Experimental report

02/12/2015

Proposal: 9-11-1697 **Council:** 4/2014

Title: Structure of DNA-bottlebrushes

Research area: Soft condensed matter

This proposal is a new proposal

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Samples: DNA-protein polymer bottlebrushes

Instrument	Requested days	Allocated days	From	To
D11	4	3	05/12/2014	08/12/2014

Abstract:

We have developed a unique well-defined model system for so-called bottle-brush polymers that consists of DNA decorated with physically attached sidechains. The sidechains are genetically engineered hydrophilic polypeptides with short positively charged binding domain that are perfectly monodisperse. For this system we propose to do a precise SANS characterization of both the brush thickness and profile (that, in view of the monodispersity of the system, makes for a good comparison with current polymer brush theories) and of the effective persistence length of the brush-coated DNA. Many conflicting theories exist for the effective persistence lengths of bottlebrushes and our model system in combination with detailed SANS studies offers an ideal opportunity to connect theories and experiments for this highly disputed problem.

1 Structure of DNA-bottlebrushes

The aim of this research proposal was to investigate the brush profile and effective stiffness of our bottlebrushes consisting of DNA as the backbone molecule and a protein polymer, $C_{400}K_{12}$, as side chains. The idea with SANS was to select the contrast to measure the DNA core alone or brush alone (or both). Calculating the scattering length density resulted in an estimated ratio of 35% D_2O/ 65%H_2O to measure only the DNA and 57% D_2O/ 43%H_2O for the protein-only contrast. A test experiment, however, already showed us beforehand that these ratios are not correct for the actual samples. The ratios to match out DNA or the protein are in fact too close to each other to use.

A back-up plan was to investigate the liquid crystalline structure of our DNA-$C_{400}K_{12}$ bottlebrushes by changing the DNA concentration and the DNA to $C_{400}K_{12}$ ratio. The data for two DNA concentration series are shown in figure 1 for stoichiometric amounts of $C_{400}K_{12}$ (a) and three times excess amounts of $C_{400}K_{12}$ (b). The denotation of $\Gamma = 10$ means that $10\times$ more mass, with respect to DNA, of the $C_{400}K_{12}$ protein polymer was required. $\Gamma = 30$ means $30\times$ more $C_{400}K_{12}$ was used. From these scattering data we extracted the distances, d, and plotted it with respect to the DNA concentration in figure 2. The information obtained from these SANS experiments is useful for other (osmotic pressure) studies on the same system. The data shows that increasing the DNA concentration obviously results in better aligned liquid crystalline samples but only for stoichiometric amounts. The samples with excess amounts of protein show a slightly disrupted liquid crystalline structure. This is also confirmed in figure 3. Figure 3(a)-(c) show nicely aligned anisotropic behaviour but figure 3(d), which is one of the samples with excess amounts of $C_{400}K_{12}$, shows a more diffuse scattering pattern.

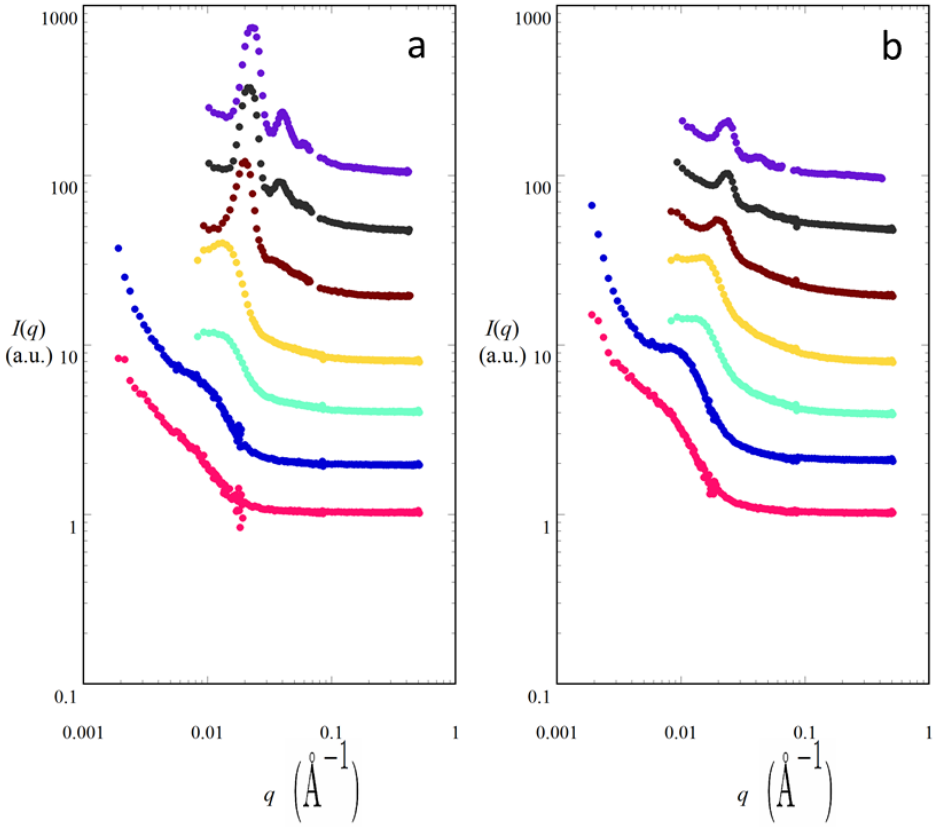

Figure 1: SANS measurements on the concentration series of DNA-$C_{400}K_{12}$ complexes for $\Gamma = 10$ (a) and $\Gamma = 30$ (b). DNA concentration from top-to-bottom: 20 mg/mL (yellow), 15 mg/mL (L blue), 10 mg/mL (purple), 5 mg/mL (orange), 2 mg/mL (green), 1 mg/ml (D blue), 0.5 mg/mL (black).

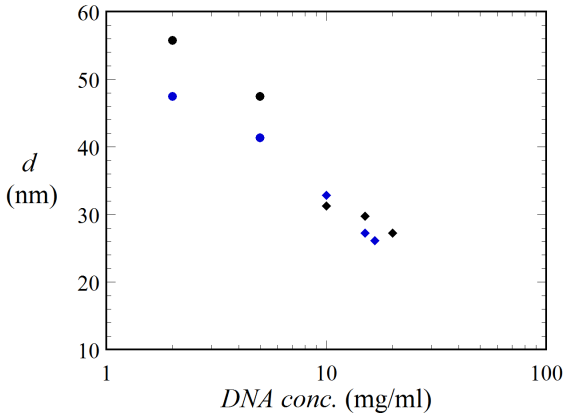

Figure 2: Distances as a function of DNA conc. according to SANS measurements. Black data points represent protein to DNA ratio of $\Gamma = 10$ (charge) and blue data points represent a charge ratio of $\Gamma = 30$. The spherical symbols correspond to isotropic samples and the diamond shaped symbols to anisotropic samples.

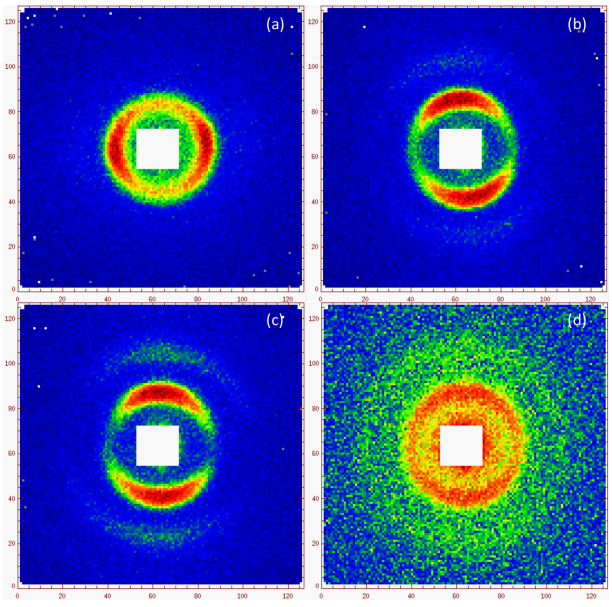

Figure 3: 2D-scattering profile of the SANS measurements of the following compositions: 10 mg/ml DNA + $C_{400}K_{12}$ ($\Gamma = 10$) (a), 15 mg/ml DNA + $C_{400}K_{12}$ ($\Gamma = 10$) (b), 20 mg/ml DNA + $C_{400}K_{12}$ ($\Gamma = 10$) (c), 15 mg/ml DNA + $C_{400}K_{12}$ ($\Gamma = 30$) (d).