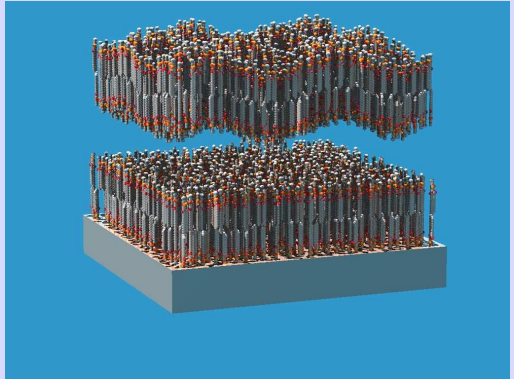


Fluctuations of a single floating lipid bilayer: a specular and off-specular reflectivity study

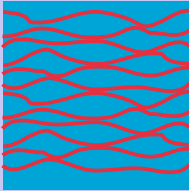
Thierry CHARITAT

Institut Charles Sadron
Université Louis Pasteur

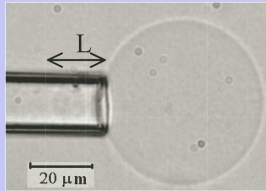
30 August 2005



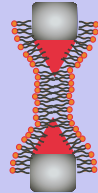
- Introduction
- Neutrons specular reflectivity study (ILL D16-D17)
with E. Bellet-Amalric, G. Fragneto, F. Graner (Grenoble),
K.Mecke (Stuggart)
G. Fragneto et al, *Europhys. Lett.* **53** (1) 100-106 (2001).
K. Mecke et al, *Langmuir* **19** (6) 2080-2087 (2003).
- X-rays off-specular study (ESRF BM32 F. Rieutord)
with A. Braslau and J. Daillant (Paris)
J. Daillant et al, *PNAS*, **102** (33), 11639-11644 (2005).



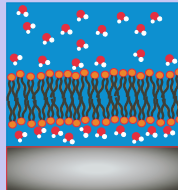
Multilamellar stack



Vesicle



Black Lipid Membrane



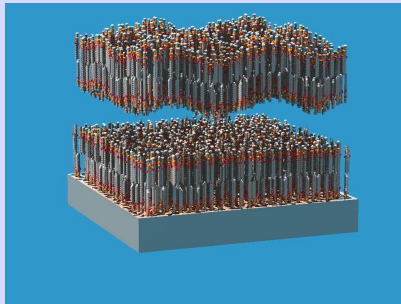
Supported bilayer

We are looking for :

- a supported membrane with controlled position :
 - to use molecular level probe as Neutrons, X-Rays, FRAP...
 - to work on a unique, accessible and non-confined bilayer
- not to strongly bound to the substrate to investigate dynamical properties

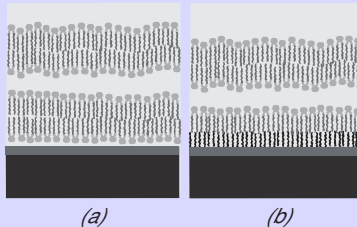
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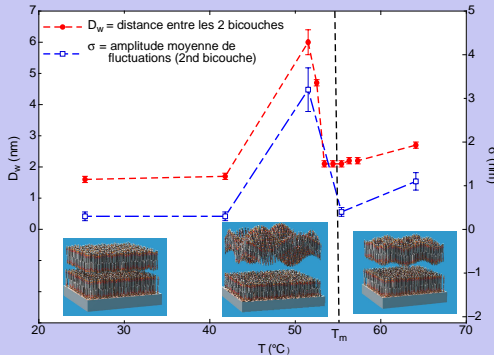
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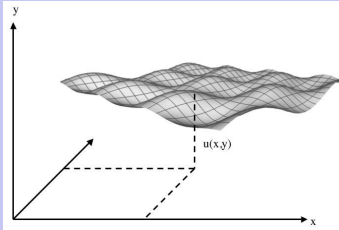
^b A. V. Hughes *et al* *J. Phys. Chem. Chem. Phys.*, 53 :100, 2001.

Going from Gel Phase ($T < T_m$) to Fluid Phase ($T > T_m$)



- Large swelling around transition temperature T_m .
- D_w increase correlated to σ_f increase.

Large increase of thermal fluctuations?

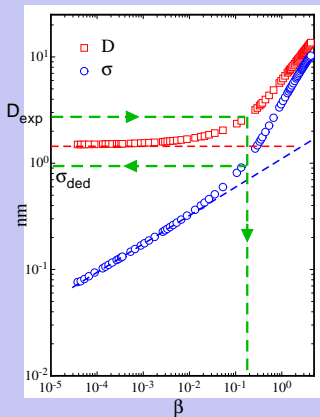


$$\mathcal{H} \simeq \frac{1}{2} \kappa \int dS (\Delta u(x, y))^2$$

in an external potential $\mathcal{U}(z)$

- Symmetrical potential : Helfrich repulsion (*Z. Naturforsch.* **1978**) $\mathcal{U}_{Helf} \simeq \frac{(k_B T)^2}{\kappa} \frac{1}{d^2}$
- Asymmetrical potential in large fluctuations regime : renormalisation theory (S. Leibler et al *PRL* **(1986)**).
- Asymmetrical potential in small fluctuations regime : self-consistent approach (with K. Mecke)

$$z \text{ and } \sigma \text{ controlled by } \beta = \frac{(k_B T)^2}{A\kappa}$$

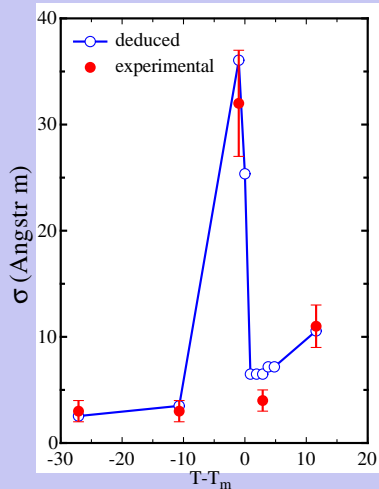


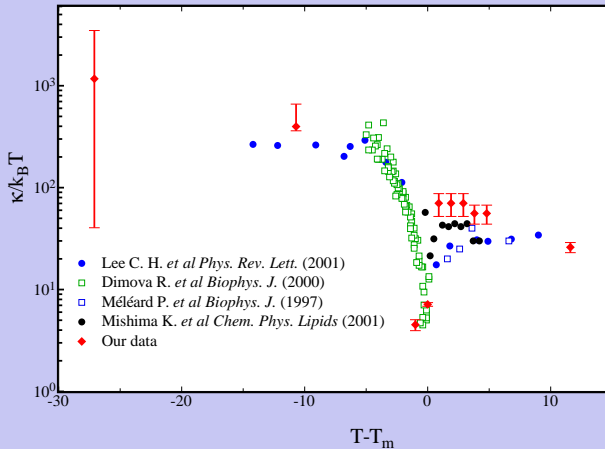
$$D_{exp} \Rightarrow \beta \Rightarrow \kappa$$

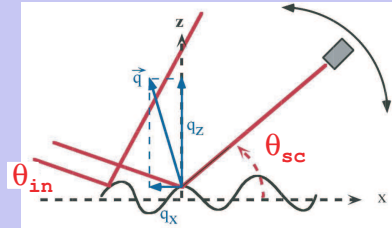


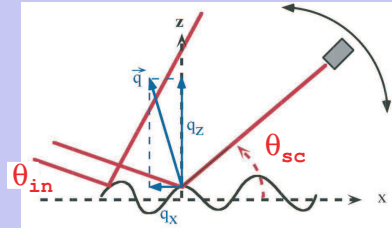
$$\sigma_{exp} \Leftrightarrow \sigma_{deduced}$$

Comparaison σ_{exp} VS $\sigma_{deduced}$









$$\frac{d\sigma}{d\Omega} = \frac{d\sigma}{d\Omega}_{spec} + r_e^2 |t_{0,1in}|^2 |t_{0,1sc}|^2 \left\langle \left| \int d^3\vec{r} \delta\rho(\vec{r}) e^{i\vec{q}\cdot\vec{r}} \right|^2 \right\rangle$$

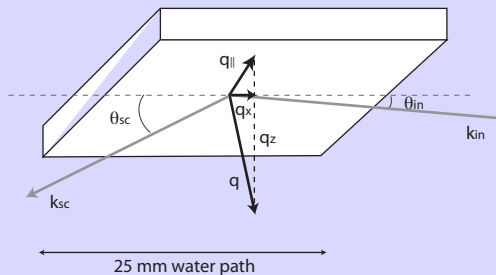
$\langle \delta\rho(r)\delta\rho(r') \rangle \overset{?}{\leftrightarrow}$ holes, protrusion, domains, ripples, fluctuations... ?

thermal fluctuations spectrum :

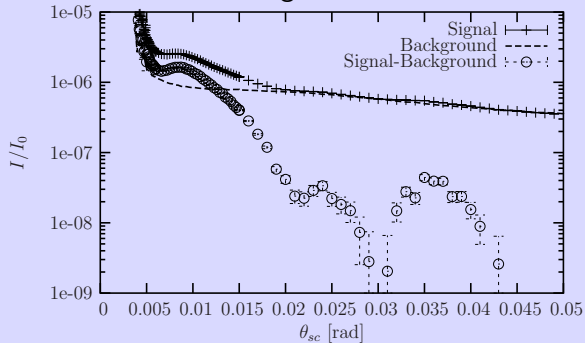
$$\langle \zeta(q_{||}) \zeta(-q_{||}) \rangle = \frac{k_B T}{A + \gamma q_{||}^2 + \kappa q_{||}^4}$$

- ESRF, BM 32, 20 keV ($\lambda = 0.62\text{\AA}$)
- Si substrate
- incidence angle $\Theta_i \simeq 1 \text{ mrad} \lesssim \Theta_c$ to limit diffusion by substrate

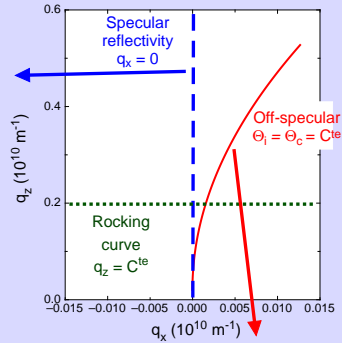
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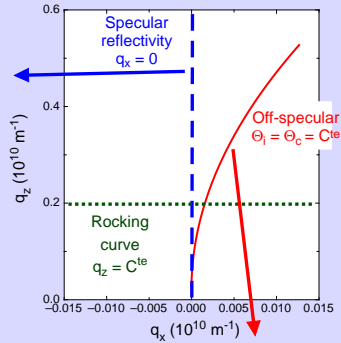
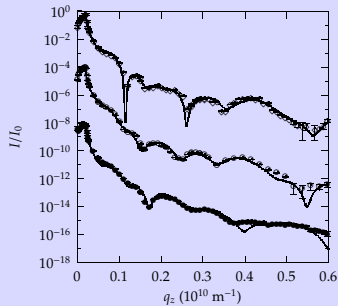


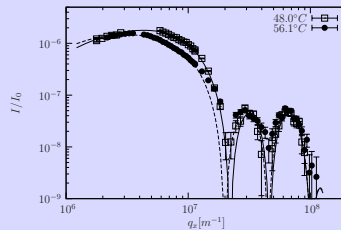
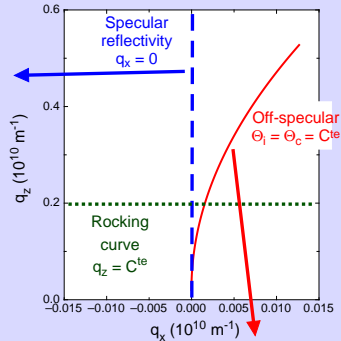
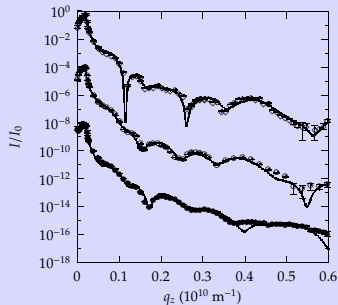
diffusion by bulk larger than diffusion by bilayer
 \Rightarrow difficult background subtraction



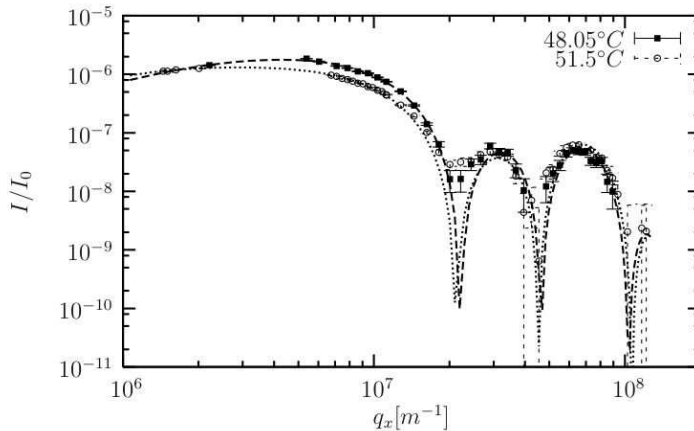
$$I_B(\theta_{sc}) = [I_{sl}(\theta_{sc}) + R(\theta_{in}) \times I_{sl}(\theta_{sc} - 2\theta_{in})] / 2$$







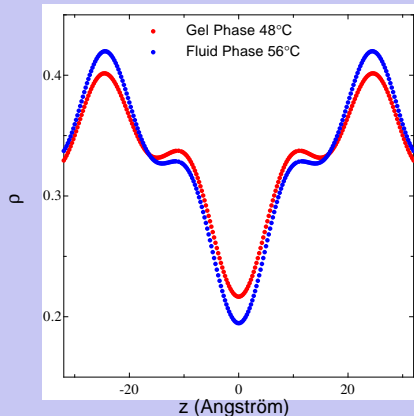
Off-specular experimental results



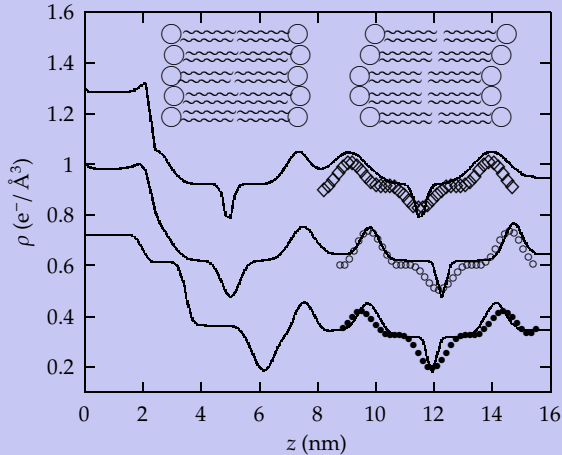
Only sensitive to second bilayer electronic density profile

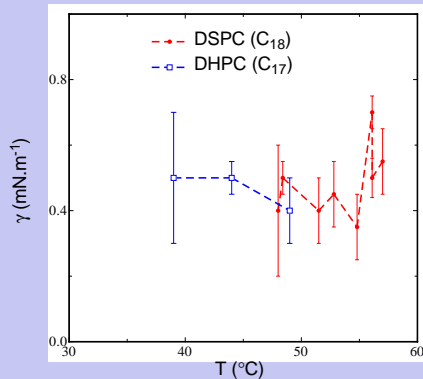
4 first Fourier coefficient

$$\Rightarrow \rho(z) - \rho_w = \frac{F_0}{D} + \frac{2}{D} \sum_{h=1}^{h_{\max}} F_h \cos\left(\frac{2\pi h z}{D}\right)$$



Coherent with specular reflectivity

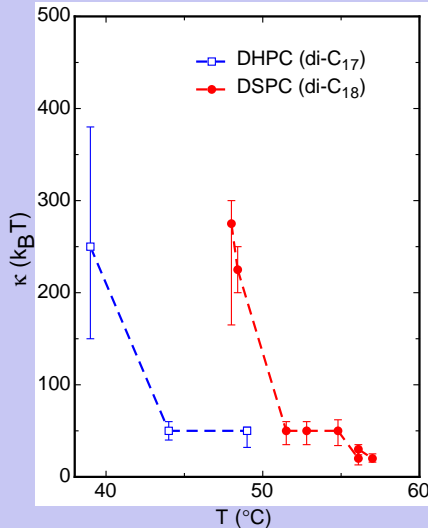




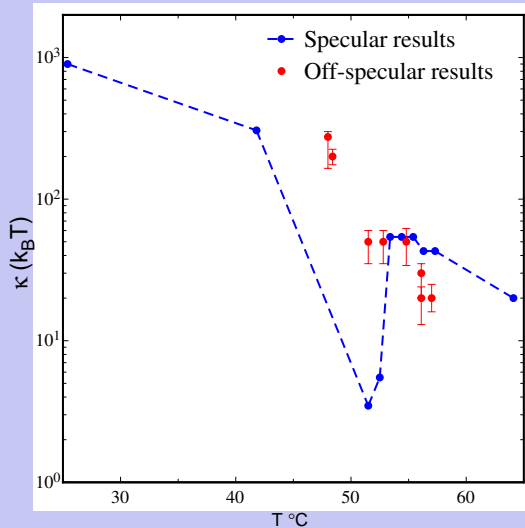
surface tension roughly constant $\sigma \simeq 0.5 \pm 0.2 \text{ mN.m}^{-1}$

- Larger than for vesicles $\sigma \sim 10^{-5} \text{ mN.m}^{-1}$ (Pécrciaux et al)
- Smaller than lysis tension $\sigma_L \sim 5 - 10 \text{ mN.m}^{-1}$

absolute measurement of bending modulus in Gel and Fluid phases



Comparison with value from self-consistent theory



Conclusions

- well controlled (composition, position) *free* supported bilayer
- Off-specular reflectivity : powerful tool to study single bilayer
- Direct measurement of Structure and Fluctuations spectrum

$$1.2 \cdot 10^6 m^{-1} < q < 1.2 \cdot 10^8 m^{-1}$$

(Pécreaux et al $0.5 \cdot 10^6 m^{-1} < q < 7 \cdot 10^6 m^{-1}$)

Perspectives

- precise off-specular study around main transition ;
- going to smaller q to investigate bilayer-substrate interaction ;
- out of equilibrium fluctuations : bilayer under electric field...

- François Graner¹
- Edith Bellet-Amalric^{2,3}
- Giovanna Fragneto, Barry Stidder²
- François Rieutord^{3,4}
- Alan Braslau, Jean Daillant, Serge Mora⁵
- Klaus Mecke⁶

¹ LSP, Université Joseph Fourier, Grenoble,

² ILL-Grenoble,

³ DRFMC-CEA, Université Joseph Fourier, Grenoble,

⁴ ESRF, Grenoble,

⁵ SPEC-CEA, Université Orsay, Paris,

⁶ MPI, Stuttgart