ESRF	**Experiment title: Bending rigidity studies of phospholipids membranes at a different stages of insertion of antimicrobial frog peptide ((PGLa)) into the layer.**	**Experiment number**: SC-1302
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Report:

Previously we reported [1,2] about effect of presence of active antimicrobial frog peptide PGLa from the skin secretion of the South African clawed frog, *Xenopus laevis* on the lateral and normal structure of the bacterial and eucaryotic cell membranes mimicked with phospholipidic monolayers of DPPG, DSPG and DPPC, DSPC. Glycerol phospholipids (-PG) DPPG, DSPG mimic bacterial cell membranes whereas choline phospholipids (-PC) mimic eucaryotic cell membranes. During these studies we found additionally to our primary aim that depending on the surface pressure and the area per a lipid molecule the antimicrobial peptide molecules can reversibly insert into or be extruded out of the -PG monolayer. However in spite of the same area per lipid molecule required for extrusion peptides from the monolayer the surface pressure in the membrane is higher in case of presence of peptide in comparison with lipid monolayer only. This may indicate that bending rigidity is different for these two cases. Change of bending rigidity changes the spectrum of surface capillary waves and as result diffuse scattering intensity measured in the grazing incidence geometry [3].

In present work we performed systematic measurements of diffuse scattering intensity measured in the grazing incidence geometry for two types of phospholipid monolayers DSPC and DSPG formed at air/ buffer interface. Monolayers were spread on the surface of phosphate buffer in a Langmuir trough especially designed for X-ray measurements with possibility to control surface pressure, area per molecule and temperature. Both types of system was studied with and without the presence of antimicrobial frog peptide PGLa at surface pressure $\pi = 15$ mN/m and $\pi = 30$ mN/m. The lower surface pressure is below the critical pressure (~ 25 mN/m) of the peptides extrusion from the membrane whereas the higher surface pressure is above. All measurements were done at the wavelength λ=1.536 Å. Measurements of one curve took typically about 9 hours. On the next figure we present reduced summary of the results.

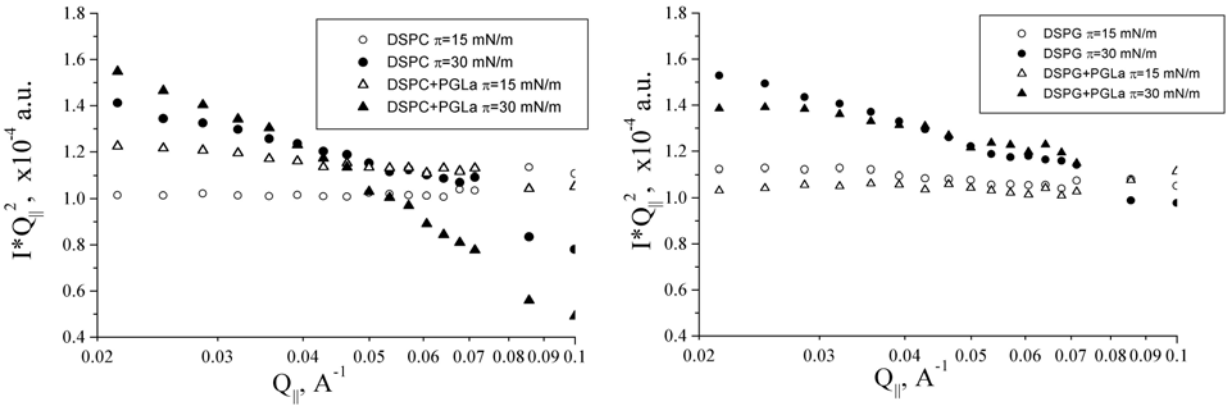

Figure. Grazing incidence diffuse scattering on monolayer of DSPC (left) and DSPG (right) with (triangle) and without (circle) presence of PGLa at surface pressure π=15 mN/m (open symbols) and π=30 mN/m (solid symbols).

The difference in the behavior of the two systems is clearly seen. Upon adding of peptides to DSPG monolayer at both surface pressures the measured curves shift downwards in respect to the curves measured without presence of the PGLa. This is clear indication of increase of bending rigidity of the monolayer. Increase of surface pressure in a monolayer change the surface tension and as the result increase diffuse scattering signal. The situation with DSPC monolayer is absolutely opposite to the DSPG monolayer. Upon adding of peptides the curves shift upwards in respect to the curves measured without presence of the PGLa. This indicates the reduction of the overall bending rigidity of the monolayer. From our previous studies we know that DSPC molecules do not interact with PGLa molecules and form two phase system on the air/water interface. The PGLa molecules are partially surface active and occupy part of available surface area. We expected that monolayer of peptides molecules reduce bending rigidity of air/water interface. Our control measurements (not shown here) demonstrated that diffuse scattering signal form air/water interface in presence of PGLa only became higher the curve of DSPC+PGLa at 30 mN/m. Due to the phase separation of DSPC and PGLa we measure averaged effect and observe increase diffuse scattering (decrease of bending rigidity) of the mixture. Quantitative analysis of the curves will be done later and published somewhere else.

1. Report on experiments SC-517 and SC-691
2. Konovalov O., Myagkov I., Struth B., Lohner K. European Biophysics Journal, 2002, v.31, pp.428-437
3. C. Fradin, A. Braslau, D. Luzet, D. Smilgies, M. Alba, N. Boudet,K. Mecke, J. Daillant, Nature, 2000, v. 403. pp 871-874