SANS and Neutron Reflection on the Mechanosensitive Channel Protein MscL

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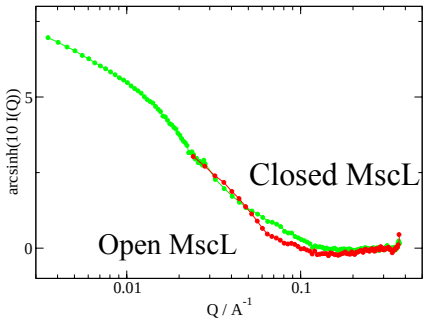
The mechanosensitive channel protein MscL is part of a sophisticated defence system, which bacterial cells have developed to tackle osmotic pressure. In the event of osmotic shock, this membrane protein can release pressure differences by opening a pore of several nanometres cross section in the cell membrane. The formation of the pore is accompanied by considerable conformational changes in the protein and alteration of its overall dimensions. We used neutron scattering and reflection techniques to characterize these structural changes in the mechanosensitive channel MscL.

Contrast matching and variation techniques render neutron scattering and reflection a powerful technique in the study of membrane systems. Using different levels of deuteration, the neutron scattering contrasts of MscL protein, the membrane environment and the buffer were adjusted and the different components of the system studied individually this way. To achieve a high scattering length density contrast, the protein was per-deuterated in the framework of the D-lab at ILL, Grenoble, and reconstituted in hydrogenous lipid bilayers. To characterize the protein in its closed and open state, measurements were performed before and after the addition of lysolipids which trap MscL in its open state.

In neutron reflection experiment both, supported bilayers of DMPC membranes without and with reconstituted MscL protein were established and their scattering length density profiles across the membrane could be obtained. The results show a bilayer with extended



e MscL containing membrane.

ANS experiments were conducted using MscL constituted unilamellar DOPC vesicles. The influence MscL on the lipid vesicles was studied as well as the otein itself, using suitable contrast matching conditions r the buffer. The addition of lysolipids to open the annel resulted in significant changes in the scattering haviour. Further work and data analysis is in progress to interpret these findings in terms of potential conformational changes.