Elasticity of crystalline $\beta$-sheet monolayers

H. Isenberg$^1$, V. Vaiser$^1$, K. Kjaer$^2$ and H. Rapaport$^1$

Department of Biotechnology Engineering, Ben-Gurion University of the Negev, Beer-Sheva, 84105 Israel; Niels Bohr Institute, University of Copenhagen, DK-2100 Copenhagen, Denmark.

Novel biomaterials composed of amphiphilic $\beta$-sheet molecular assemblies have been engineered in a bottom to top approach to form a variety of supramolecular architectures. Advanced understanding of peptide assemblies at the molecular level would enable utilization of peptides in applications that require nanometer-scale precision. Here we present an in-situ grazing incidence X-ray diffraction (GIXD) study showing that the ordered $\beta$-sheet assemblies of amphiphilic peptides Pro-Glu-(Phe-Glu)$_5$-Pro exhibit elasticity, apparent from a quasi-reversible compression and expansion of the 2-D crystalline unit cells at the air-water interface (Figure 1c). The alternating hydrophilic (Glu) and hydrophobic (Phe) amino acids along the peptide backbone induce the $\beta$-strand conformation at air-water interfaces (Figure 1a) [1]. Phe was selected as the hydrophobic amino acid for its relatively large side-chain that is advantageous in X-ray diffraction measurements. Phe side chains may also form favorable phenyl-phenyl interactions between neighboring strands. The stability of these $\beta$-sheet monolayers at the air-water interface has also been attributed to a possible hydrogen bonded arrays formed by Glu side chains.

The surface pressure vs. mean molecular area isotherm in Figure 1c shows the film compression and expansion. The compression isotherm starts at a low surface pressure region that extends down to the peptide limiting area per molecule. At this point along the isotherm, the water interface is essentially fully covered by the peptide molecules. Further compression leads to a steep increase in surface pressure which is followed by a collapse at surface pressure $\sim 25$ mN/m. The collapse state is commonly attributed to the formation of ordered or disordered multilayer structures. In general the expanding film exhibits mean molecular areas that are smaller than during compression, suggesting the irreversible formation of molecular aggregates along the collapse region of the isotherm.

GIXD measurements indicate that on compression the repeat distance, that corresponds to the long axes of the peptide strands (0,1), may decrease by up to 37% (Figure 1b). Upon expansion the compressed $\beta$-sheet assemblies revert elastically to their original conformation. The interstrand repeat distance along the peptide hydrogen bonds apparently does not change upon film compression and expansion. Based on the GIXD data, at surface pressures higher than $\sim 3$mN/m, beyond the peptide limiting area per molecule, the compressibility of the crystalline $\beta$-sheet, $C_c = -(\partial \ln d_{0,1}/\partial \pi)$, where $d_{0,1}$ is the crystalline $d$-spacing, is $\sim 8$ m/N. The out of plane Bragg rod diffraction patterns imply that in the compressed state the $\beta$-strands buckle up in reaction to the increase in surface pressure (Figure 1d). At low surface pressure the 2-D compressibility of the crystalline $\beta$-sheet was estimated at $\sim 32$ m/N and attributed to inter-domain rearrangements.

Whether on compression or expansion of the film, the (0,1) Bragg peaks occur at similar $q_{xy}$ values, implying that under surface pressure the crystalline $\beta$-sheet monolayer film deforms elastically and approximately reversibly. Nevertheless, Figure 1b depicts a continuous decrease in the intensity of the (0,1) Bragg peak all along the compression and the expansion isotherm, indicating a destruction of the ordered peptide domains, which occurs probably due to shear stresses, interfacial forces resulting from film compression and expansion and beam damage.
This work was supported by the European Community - Research Infrastructure Action under the FP6 "Structuring the European Research Area" Programme (through the Integrated Infrastructure Initiative "Integrating Activity on Synchrotron and Free Electron Laser Science"). Contract RII3-CT-2004-506008. We also acknowledge support from the Carlsberg Foundation and from the DANSYNC programme of the Danish Natural Science Research Council.

References