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<tr><td>ESRF</td><td>Experiment title:
Polymorphism of the planar phospholipid membrane at the cesium-enriched silica hydrosol substrate.</td><td>Experiment number:
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<tr><td>Shifts:
18</td><td>Local contact(s):
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Report: During this round of experiments we studied spreading of various phospholipids at the surface of silica hydrosols substrates including those enriched by both Rb^+ and Cs^+. We used developed by us method for obtaining macroscopically planar phospholipid multilayers on top of a liquid substrate surface [1,2]. We have collected publishable sets of data on the ordering of DMPS (1,2-dimyristoyl-*sn*-glycero-3-phospho-L-serine), DPPC (1,2-dipalmitoyl-*sn*-glycero-3-phosphocholine) phospholipid molecules at the surface of the hydrosol substrates with 5-nm and 7-nm particles of SiO_2 at varies alkali ions (Na^+, Rb^+, Cs^+) concentrations.

According to our reflectivity data, at room temperature both DMPS and DPPC molecules form at the hydrosol's surface lamellar liquid-crystalline multilayer structures (stack of bilayers). The measurement of angular dependences of the reflection coefficient R of photons with energy ~ 70 keV (wave-length $\lambda \approx 0.17$ Å) for multilayers of DMPS (see curve 1 at Figure 1a) and DPPC at the surface of the hydrosol substrates were carried out in the range of the scattering vector $q_z = (4\pi/\lambda)\sin\mu$ (where μ is the angle of incidence) from 0 to 0.8 $Å^{-1}$ (8-10 orders of the magnitude of R). In addition, we obtained data for the dependences of R on both time and temperature in the interval from 25°C to 45°C.

The lateral structures of multilayers were probed by both off-specular scattering and grazing-incidence X-ray diffraction methods (see Figure 1c and 1d). We carried out measurements of diffuse scattering background for the above mentioned systems in a wide range of the detector angle, γ, that is from 0 to ~1 deg. For the scattering measurements we used the detector scans at the fixed grazing angle, μ, of the probe beam (at 0.012 deg or 0.02 deg). We observed sharp peaks for both phospholipid liquid-crystalline multilayers. Finally, according to the experimental data as temperature rises the phospholipid multilayers undergo phase transition from solid to liquid state. We applied grazing incidence diffraction method to study in-plane correlations in the multilayers to trace the phase transition in them from the hexagonal (liquid-crystalline) phase to a liquid phase that is related to the melting of hydrocarbon chains of phospholipid molecules.

Preliminary results:

1. For the preliminary analysis of the reflectivity data we have applied free-form approach (modelless) for the reconstruction of the dielectric function $\varepsilon(z)$ [3, 4]. Lines in Figure 1b demonstrate the electron density profiles for the DMPS multilayer obtained by this method. Solid line is for the liquid-crystalline state at room temperature and dashed line is for the liquid state at temperature above the transition $T_c \approx 35$°C. These results allow us: (a) to compare the near surface structure of membranes with existing biophysical models; (b) to obtain new quantitative information about lipid mesophases that is necessary for the better understanding of the physical and chemical properties of biological membranes.

2. Further joint quantitative analysis of high-resolution reflectometry and off-specular scattering measurements for DMPS and DPPC layers will be given both within the framework of the modelless and

model approaches. On their basis, the interlayer correlations in the lipid multilayers on the hydrosol substrate will be studied.

3. Applying the free-form approach to the reconstruction of the profiles of electronic density from temporal dependences of reflectivity, we plan to trace the process of ordering of the multilayer of both electroneutral DPPC and positively charged DMPS bilayers at the hydrosol's surface. The information obtained on kinetics and phase transitions in the lipid multilayers, which model natural biological membranes, is crucial for solving many biophysical problems.

In summary, X-ray scattering methods available at ID31 offer a fundamentally new experimental material that allows establishing the geometric characteristics and distributions of electron density at interface boundaries with lipid based structures of increasing complexity. For the description of three-dimensional surface structures from X-ray scattering data with high spatial resolution we develop on the basis of perturbation theories both a self-consistent free-form approach and a model approach in the distorted wave Born approximation (DWBA). In particular, the preliminary analysis of the data obtained by us demonstrated the unique sensitivity of the ID31 equipment for the study of the phase transitions in the phospholipid multilayers. The usage of high energy beam at ID31 appeared to be very useful for the reduction of the radiation damage to the samples. Overall, ID31 offers outstanding capabilities to carry out studies of such liquid surfaces.

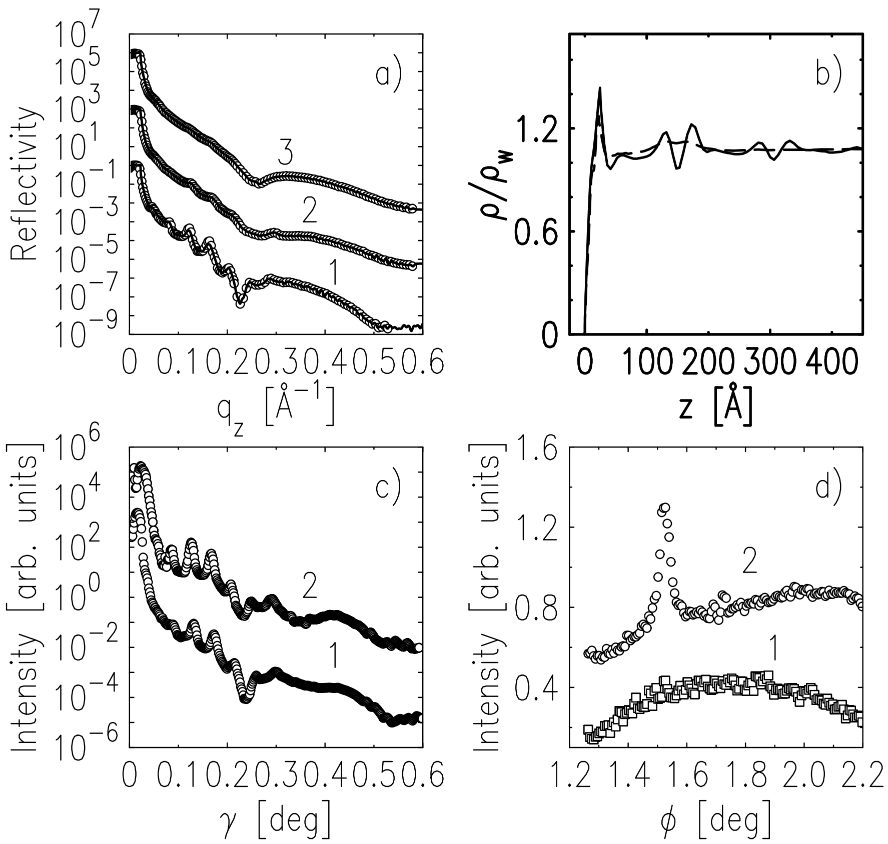

Figure 1. a) X-ray reflectivity as a function of q_z from the surface of silica hydrosol of 5-nm particles: 1,2 correspond to DMPS multilayer; 3 corresponds to DMPS bilayer. Lines correspond to modelless approach; b) Example of the reconstructed electron density profile of DMPS multilayer in the liquid-cristalline state (solid line) and in the liquid sate (dashed line) on top of the hydrosol; c) X-ray diffuse scattering from lipid multilayer as a function of the out of plane angle, γ, at different incident angles, μ: 0.012 deg (1), 0.02 deg (2); d) Intensity of grazing incidence diffraction for the phospholipid multilayer as a function of the in-plane angle, φ, at 40°C (1) liquid and 25°C (2) liquid-crystal.

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