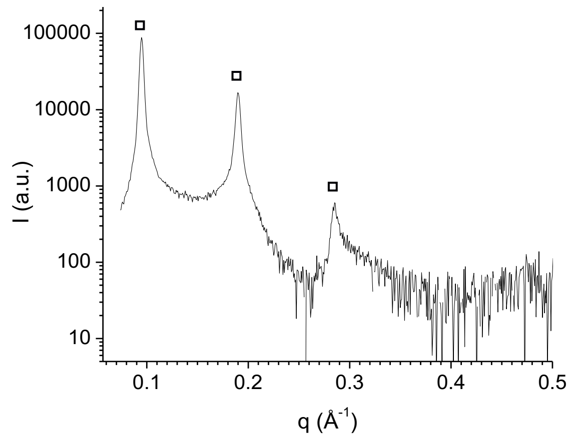
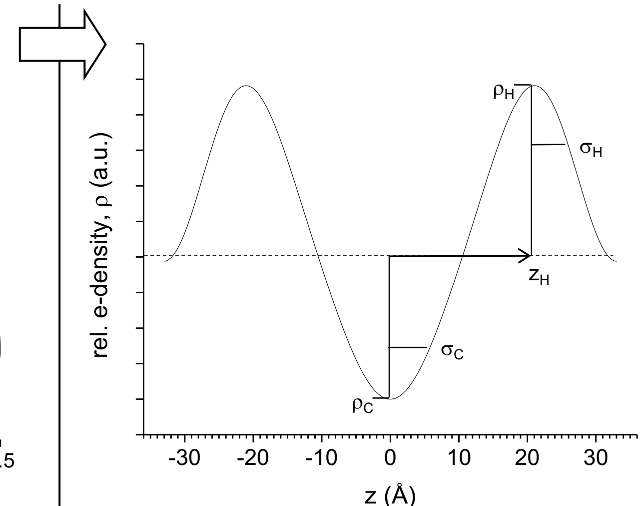
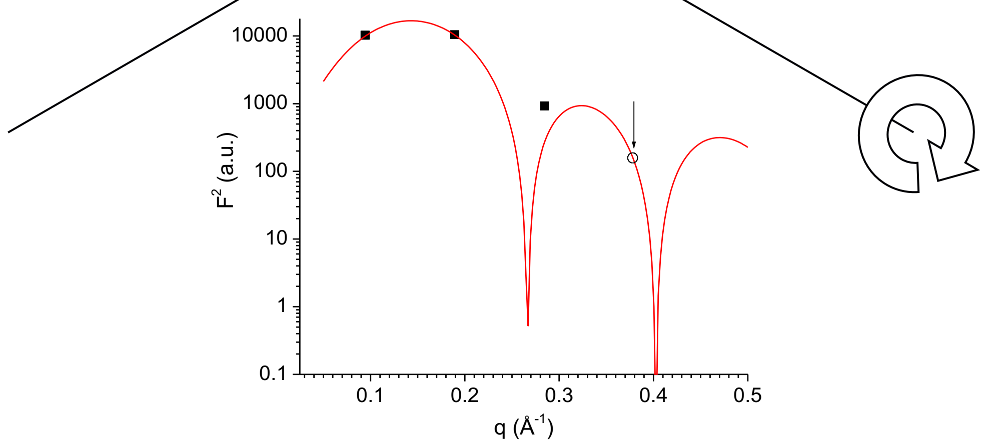
Interpolation of „Poor“ Data



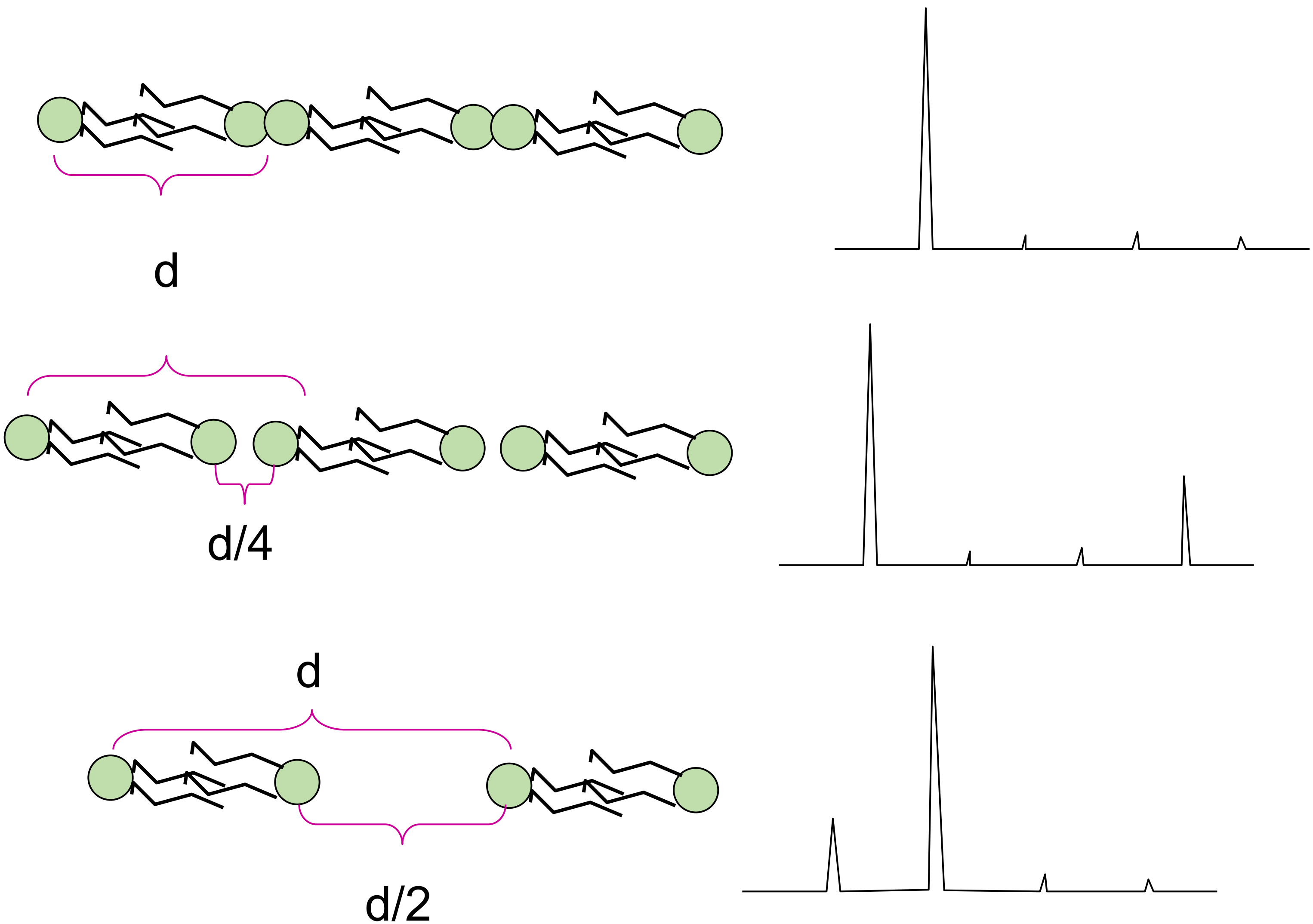


$$\rho(z) = \rho_s + \sum_{h=1}^{h_{\max}} \alpha_h F_h \cos\left(\frac{2\pi h z}{d}\right) \quad (1)$$

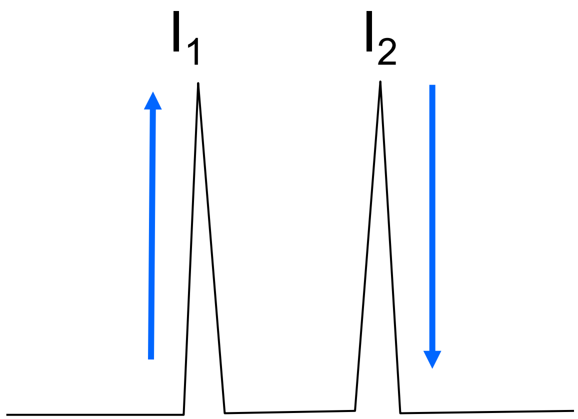


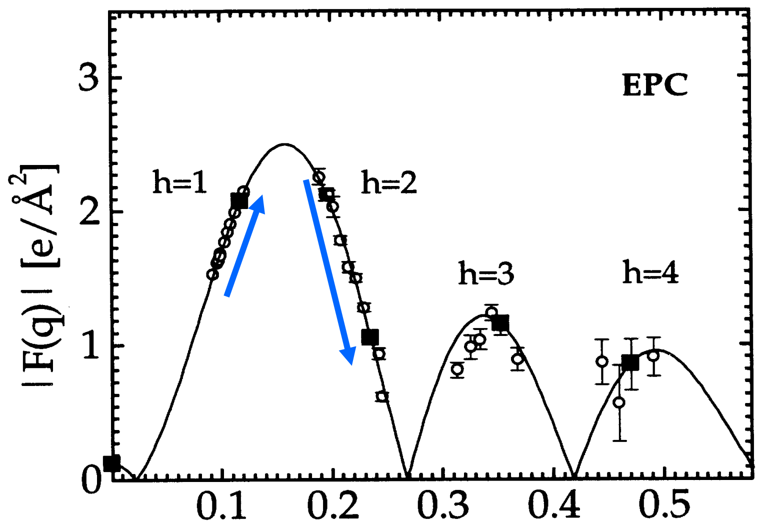
$$F(q)^2 = 2\pi\left[2\sigma_H \exp\left(-\sigma_H^2 q^2/2\right)\cos(q z_H) - \sigma_C \rho_r \exp\left(-\sigma_C^2 q^2/2\right)\right]^2 \quad (2)$$

Playing with Contrast



Taking a Look to F1 and F2





Decreasing of Hydration

$$|F(q)|^2 = 2\pi[2\sigma_H \exp(-\sigma_H^2 q^2/2) cos(q z_H) - \sigma_C \rho_R \exp(-\sigma_C^2 q^2/2)]^2$$

2 Peak Estimation of Bilayer Thickness

$$z_H = \pm \frac{d}{2\pi} \cdot \arccos\left(\frac{c_1 - \sqrt{8\,(r_F\,c_3)^2 + 8\,(c_2 - r_F\,c_4)\cdot(r_F\,c_3) + c_1^2}}{4\,r_F\,c_3}\right) \quad = \Phi_L$$

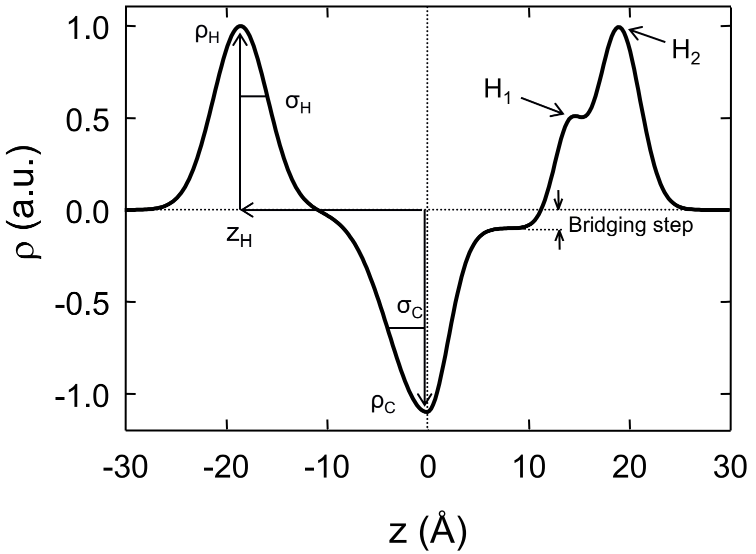
with $r_F = F_1/F_2$

$c_1 = 2\sigma_H \exp(-2\pi^2 \sigma_H^2/d^2)$

$c_2 = -\sigma_C |\rho_r| \exp(-2\pi^2\sigma_C^2/d^2)$

$c_3 = 2\sigma_H \exp(-8\pi^2\sigma_H^2/d^2)$

$c_4 = -\sigma_C |\rho_r| \exp(-8\pi^2 \sigma_C^2/d^2)$



Some Examples: POPE, POPC and DMPC

Bilayer parameters at full hydration.

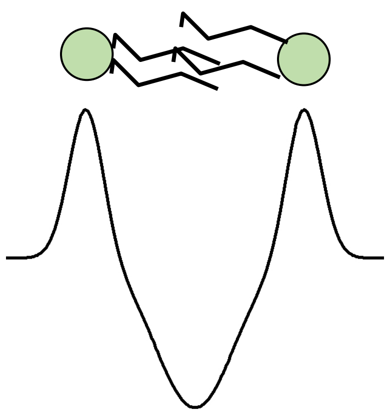
	POPE[a] (30 ° C)	**2-peak method**	**3-peak method**	**POPC[b] (2° C)**	**3-peak method**	**DMPC[c] (30 ° C)**	**2-peak method**
z_H (Å)	**20.0[a]**	**21.3**	**20.0 (19.8)**	**20.2**	**20.8 (20.4)**	**17.3**	**19.1**
σ_H (Å)	3.4[a]	3.1[d]	3.3 (4.1)	3.6	4.8 (4.6)	3.0	3.1[d]
σ_C (Å)	4.8[a]	4.4[d]	8.3 (8.9)	4.8	6.9 (7.3)	4.5	4.4[d]
$\|\rho_r\|$	1.01[a]	0.91[d]	1.01 (1.01)	0.73	1.30 (1.24)	1.13	0.91[d]

Rappolt, M. (2010):
Bilayer thickness estimations with "poor" diffraction data.
J Appl. Phys. 107, 084701, 1-7

Real and Inverse Space

Lattice & its disorder

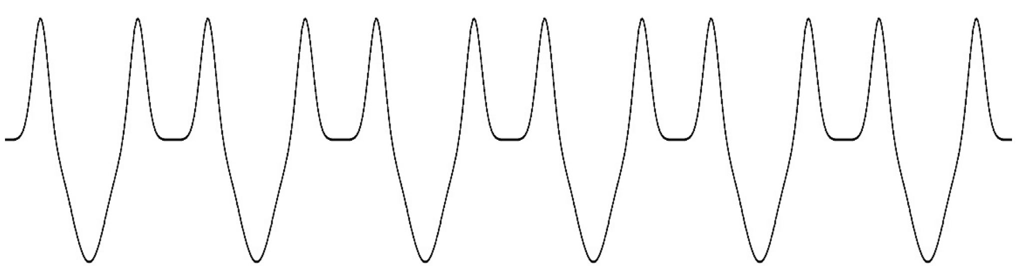
⊗

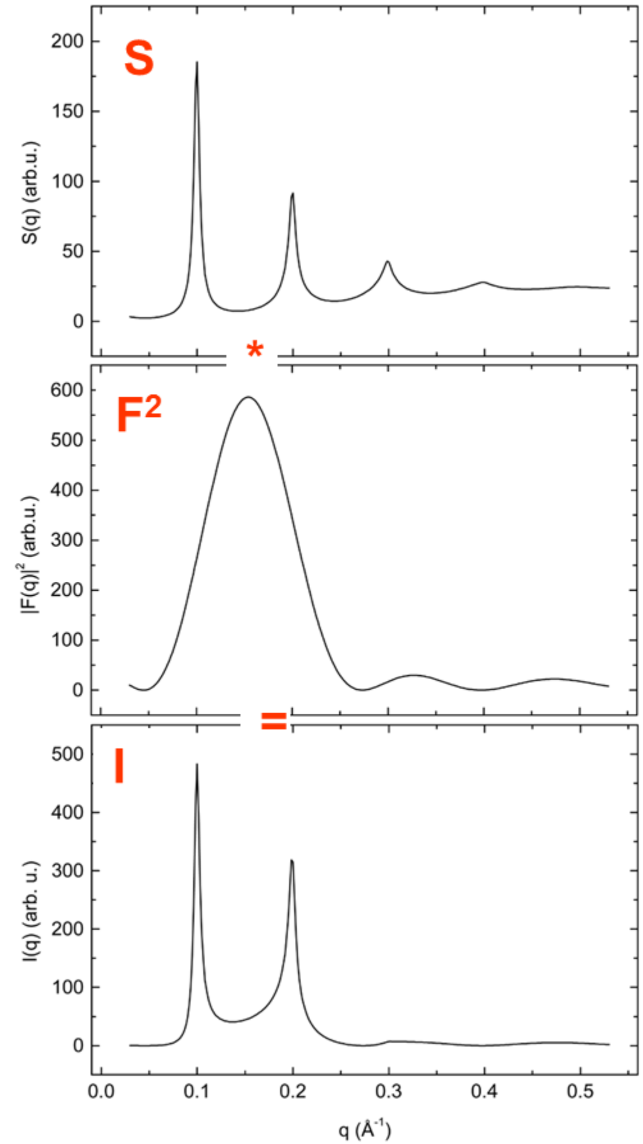


Motif

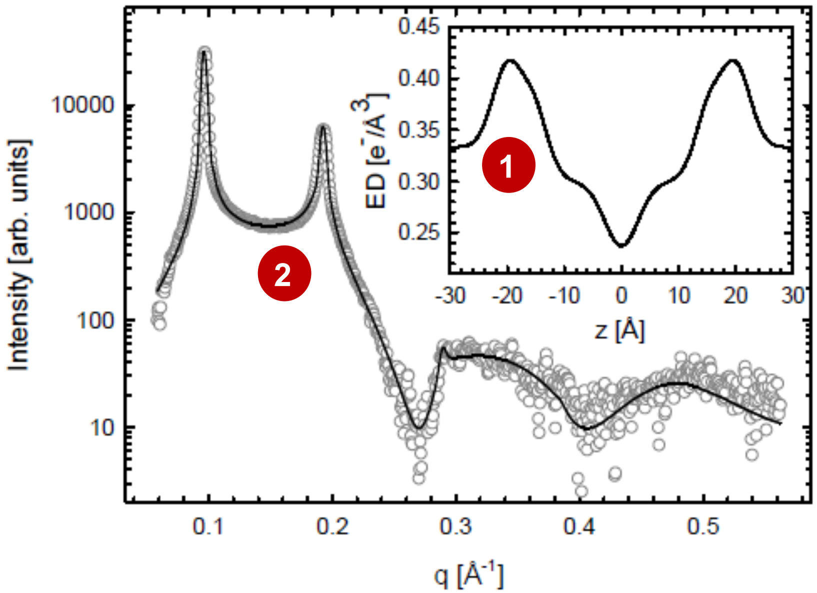
=

Crystal



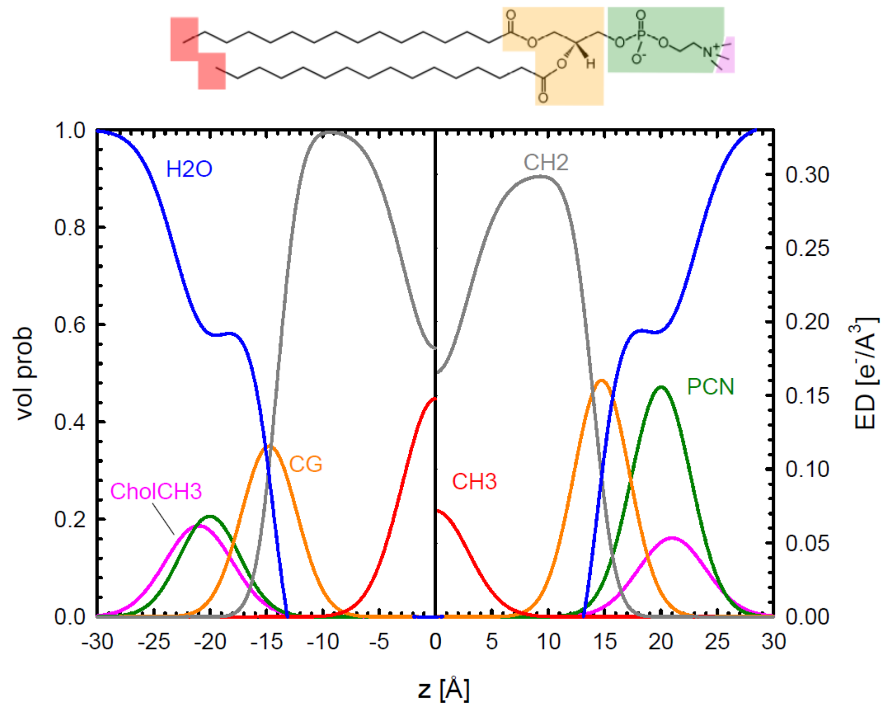


The Global Model for Fluid Lamellar Phases



G. Pabst, M. Rappolt, H. Amenitsch and P. Laggner **2000** *Phys. Rev. E* 62, 4000

M. Rappolt, P. Laggner, and G. Pabst, *Recent Res. Devel.* **2004** Biophys. 3, 363



Heftberger, P. et al. (**2013**): J. Appl.Cryst.,

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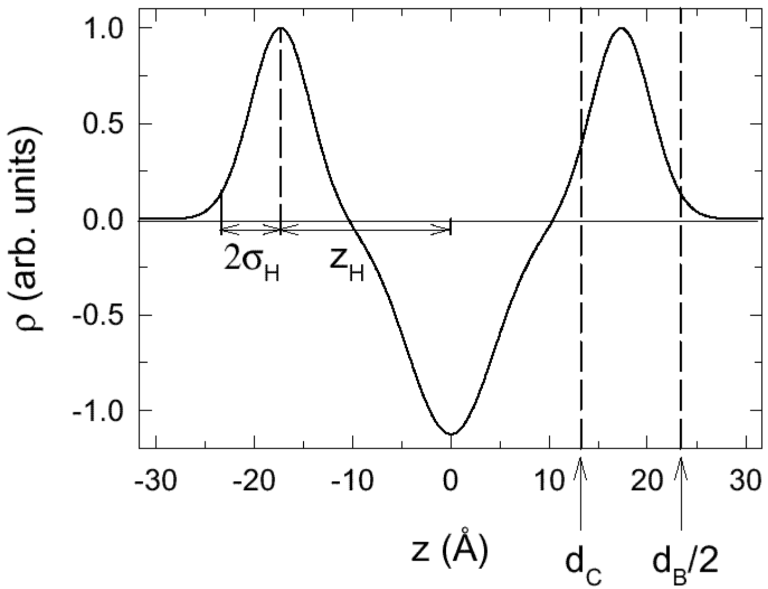
What Do You Get?



$d_B = 2\,(z_H + 2\sigma_H)$

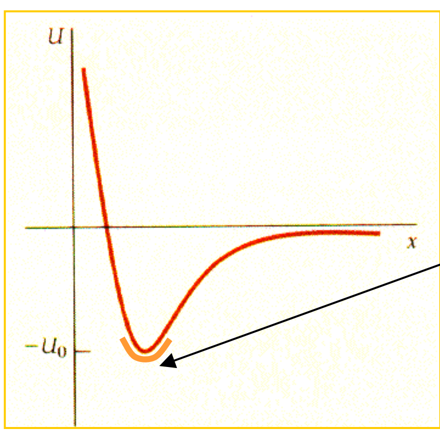
$d_W = d - d_B$

$d_C = d_B/2 - d_H$

$d_C = z_H - 4.1$ Å

$A = (V_L + V_H)/d_C$

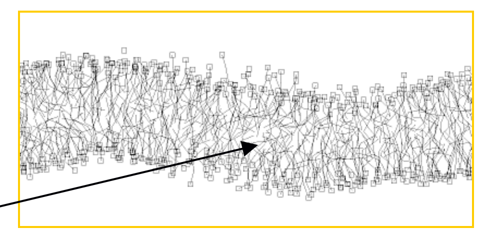
Geometrical method



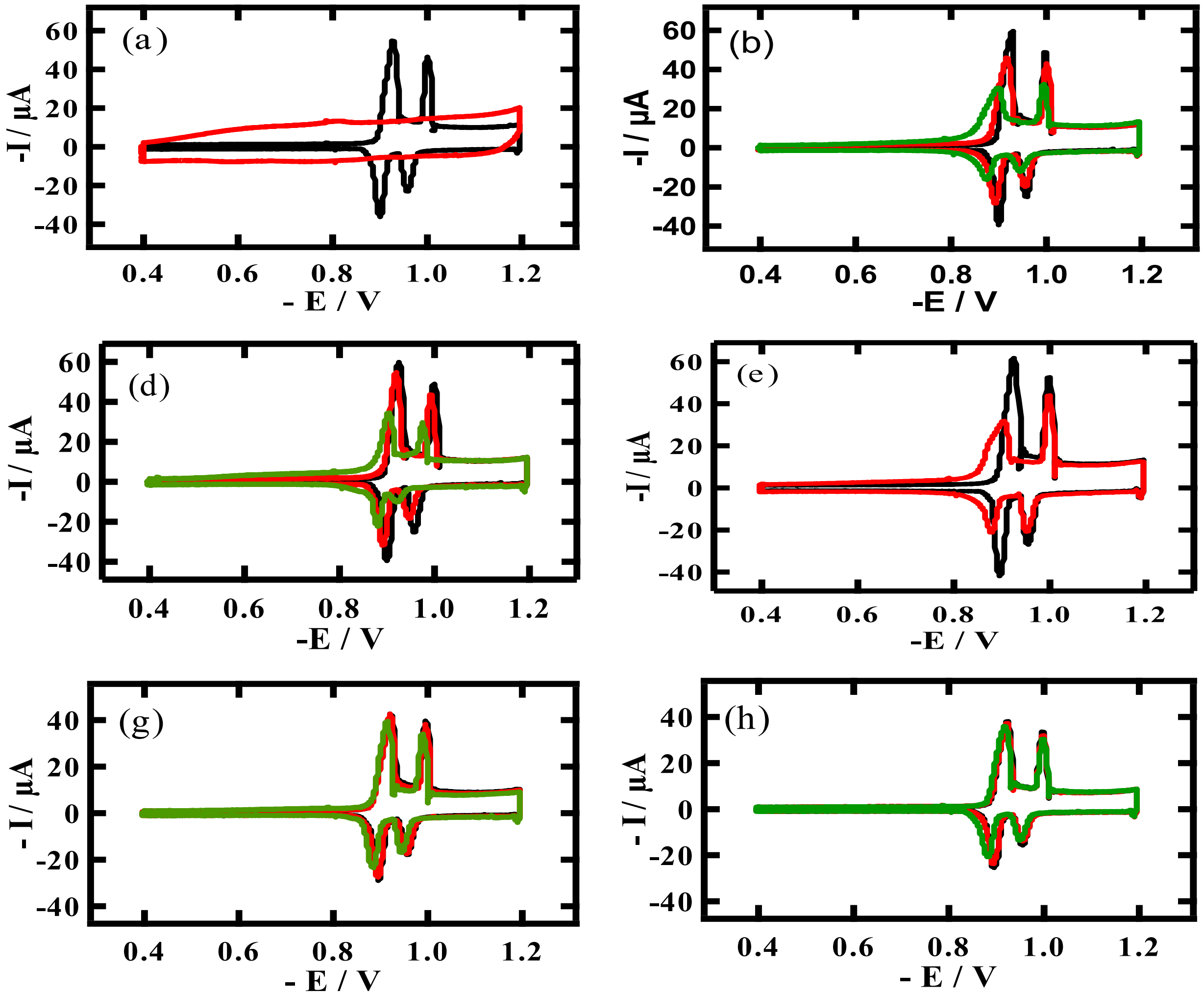
$\eta \propto 1/\sqrt{(B\,K_c)}$

B: bulk compression modulus

K_c: bilayer bending modulus



Experimental Insights into Flavonoid-Biomembrane Interactions (Didem Sanver & Andrew Nelson)



RCVs at 40 V s $^{-1}$ of a pure DOPC-coated Pt/Hg electrode (black line) in the presence of (a) **quercetin** (b) naringenin (c) kaempferol (d) hesperetin (e) **tiliroside** (f) (+) – catechin (g) **rutin** (h) naringin at concentrations of 10 μmol dm^{-3} (red line) and 35 μmol dm^{-3} (green line) in PBS at pH 7.4.

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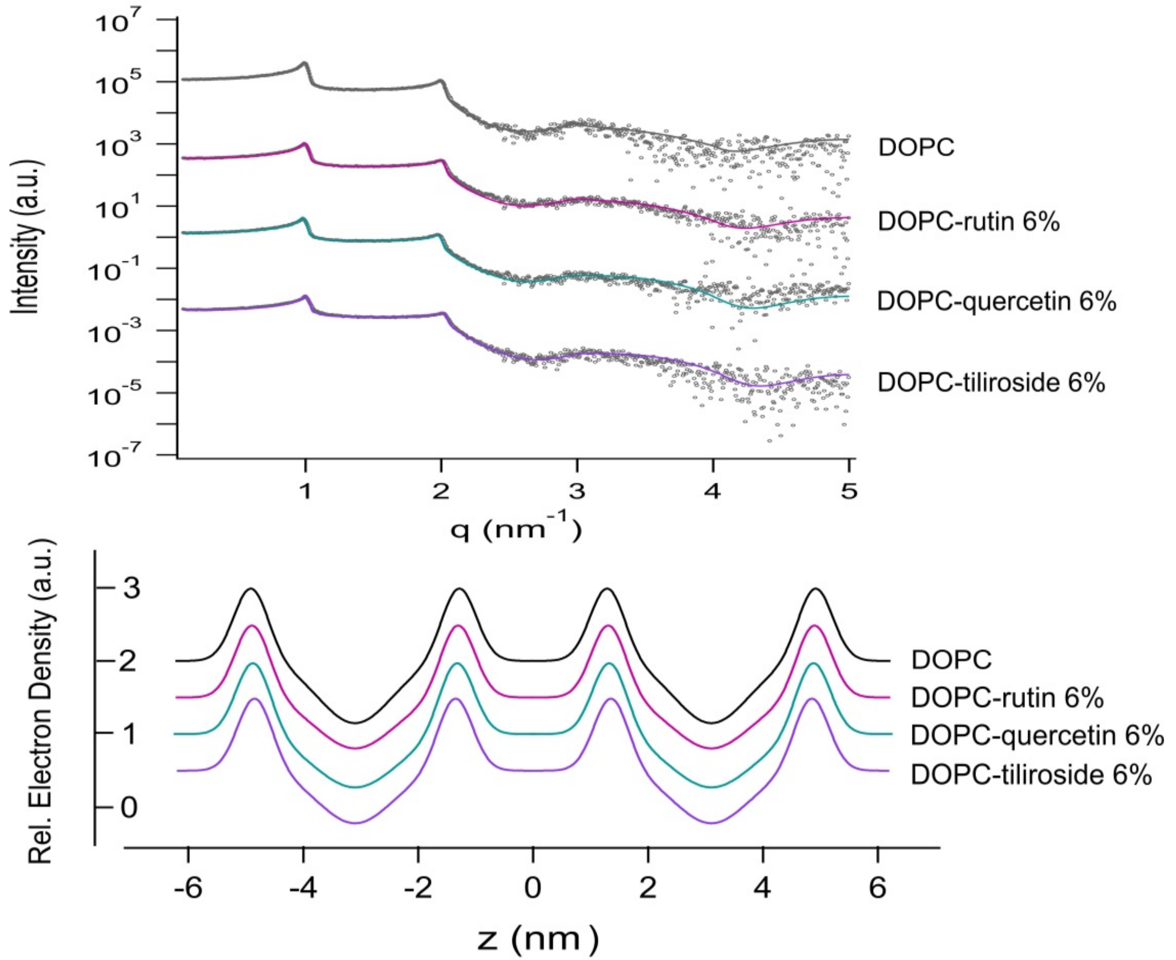
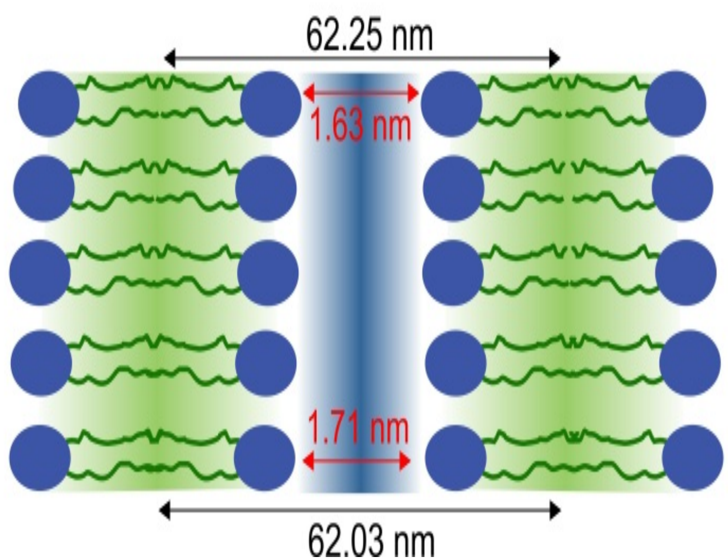
Structural data on pure DOPC MLVs and DOPC with 6% of flavonoids



Structural data on pure DOPC MLVs and DOPC with 6% of flavonoids (II)

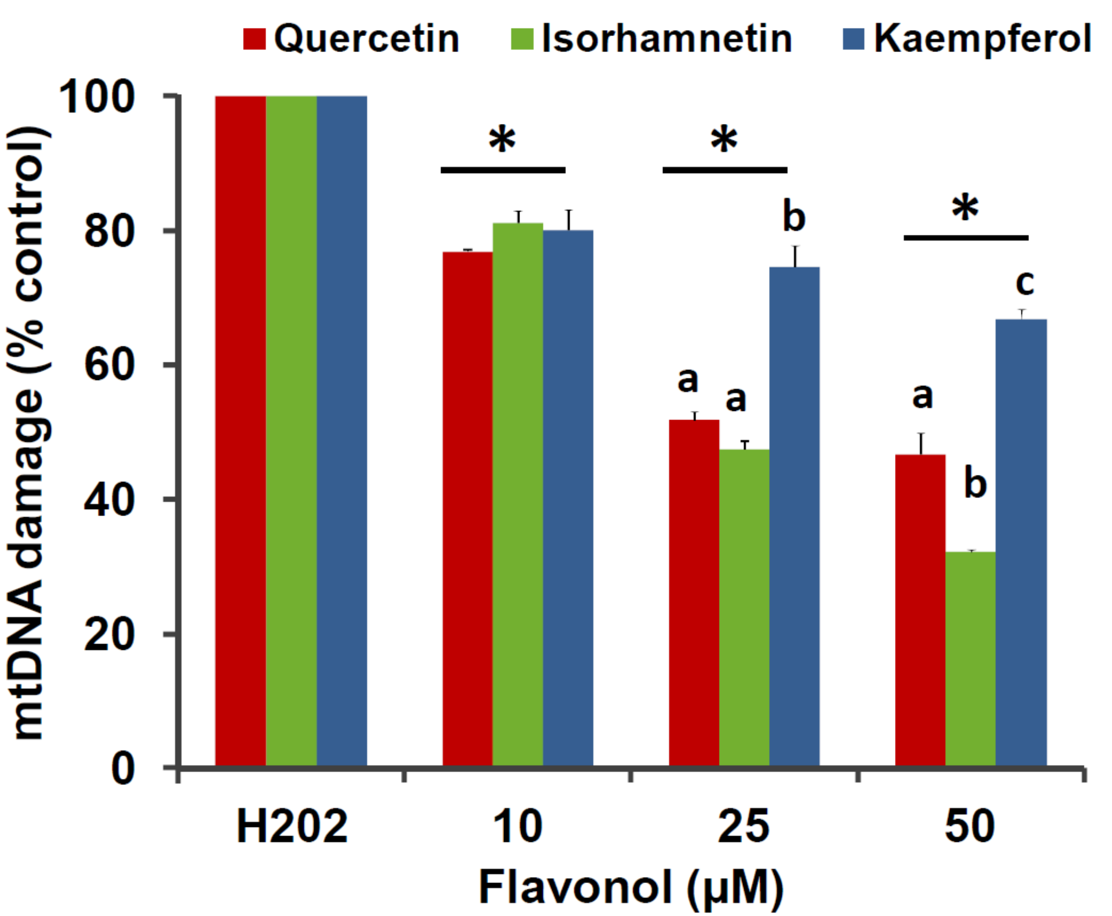
	DOPC	DOPC/Rutin	DOPC/Quercitin	DOPC/Tiliroside
d (Å)	62.48	62.39	63.13	62.03
d_{HH} (Å)	36.2	35.7	35.4	34.8
d_W (Å)	26.3	26.6	27.8	27.7
σ (Å)	5.3	6.1	6.5	6.6

Temperature ramp
15 to 65 °C



6 mol% Tiliroside

Flavonoids' Protective Properties in Mitochondrial Stress (Christine Bosch)



All flavonols markedly **attenuated the hydrogen peroxide induced mtDNA damage** in skin fibroblast cells after a pre-incubation period of 24 hours at concentrations of 10µM or higher.

At 25 and 50µM, **isorhamnetin and quercetin further reduced mtDNA damage to 47-52% and 32-47%,** respectively, whereas kaempferol showed a maximal reduction to 67% mtDNA damage.

Inhibition of H_20_2 generation by isorhamnetin was much stronger compared to quercetin and kaempferol.

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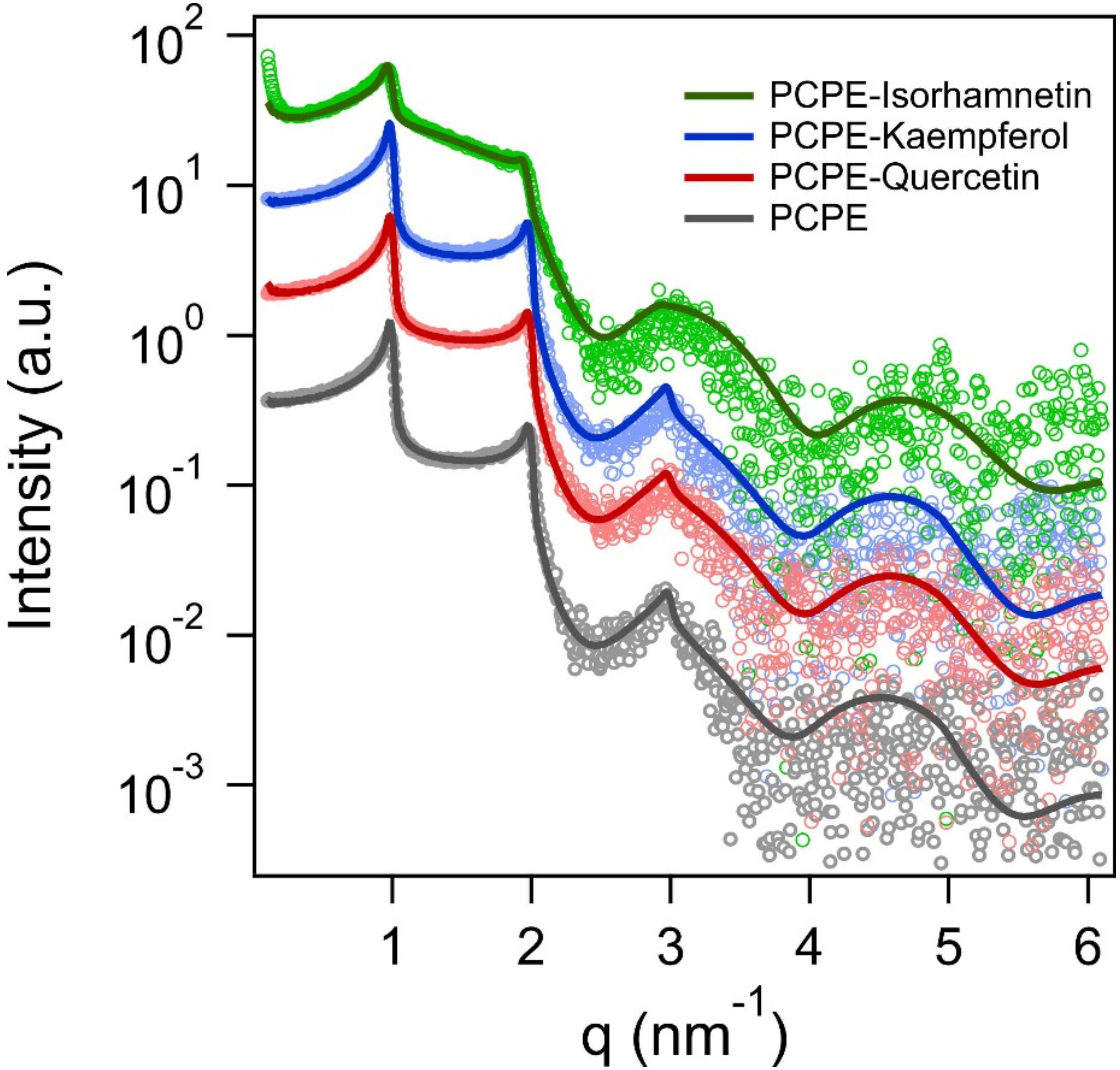
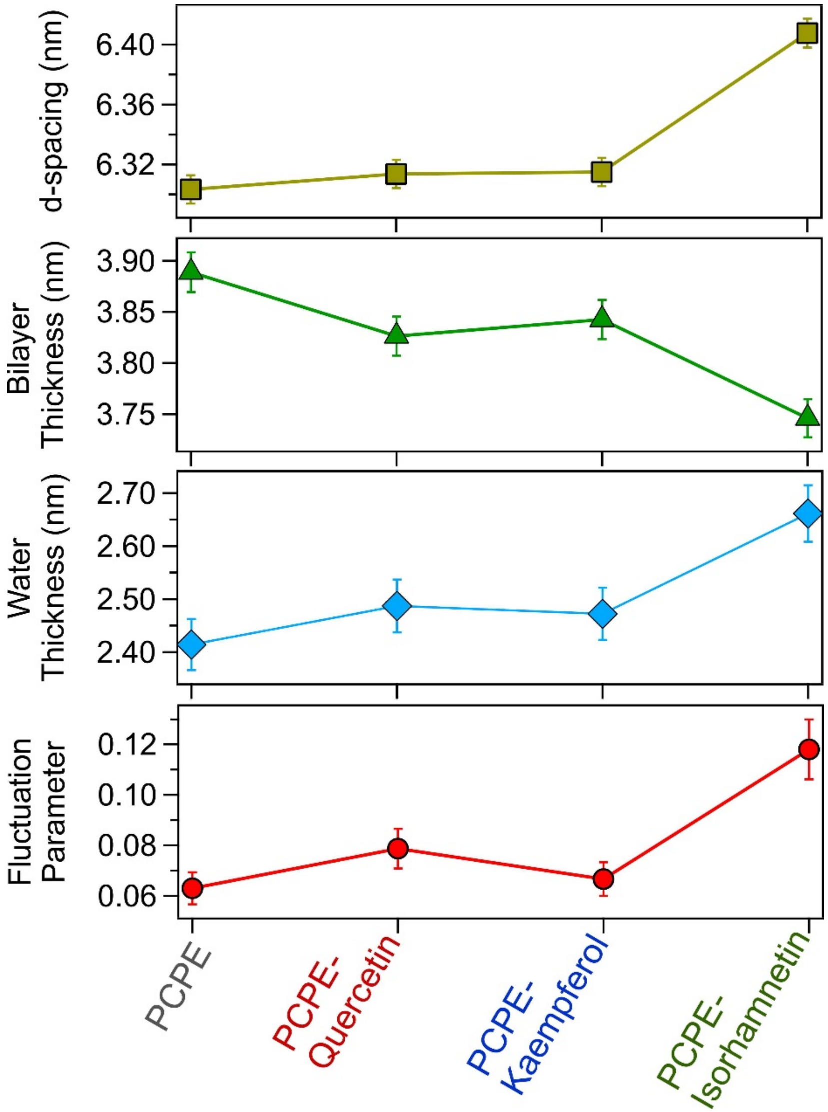
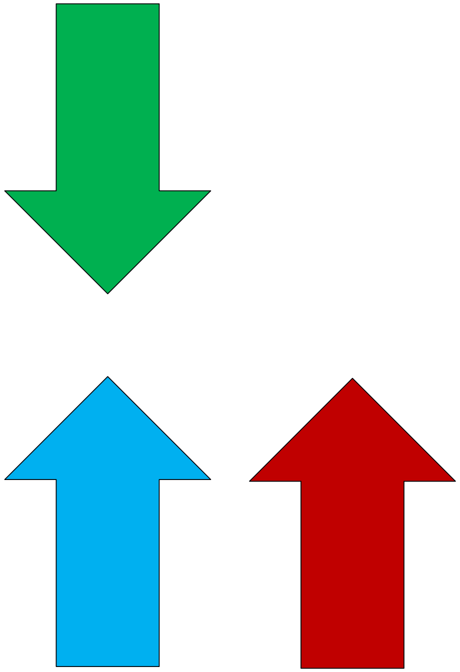
Global Fitting of the Mitochondrial Model Membranes



Structural Results on the Structural Influence of Flavonoids





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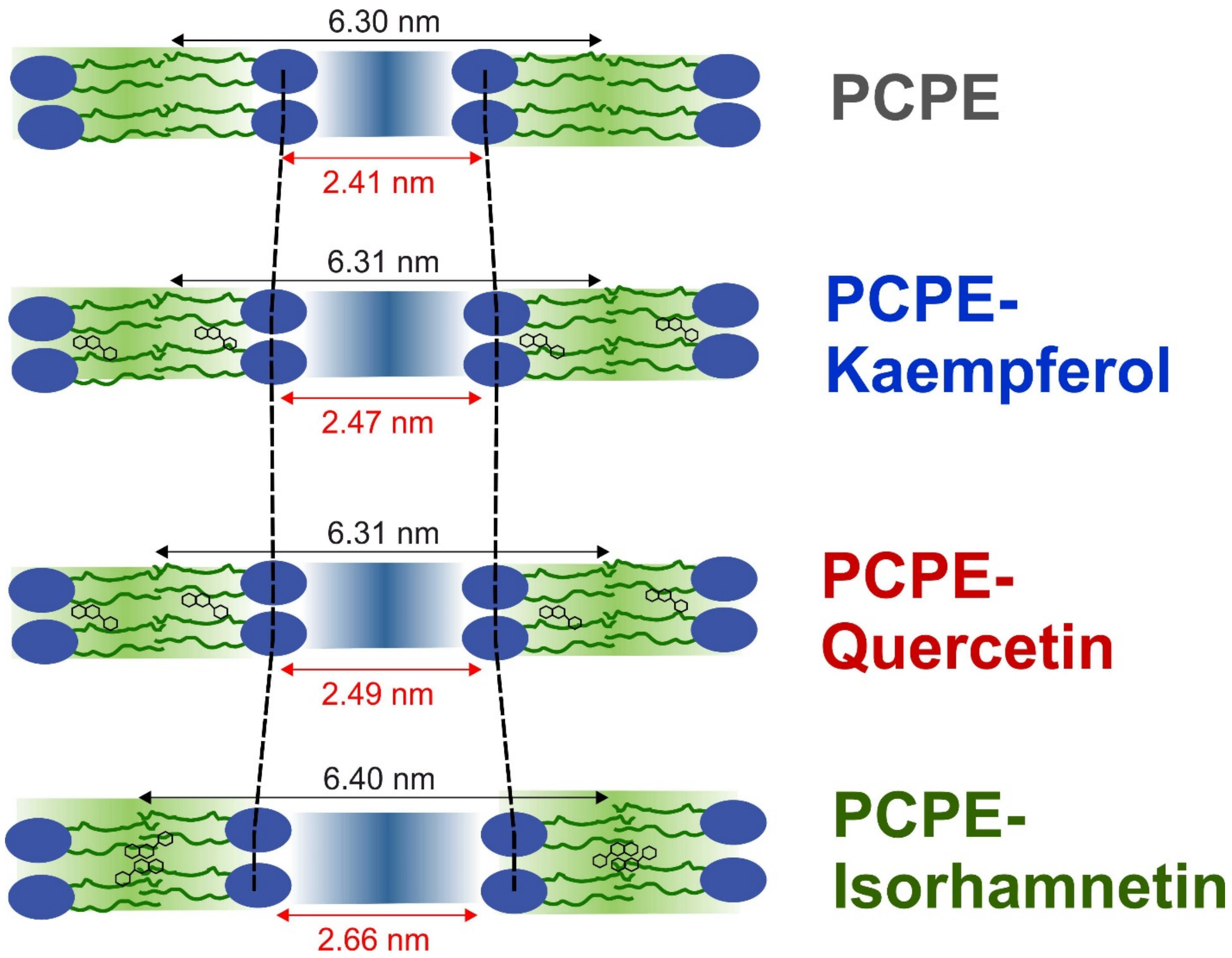
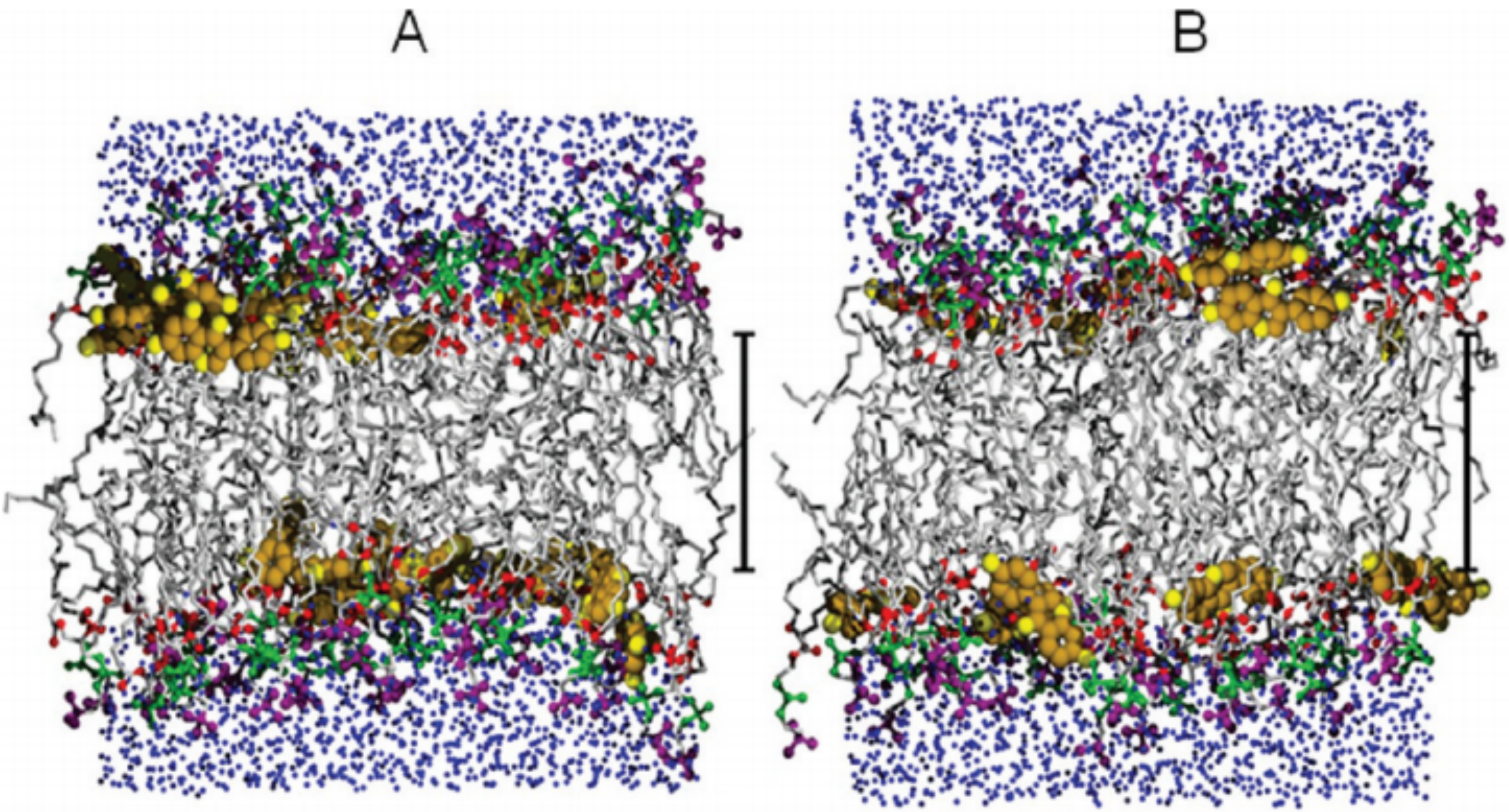
Overview on Membrane Fluidization



Outlook: Combining Structural & Simulation Data



(A) DOPC/20 mol% Genistein
(B) DOPC/ 20 mol% Daidstein

John Nagle: dx.doi.org/10.1021/jp211904j | J. Phys. Chem. B 2012, 116, 3918−3927

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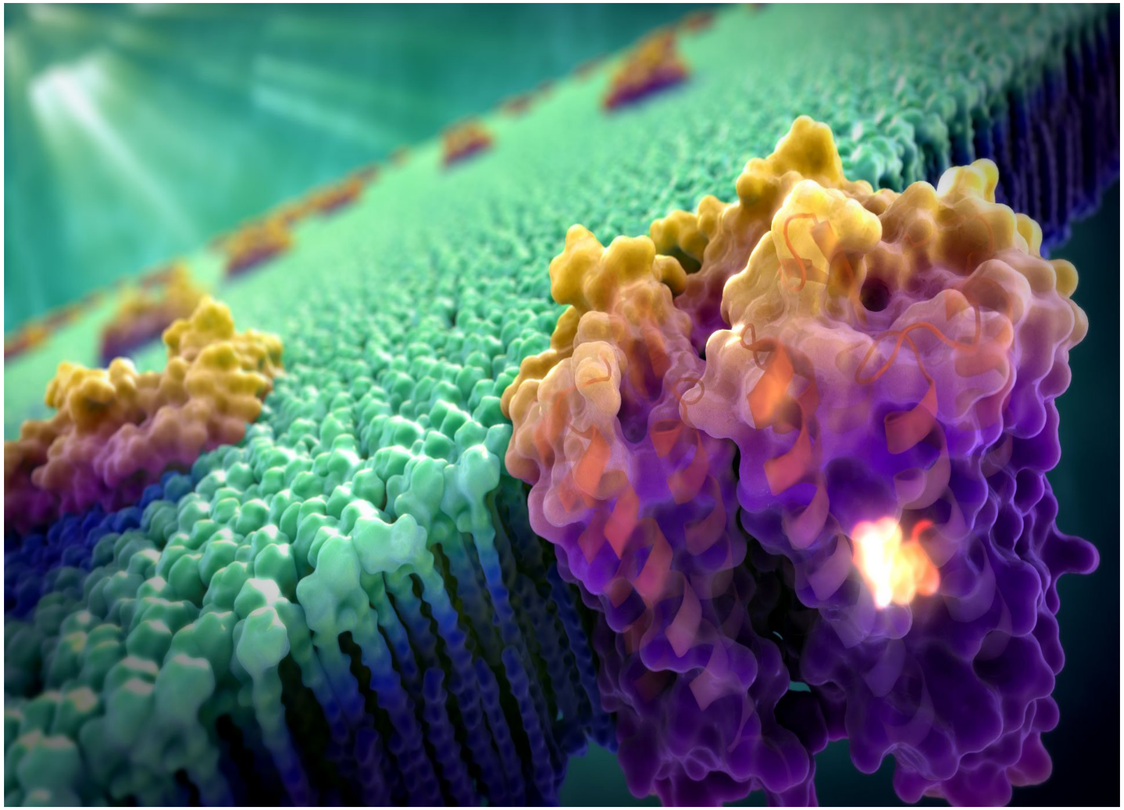
Thank You for Your Attention!



Taken from: CELAPREV.COM
Intelligent Cell Health

My special
thanks go to:

Christine Bosch, Didem Sanver and Amin Sadeghpour
School of Food Science & Nutrition, University of Leeds, UK

Andrew Nelson
School of Chemistry, University of Leeds, UK