	Experiment title: probing in situ formation of interpolyelectrolyte complexes between oppositely charged peptide and nucleotide sequences	**Experiment number**: **CH-4293**
Beamline: BM26B	**Date of experiment**: from: 6.10.2014 to: 8.10.2014	**Date of report**:
Shifts: 6	**Local contact(s)**: Giuseppe Portale	*Received at ESRF:*

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Report:

Synchrotron SAXS experiments were performed on the BM26B beam line (ESRF, Grenoble, France) using a pixel detector (1M PILATUS). The X-ray scattering images were recorded using a monochromatic incident X-ray beam ($\lambda = 0.154$ nm) covering the $0.023\ nm^{-1} < q < 1.65\ nm^{-1}$ range of momentum transfer ($q = 4\pi/\lambda \sin\theta$), with 2θ the scattering angle). During experiments some technical problems were witnessed. The detector that measures transmission was apparently broken. As a result, software that normalizes data on transmission was malfunctioning and proper subtraction of solvent background was very complicated.

The main objective of the proposed research was to shed light on the structure of complex formation of amyloid β (1-42) – a protein that is involeved in the onset of Alzheimer's disease. The primary aim is the investigation of structure of highly diluted interpolyelectrolyte complexes (IPECs) upon incubation of the amyloid β (1-42) peptide sequence (Aβ42) with different polyions, namely positively charged poly(ethylene imine) (PEI) and negatively charged poly(acrylic acid) (PAA) or short single stranded nucleotide sequences. These objectives will be achieved by the characterization of the properties of the complexes in terms of aggregation rate, size and morphology.

The amyloid sequence is composed of three positively charged amino acid lysine and arginine residues and three partially charged histidine residues along with 6 negatively charged residues. Since no specificity of the nucleotide sequence composition towards interaction with the peptide sequence has been observed, we therefore investigated the structure formation upon incubation with sodium chloride (NaCl), positively charged poly(ethylene imine) (PEI), negatively charged poly(acrylic acid) (PAA) and with 3-mer (5'-GAG-3'), 6-mer (5'-TCTGAG-3') and 12-mer (5'-AAAGAGAGAGAG-3') nucleotide long sequences which are negatively charged owing to the presence of the phosphate groups along the backbone in these physiological-like conditions. Conjugation of poly[2-(3-butenyl)-2-oxazoline] (PBOX) with the 12 nucleotide-long nucleotide sequence (5'-CTCTCTCTCTTT-3', 5'-$(CT)_5T_2$-3'), was studied as well.

SAXS experiments conducted on pure nucleotide long sequences revealed the presence of small objects of coil-like conformation. Such objects were attributed to unimers of nucleotides (Figure 1a).

The scattering curves of PBOX-g-DNA copolymers could be analyzed with the form factor of a hollow cylinder of finite length (8600 nm) in combination with that of a Gaussian coil. The best possible fit was achieved with structures of 17 nm inner radius and outer radius of 76 nm (Figure 1b).

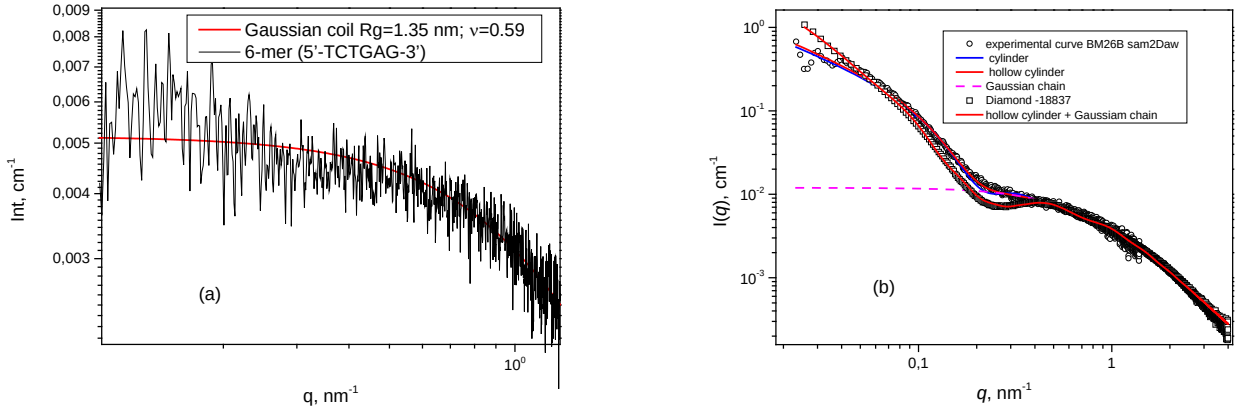

Figure 1. SAXS data from (a) unimers of 6-mer (5'-TCTGAG-3') and (b) PBOX-g-DNA copolymers.

Additional SAXS experiments conducted at Diamond Light Source supported those conclusions (Figure 1b). The presence of hollow fibers was independently proven by TEM method. (Figure 2)

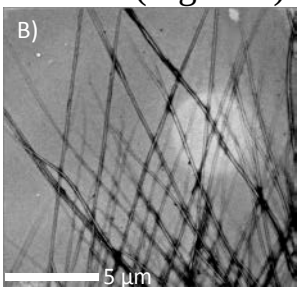

Figure 2. Fibers morphology. TEM imaging.