ESRF	Experiment title: Characterization of the temperature dependent peptide incorporation in phospholipid multilayers using in-situ grazing incidence scattering and x-ray reflectivity.	Experiment number: SC-4659
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Report:

Introduction:

Phospholipid bilayers are a main component of membranes of biological cells. Solid supported multilayers of the phospholipid 1,2-dimyristoyl-sn-glycero-3-phosphocholine (DMPC) can be used as a model system for such membranes. Interactions of the membranes with peptides and polymers is essential for the function of the cells. Therefore, we have studied the structural change of DMPC caused by the biological relevant peptides Melittin, Alamethicin, and Amyloid beta.

Results and Discussion:

The beamtime was aimed at the characterization of structural changes of DMPC caused by mixing it with three different peptides (see above). Since these structural changes of the DMPC multilayer systems are expected to be concentration dependent, we measured a concentration series from 0.01 mol% to 2 mol% for all of the three peptides in addition to the pure DMPC sample. The samples were doctor bladed in situ onto silicon substrate at the ID10 beamline and simultaneous time resolved Grazing-Incidence Wide-Angle X-ray Scattering (GIWAXS) measurements were performed to study possible difference in film formation caused by the peptides. Afterwards, the films were hydrated using our custom made in situ sample cell (paper in preparation) and time resolved X-ray reflectivity (XRR) measurements were done. Since the analysis of the data is still in progress, we only show preliminary data in this report. Figure 1 shows an exemplary comparison of dry and hydrated pure DMPC, DMPC mixed with 1 mol% Melittin, and with 1 mol% of Alamethicin. In the dry states all three reflectivity curves show the typical Bragg peaks corresponding to the average distance of the phospholipid bilayers. After hydration the pristine DMPC sample consists of two phases: a fully hydrated and a partially hydrated phase. This can be

seen by the twofold splitting of every diffraction order. Whilst the XRR curves of the mixture of DMPC and Alamethicin shows a similar behavior as the pure DMPC in regards to the Bragg like scattering, a typical thin film oscillation not present in the measurements of the other samples can be observed. The mixture of DMPC and Melittin – as expected – doesn't show a thin film oscillation, since the overall thickness of the multilayer stack is too large. In this sample multiple Bragg peaks corresponding to a variety of periodic distance were apparent. A more detailed analyses of the scattering data of the concentration series during doctor blading and hydration is work in progress.

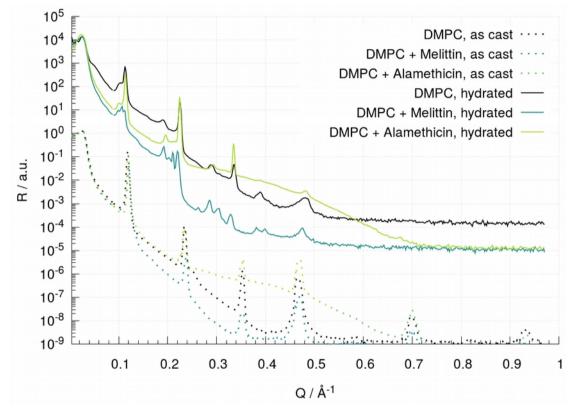


Figure 1: XRR of pure DMPC and of DMPC-peptide-mixtures measured at 30 °C. For both peptides (Melittin and Alameticin) a concentration of 1 mol% was used. The samples were measured before hydration (dashed curves) and at the end of the hydration process (solid curves, shifted upwards for clarity).

Experimental:

We have performed GIWAXS and XRR measurements using a photon energy of 22 keV. For the GIWAXS measurements we were using a Maxipix CdTe 2x2 area detector with a sample to detector distance of 957.13 mm, an angle of incidence of 0.065°, and a time resolution of 0.1 s. A Mythen MCA 2 detector was used for XRR. The cell was pumped with nitrogen during every measurement to reduce air scattering and radiation damage.