Influence and function of different ceramides on the nanostructure of *Stratum Corneum* model systems.

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Introduction:

The Stratum Corneum (SC) is the outermost layer of the skin. It is responsible for the skin barrier function and therefore the main problem when addressing drug penetration in topical delivery systems. The SC is mainly constituted by corneocytes (dead cells) surrounded by a lipid matrix, mainly constituted by ceramides (40% of the SC lipids), free fatty acids and cholesterol. The uniqueness of ceramides regarding other lipids is responsible for a special phase behavior of the SC different from the other biological membranes. Ceramides are composed of sphingosine and a fatty acid. The head groups are only slightly hydrophilic, and the two chains are long and have usually different length. Our aim was to enlarge the understanding of the role of the different constitutional ceramides (Ceramide AS, NS, AP and NP), mainly the role of the weakly polar head groups, by studying their phase behaviour and lateral structure in Langmuir monolayers using Grazing Incidence X-Ray Diffraction (GIXD) and X-Ray Reflectivity (XR).

Experimental method:

All systems studied were spread from chloroform:methanol (7:3) solution directly onto the water surface of the Langmuir trough placed in a hermetically sealed container, which is filled with helium. GIXD and XR measurements were performed along an isotherm at certain defined lateral pressures. While XR gives the averaged electron density profile normal to the interface, GIXD is the important method for structure determination.
The energy used was 10 keV and the measurements were performed at an angle of incidence of 0.105°, which is ~85% of the critical angle for total reflection. A Pilatus (2D-detector) was used for GIXD and a Mythen (1D-detector) for XR experiments.

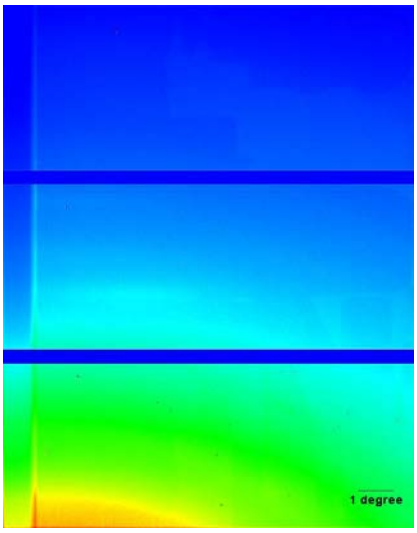
Results:

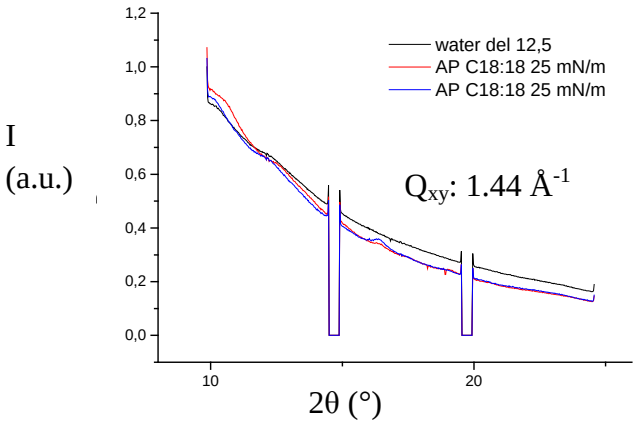
The GIXD setup used during our beamtime with a 2D-detector did not resolve all the peaks needed to determine the structure of our important molecules. We tested the setup with octadecanol, for which we could observe the Bragg peaks (even if the diffraction signal is rather weak) described in the literature for the respective surface pressure. However, in the case of our ceramides, only one Bragg peak was seen even after several trials to subtract the corresponding background. In a posterior beamtime (Proposal SC – 4016) a Mythen 1D-detector was used. We could prove that almost all ceramides studied exhibit a “pseudo-crystalline” monolayer state, being difficult to correctly index all the Bragg peaks with their respective Miller indices. Comparing to the data obtained in posterior beamtime (see Figure 1 below), where a 1D-detector was used, the GIXD data obtained cannot be used for evaluation.

Figure 1: GIXD results for Ceramide [AP] C18:18

a) @ 25mN/m on a water subphase obtained with a 2-D (Pilatus) detector.
b) Comparison of vertical cuts: bare water surface (background) and Ceramide [AP] C18:18 monolayers using different ranges.

(a) (b)





c) GIXD results for Ceramide [AP] C18:18 @ 20 mN/m on a water subphase obtained with a 1D (Mythen) detector