Structure study of peptide-lipid membrane interactions

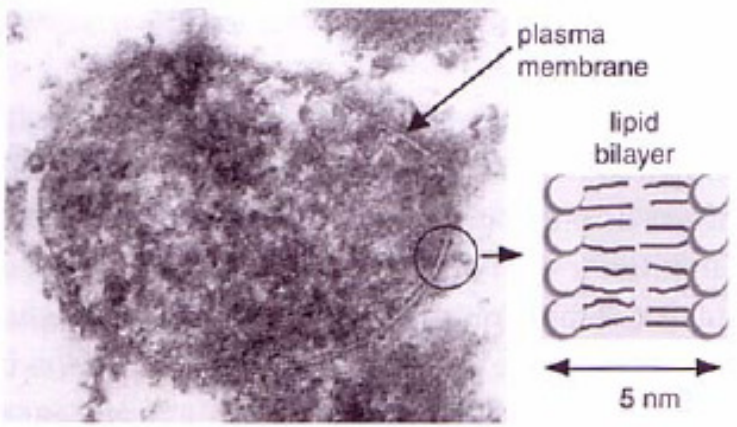
Pia Kjaer Nielsen

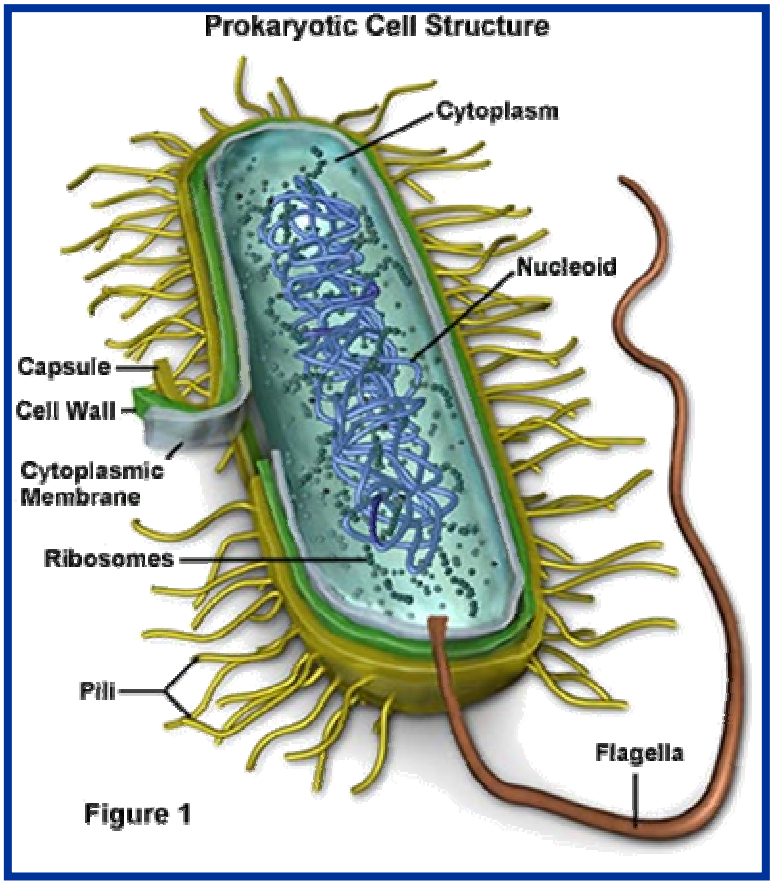
University of Southern Denmark, Odense
Department of Physics and Chemistry & MEMPHYS

DESY summer student 2006, HASYLAB

Lipid - an essential biomolecule

The border of all cells is constituted by **lipid membranes**





Structure of a phospholipid

Glycerol

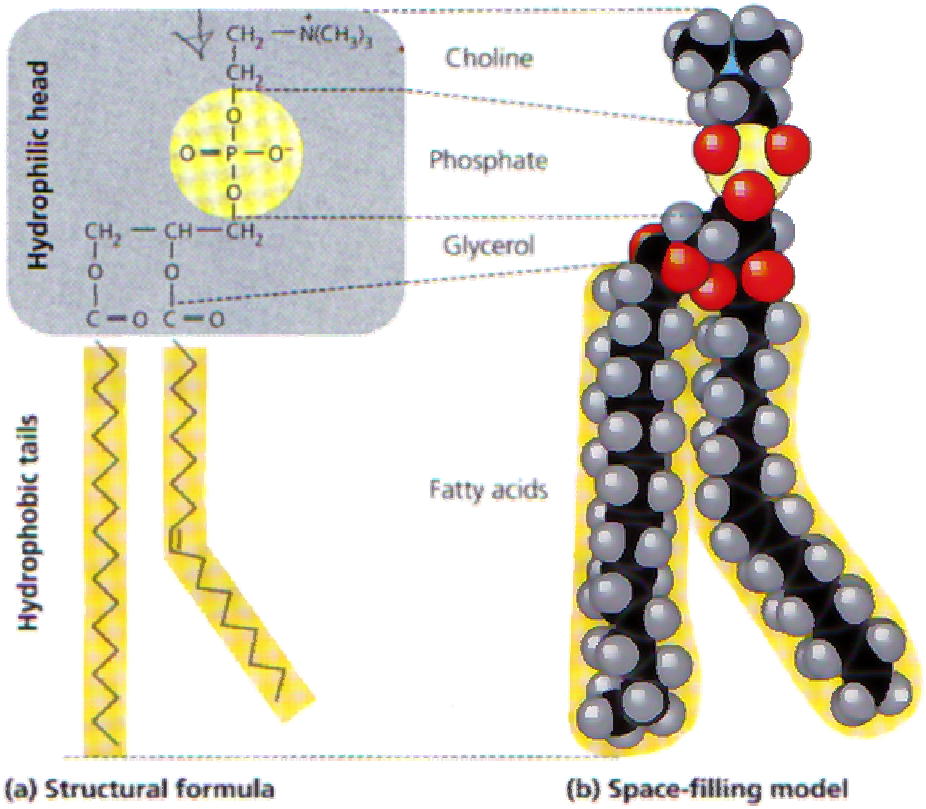
Tails: Fatty acid chains

Phosphate

Head:

- glycerol
- choline
- ethanolamine
- ...

(all polar, some charged)

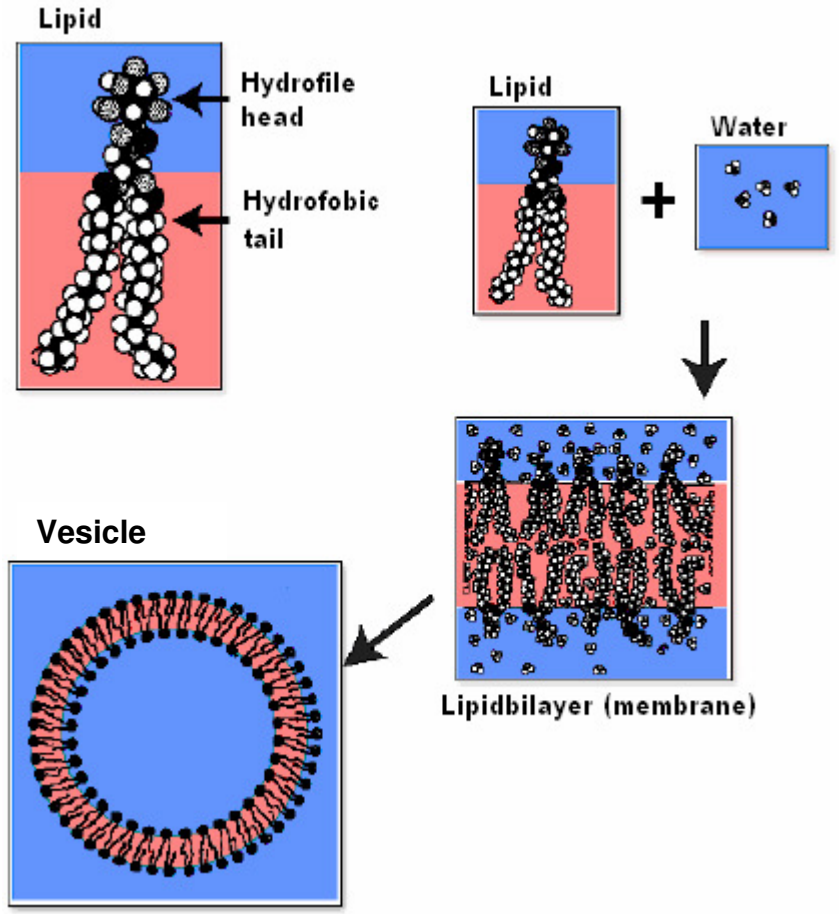


Self assembly

Lipids are amphiphiles

⟶

they form
self-assembled structures
in aqueous solution.



Aggregates

Lipid	Critical packing parameter $v/a_0 l_c$	Critical packing shape	Structures formed
Single-chained lipids (surfactants) with large head-group areas: *SDS in low salt*	< 1/3	Cone	Spherical micelles
Single-chained lipids with small head-group areas: *SDS and CTAB in high salt, nonionic lipids*	1/3-1/2	Truncated cone	Cylindrical micelles
Double-chained lipids with large head-group areas, fluid chains: *Phosphatidyl choline (lecithin), phosphatidyl serine, phosphatidyl glycerol, phosphatidyl inositol, phosphatidic acid, sphingomyelin, DGDG[a], dihexadecyl phosphate, dialkyl dimethyl ammonium salts*	1/2-1	Truncated cone	Flexible bilayers, vesicles
Double-chained lipids with small head-group areas, anionic lipids in high salt, saturated frozen chains: *phosphatidyl ethanolamine, phosphatidyl serine + Ca^{2+}*	~1	Cylinder	Planar bilayers
Double-chained lipids with small head-group areas, nonionic lipids, poly (*cis*) unsaturated chains, high *T*: *unsat. phosphatidyl ethanolamine, cardiolipin + Ca^{2+} phosphatidic acid + Ca^{2+} cholesterol, MGDG[b]*	> 1	Inverted truncated cone or wedge	Inverted micelles

size and charge of the head and length of the tail differ

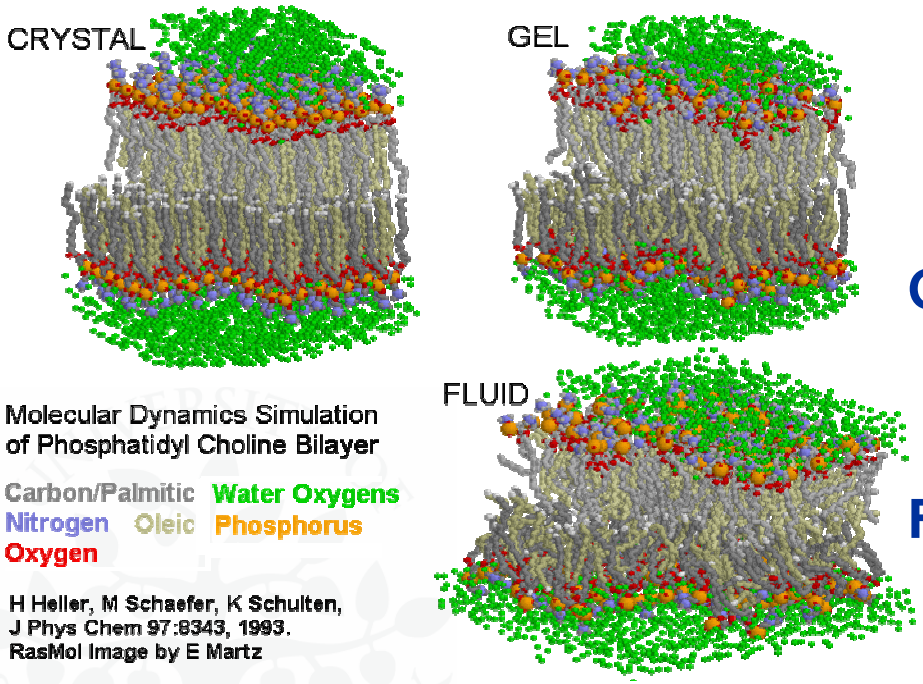
→

lipid molecules exhibit diverse average molecular shapes

→

form aggregates of various structural properties

Lipid phases



Solid: long range order (crystalline), almost no mobility

Gel: less order, more mobility

Fluid: only short range order, high mobility

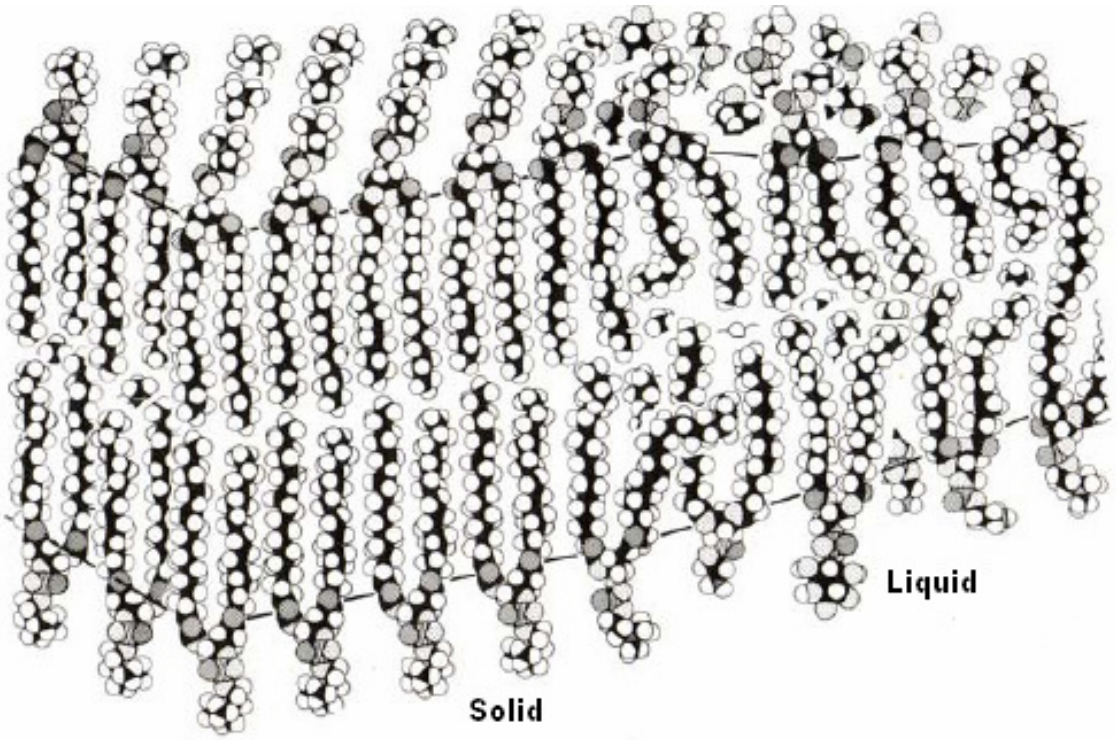
Phase transitions of lipid membranes

Lipid membranes exhibit phases transitions depending on changes in

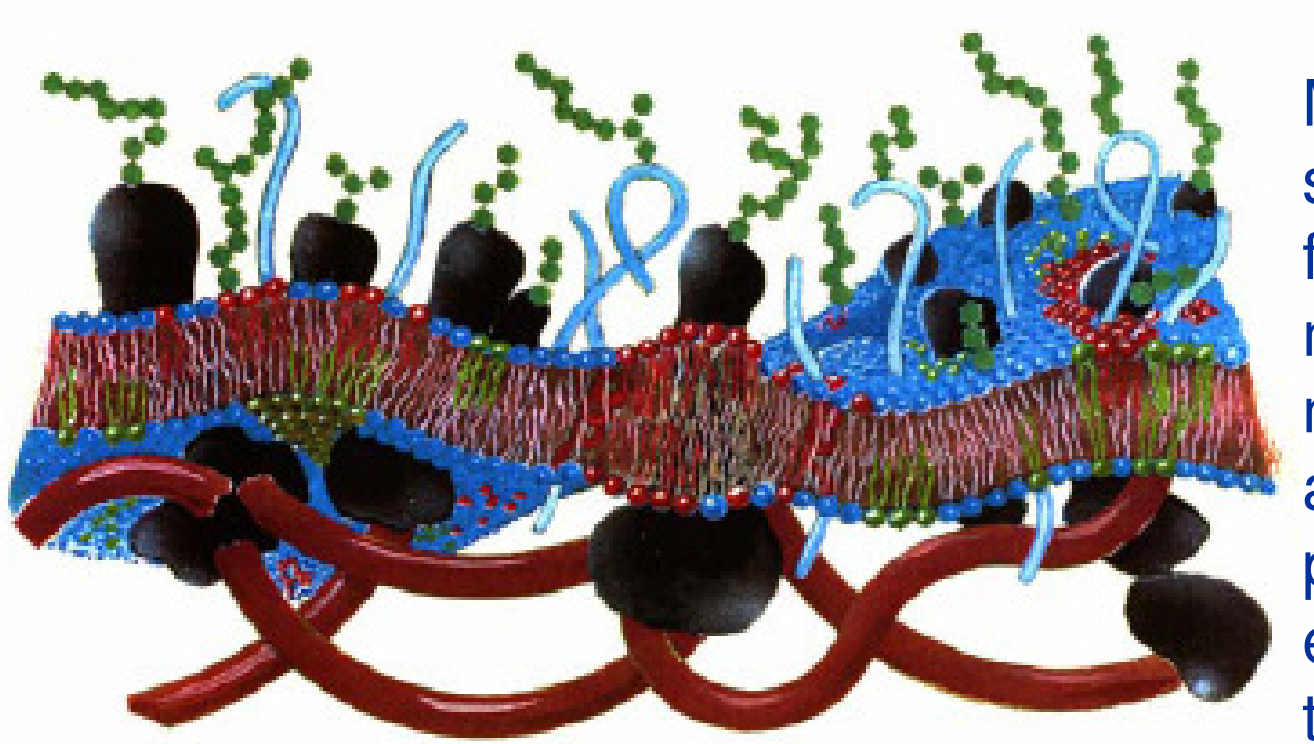
- **temperature ("thermotropic")**
- **composition ("lyotropic")**

or

- **pressure ("barotropic")**



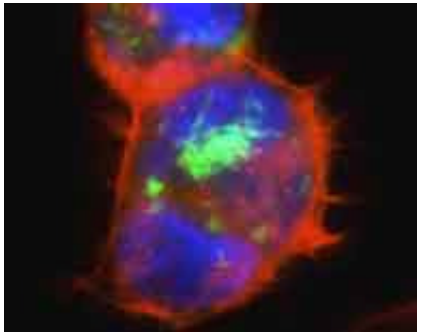
Membrane protein



Membranes may serve a matrixes for non-lipid molecules: e.g. macromolecules as membrane proteins, that are embedded across the bilayer

The need for new drugs

Due to the increasing resistance towards penicillin and other antibiotics, the search for new antibacterial drugs has started.



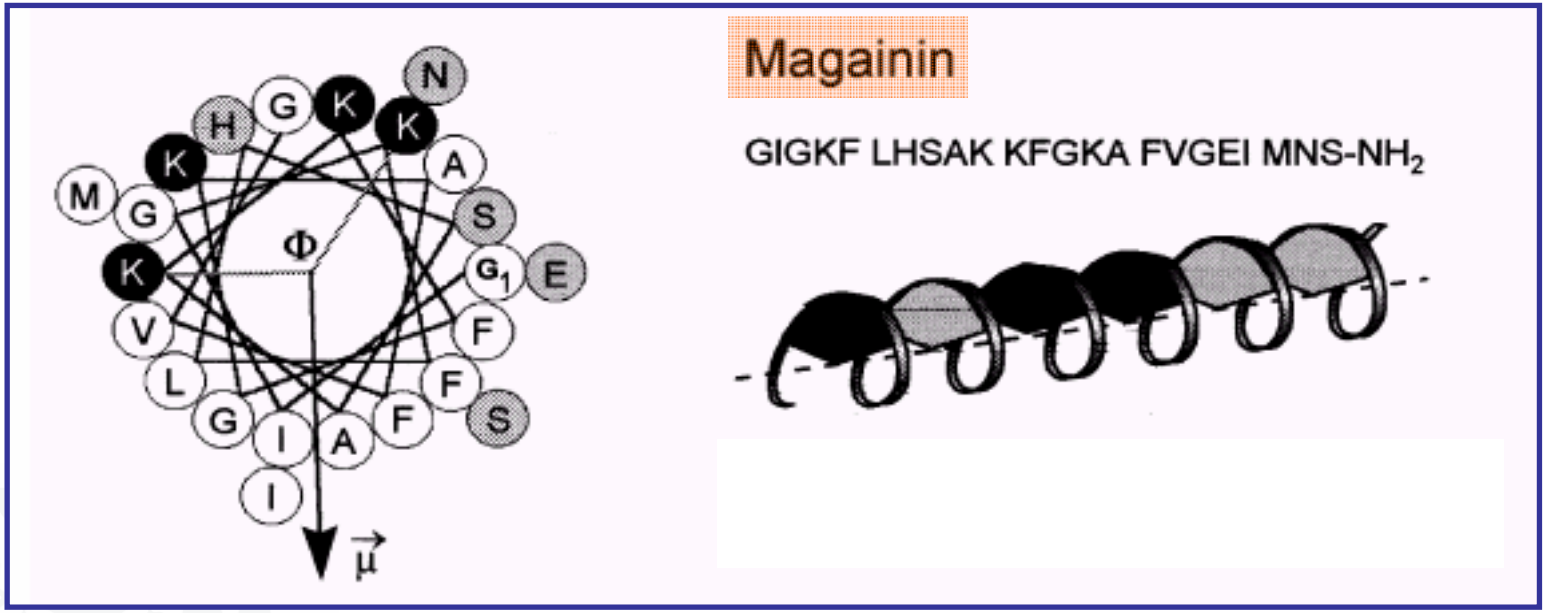
Green Salmonella bacteria

candidates: membrane active peptides as, e.g. natural toxins which:

cholera disease bacterium

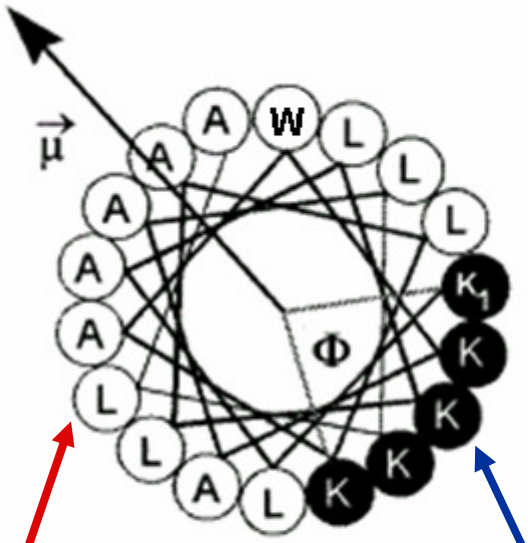
- **interact with membranes by being amphiphilic**
- **cause membrane breakdown by inducing leakage**

Magainin
(a natural toxin from the frog)



- **Very toxic**
- **Defense against surrounding bacteria**
- **Potential antibiotic**

Artificial counterpart: KLA class peptides



KLA - Model

KLALK LALKA LKAAL KLA-NH_2

hydrophobic face

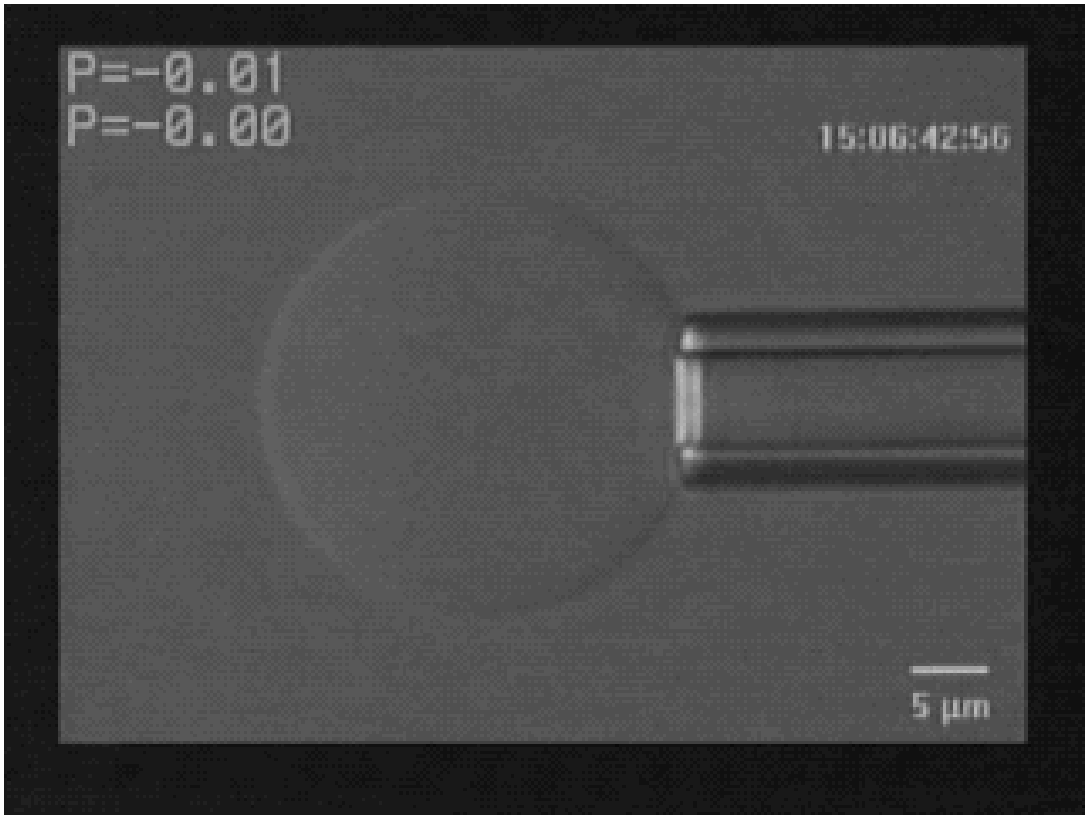
hydophilic face

KLA1 in a membrane

highly membrane active and toxic:

potential drug for

- **anti-bacterial activity**
- **cancer therapy**

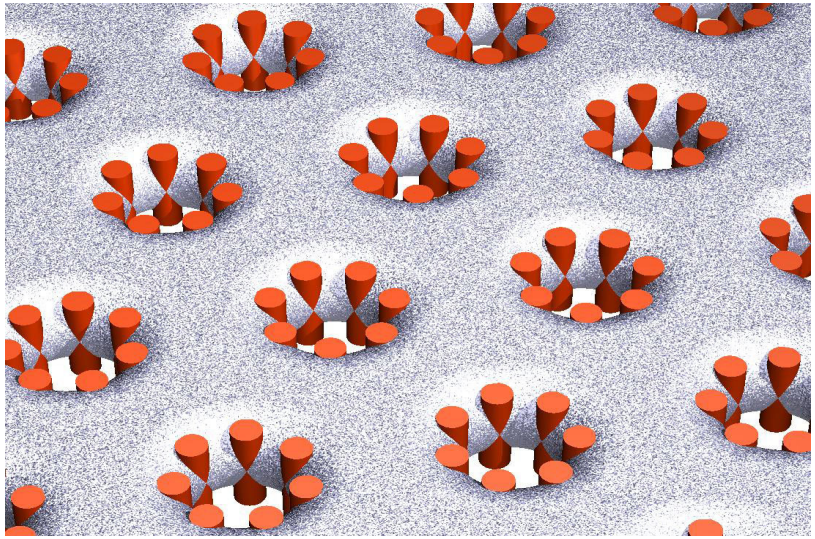


peptide induces leakiness

- vesicle is squeezed empty upon slight suction !!!

The Mystics of KLA

Strange enough!

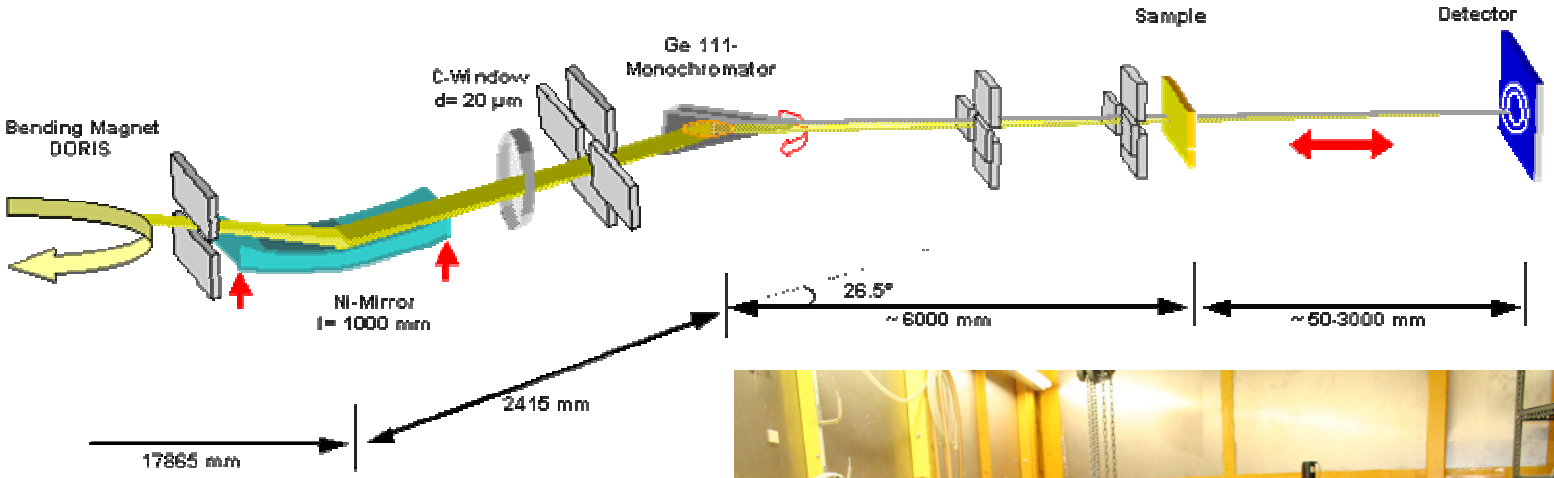


- **the membrane persists**
- **but forms a pore sieve !**

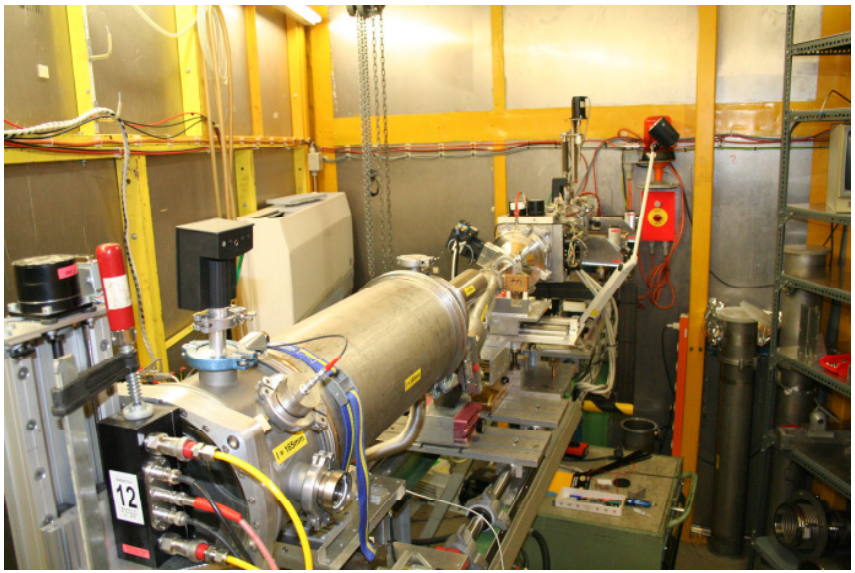
→

- **there must be a structural change within the membrane**

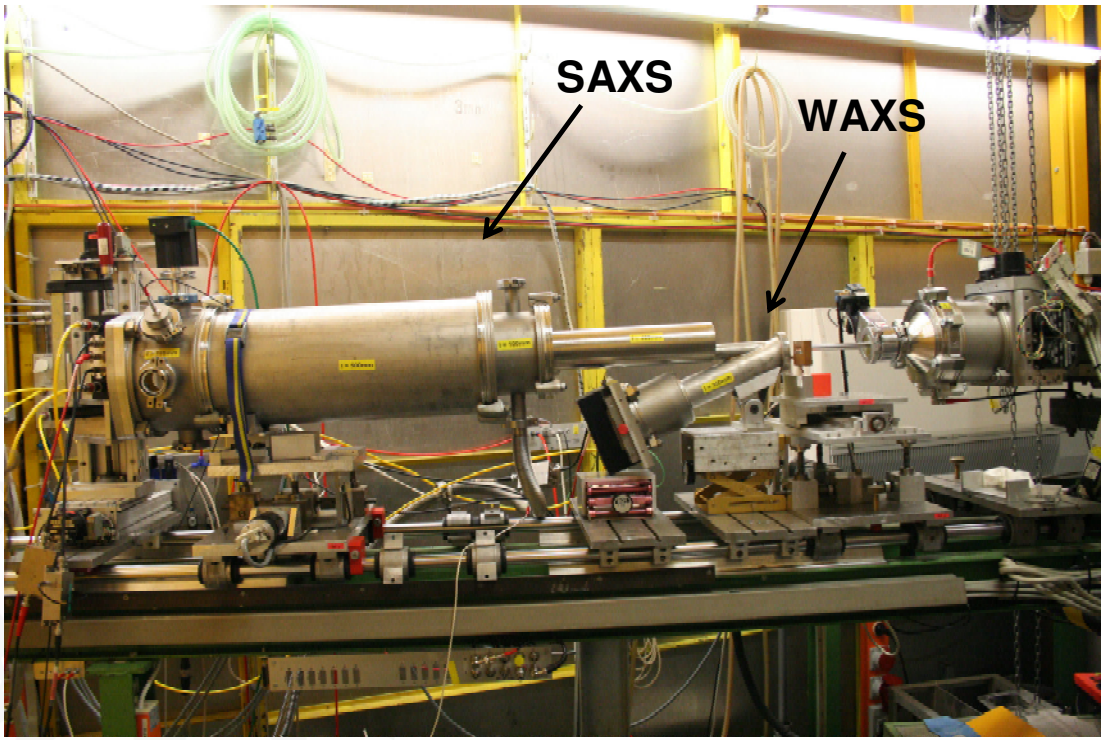
Beamline A2



- Bending magnet beamline
- 8 keV
- SAXS and WAXS



SAXS and WAXS



Small Angle X-ray Scattering:

- Large distance from sample to detector
- Detects larger structures in the sample

Wide Angle X-ray Scattering

- Small distance from sample to detector
- Detects smaller structures in the sample

Scattering

Wave: Green

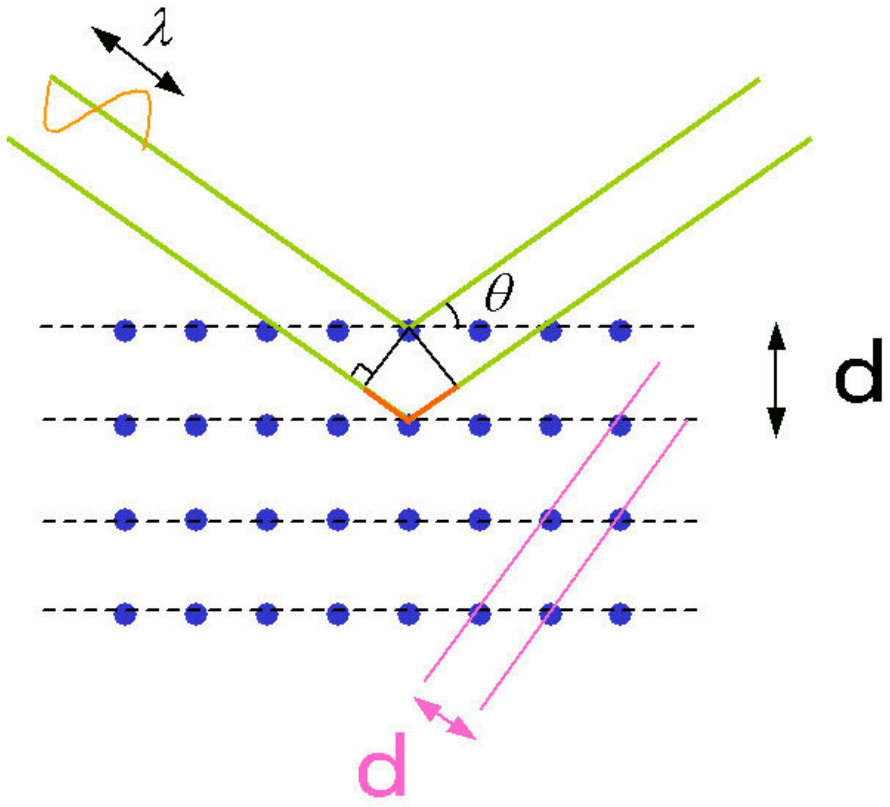
SAXS: Black

WAXS: Pink

Bilayers: blue dots

d = 1/s

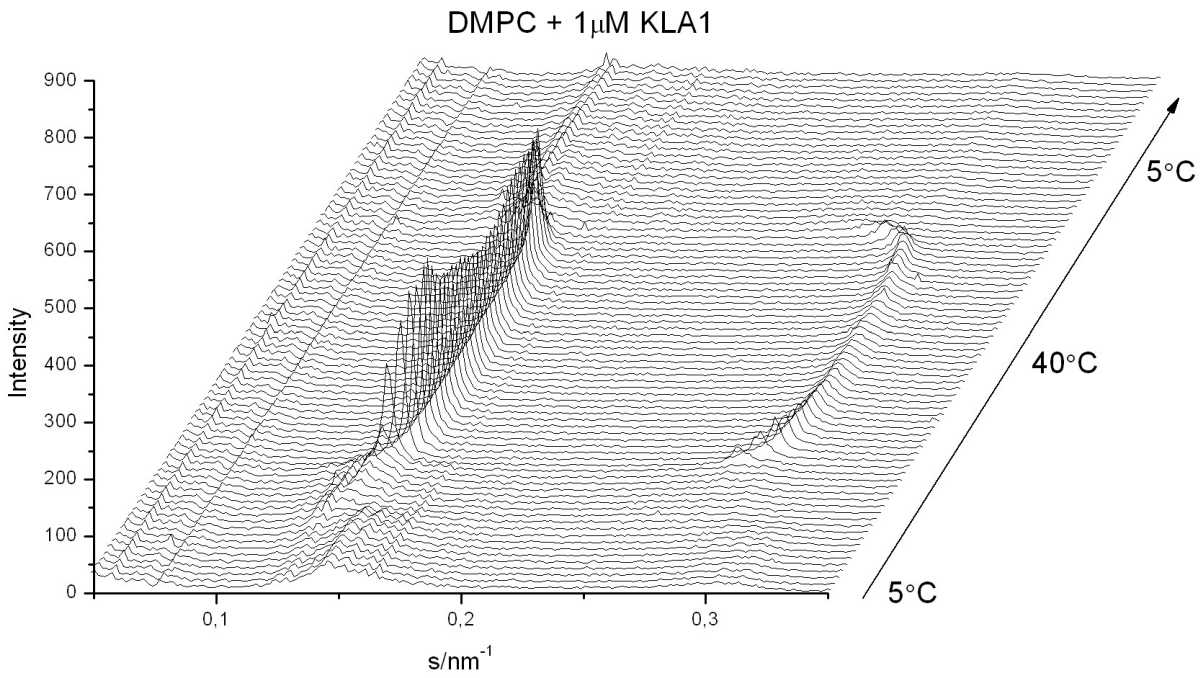
λ = 2d sin(θ)



The experiment

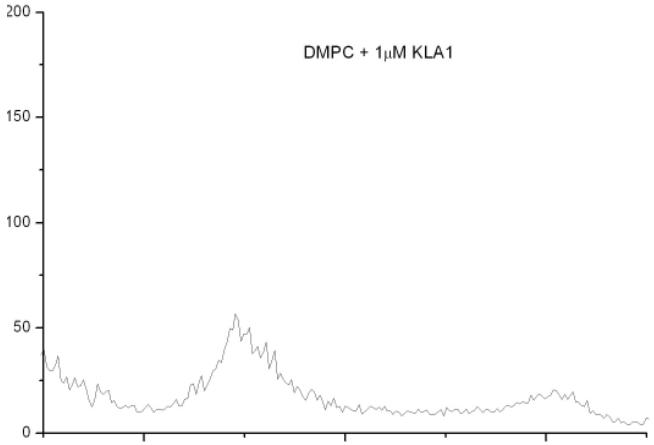
- Samples: Lipids and peptide in aqueous suspension
- Measured with change in temperature: 5ºC to 40ºC and back to 5ºC

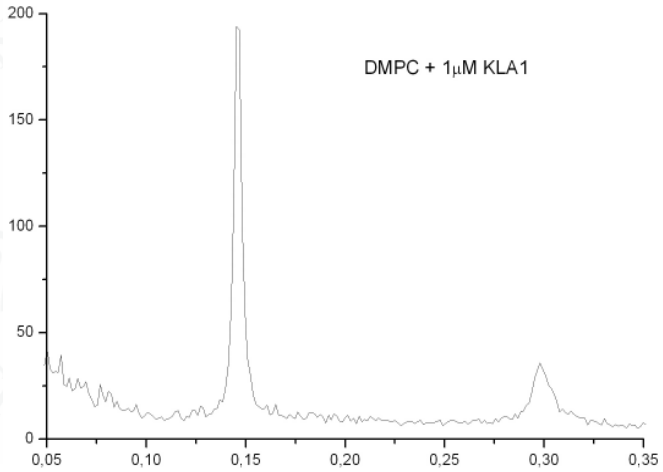
Diffraction pattern

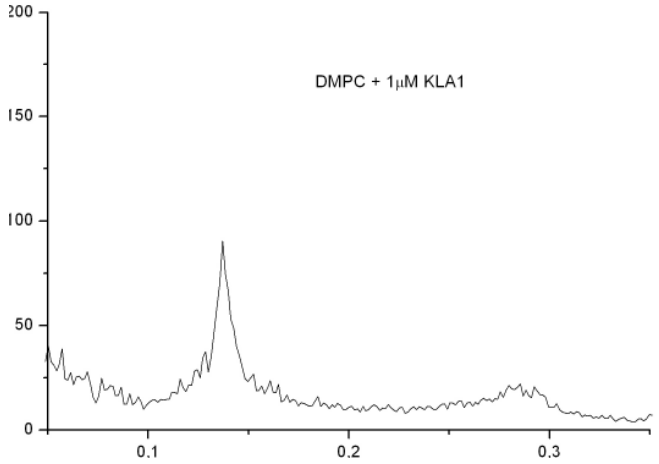


- Change in structure due change in temperature
- Reversible structure change
- Lower temperature → larger structure
- Higher temperature → smaller structure

Analysis







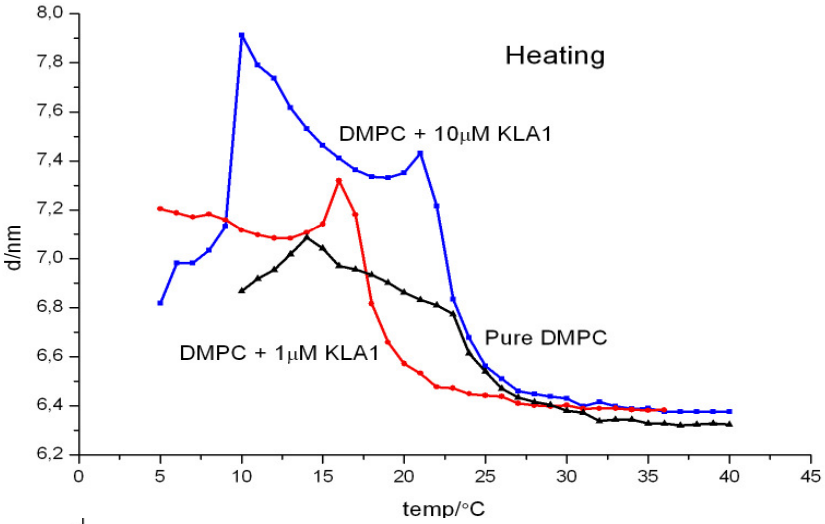
Comparing patterns of different temperatures

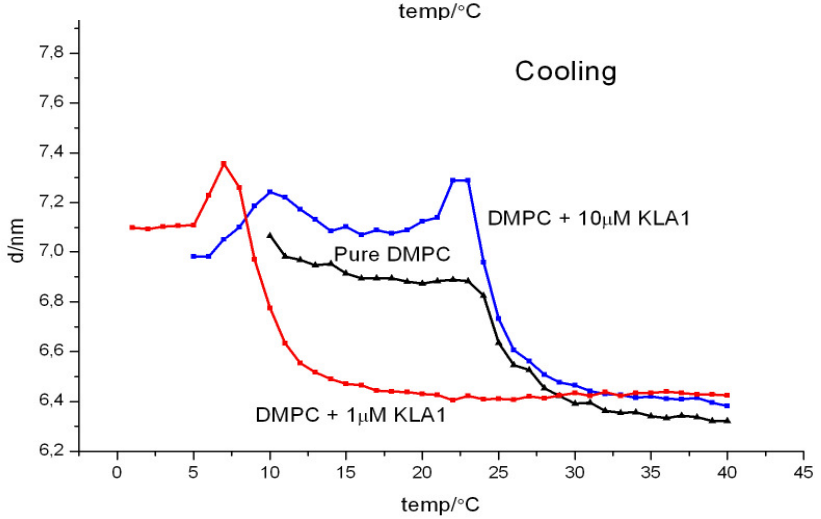
- **before transition**
- **during transition**
- **after transition**

Analysis

Comparing the heating and cooling of the samples shows:

- **Change in structure – size of bilayer**
- **Change in transition temperature**
- **Change in phase transition**





Thanks

- **Dr. Sérgio S. Funari**
- **Ms. Maria Hanulova**
- **Prof. Beate Klösgen**

…and Cornelius for the coffee and Monika for moral support

Thank you for your attention