<table>
<tr><td rowspan="1"></td><td>Experiment title: Structure and fluctuations of a single floating bilayer</td><td>Experiment number:
32-2-632</td></tr>
<tr><td>Beamline:
BM 32</td><td>Date of experiment:
from: 23/09/2005 to: 01/10/2005</td><td rowspan="2">Date of report:
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Received at ESRF:</td></tr>
<tr><td>Shifts:
18</td><td>Local contact(s):
Jean-Sebastien Micha</td></tr>
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Report:

The aim of the experiment was to investigate the precise structure and fluctuations close to equilibrium, of a phospholipid membrane in a very ideal case: that of a single bilayer floating 2-3 nm above a first bilayer grafted on a silicon substrate. Previous neutron reflectivity experiments showed that such systems are stable both in the gel and the fluid phases, and that an important swelling occurs around the so-called main transition temperature. We interpret this as a drop of the bending rigidity modulus of the membrane κ [1].

Earlier X-ray experiments (32-2-76, 32-2-89 and 32-2-129) enabled us to show that grazing-incidence X-ray scattering can give access to the thermal fluctuation spectrum of the membrane

$$< z(q)z(-q) >= \frac{k_B T}{U'' + \gamma q^2 + \kappa q^4}$$

where U'' is the second derivative of the membrane-substrate potential, γ the surface tension of the membrane and κ its bending modulus. After rigorous background substraction, we were able to caracterise the membrane fluctuation spectrum over a wide submicrometric range ($1\ \mu m^{-1} < q_z < 0.1\ nm^{-1}$), leading to the determination of γ and κ for the free-standing bilayer, in both the gel and the fluid phases. However, we could only determine an upper limit for the potential experienced by the membrane due to the lack of data at small in-plane wave-vector transfer [2].

One aim of this experiment was to focus on small values of q in order to be able to determine the potential U more precisely and to investigate in detail the gel to fluid transition region, where neutrons showed that important changes occur. The instrumental resolution was improved through the use of smaller slit openings coupled with a larger (50 mm !) sample size. Even though the experiments were carried out using the synchrotron beam in the 16-bunch mode, we were nevertheless able to obtain excellent results.

Experiments were carried out on DSPC bilayers and double-bilayers samples, deposited on silicon substrates. The first layer was always covalently linked to the silicon. All samples, including bare silicon wafers and first grafted layers, were characterised by specular reflectivity measurements.

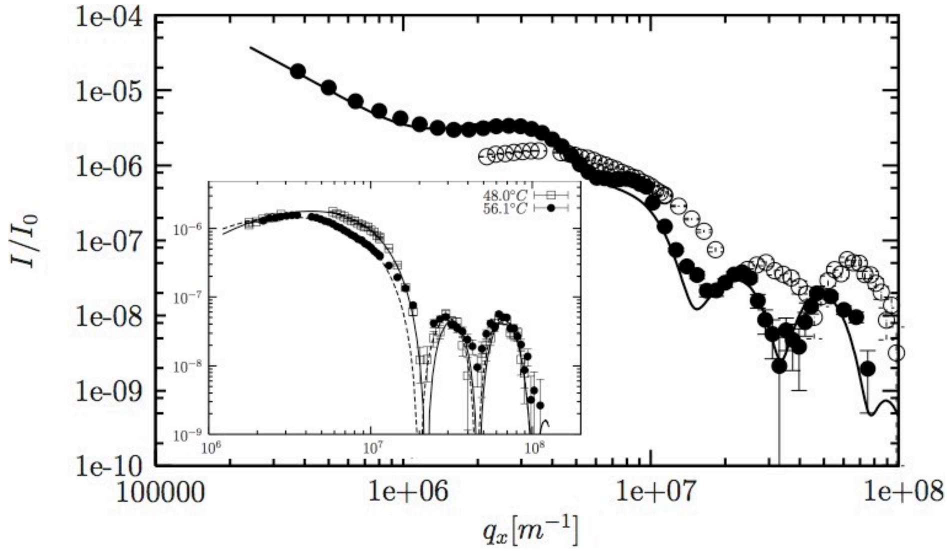

Figure 1: Grazing-incidence diffuse scattering curve in the fluid phase: open circle = previous experiment [2]; closed circle = this experiment. Note that we improved the resolution a low q values. The oscillation around 6.10^6 m^{-1} is caracteristic of correlations between the two layers. The solid line is a preliminary fit in the strong correlation limit. In insert, data and fits obtained in previous experiment in Gel and Fluid Phase ([2]).

The optimisation of experimental conditions give access q data at lower values of wave-vector, enabled us to distinguish particular features of diffuse scattering curves that could not be seen in the first experiments. Preliminary theoretical studies showed that this features are related to a crossed-term in the potential due to coupled fluctuations of the two membranes at large length-scales; The fluctuations appear to be uncoupled at short length-scales. We also investigated in details the transition region around melting temperature T_m, showing strong effect on the bilayer fluctuations. A detailed analysis remains to be performed to give quantitative variations of the physical parameters U", γ and κ at the transition.

The beamline BM32 was working well (although the 16-bunch lead to a doubling of the acquisition time, as expected) and the beamtime was used at 100%. Sample preparation was also satisfactory, according to both quantitative (transfer rates reproducibly > 95%) and qualitative criteria.

[1] G. Fragneto, T.Charitat, E. Bellet-Amalric, R. Cubitt and F. Graner, *Swelling of phospholipid floating bilayers: the effect of chain lengt,* Langmuir **19**, 7695-7702 (2003).

[2] J. Daillant, E. Bellet-Amalric, A. Braslau, T. Charitat, G. Fragneto, F. Graner, S. Mora, F. Rieutord, B. Stidder *Structure and fluctuations of a single floating lipid bilayer*, PNAS **102** 11639-11644 (2005).