MLF Experimental Report	提出日 Date of Report 2017 Feb 27
課題番号 Project No. 2017B0138 実験課題名 Title of experiment Quantitative evaluation of uniformity of various polymer networks by small angle neutron scattering (SANS) with contrast variation technique 実験責任者名 Name of principal investigator Xiang Li 所属 Affiliation The University of Tokyo	装置責任者 Name of responsible person 高田　慎一 装置名 Name of Instrument/（BL No.） BL-15 Small and Wide Angle Neutron Scattering Instrument (TAIKAN) /BL-15 実施日 Date of Experiment 20/1/2018-23/1/2018

試料、実験方法、利用の結果得られた主なデータ、考察、結論等を、記述して下さい。（適宜、図表添付のこと）
Please report your samples, experimental method and results, discussion and conclusions. Please add figures and tables for better explanation.

1. 試料 Name of sample(s) and chemical formula, or compositions including physical form.

PEG/DNA conjugate in D_2O/H_2O buffer

1. Tetra-functional poly(ethylene glycol)s (M_w = 40,000, conc. = 1 mM)
2. 16mer sense- and anti-sense DNA strands
3. Buffer {D_2O 63.2%, H_2O 36.8 %, sodium phosphate 25 mM, ethylenediaminetetraacetic acid 1 mM, pH 7.4}

2. 実験方法及び結果（実験がうまくいかなかった場合、その理由を記述してください。）
Experimental method and results. If you failed to conduct experiment as planned, please describe reasons.

By reacting Tetra-PEG with DNA, DNA-terminated Tetra-PEG (Tetra-PEG/DNA) were synthesized (Figure 1). There are two types of Tetra-PEG/DNA. One is sense-DNA/PEG conjugate and another is anti-sense-DNA/PEG conjugate. By mixing these two types tetraPEG/DNA conjugate at room temp., the gel was formed in the 1mm thick demountable cell due to the physical bond between sense and anti-sense DNA strands.

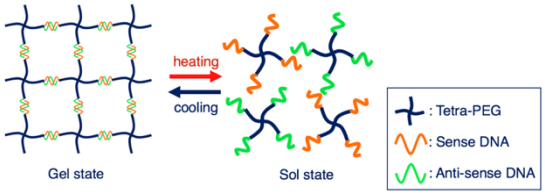

Figure 1. Schematic of PEG/DNA conjugate and its gel

Because strength of physical bond between DNA depends on temperature, the sol-gel transition occurrs by changing the temperature. The sol-gel transition point is about 55 ℃ by rheological measurement in lab.

2. 実験方法及び結果（つづき） Experimental method and results (continued)

We have measured this Tetra-PEG/DNA gel by using small-angle neutron scattering (SANS) at different temperatures from 5-75ºC for cycles to check the structure due to sol-gel transition and the hysteresis.

Every sample was well sealed and place in the 10 positions auto sample changer with temperature control. The run time for each sample was 1.5 hour and it took about 0.5 hour for the temperature to stabilize at each temperature. At the temperature lower than 15ºC, we sealed the sample changer with dried nitrogen gas to prevent condensation on the sample window. Figure 2 shows the SANS profiles of tetraPEG-sene-DNA/tetraPEG-anti-sense-DNA mixture and Figure 3 shows the profiles of tetraPEG-sense-DNA. In Figure 2, the scattering peaks were observed at $q \sim 0.03$Å^{-1}, which is because of the electrostatic interaction between DNAs as expected. There was no hysteresis after cycles of heating and cooling, indicating the very site specific and reproducible bonds between DNAs unlike the other physical gels like gelatin or agarose. In Figure 3, the scattering peak at $q \sim 0.03$Å^{-1} was also observed and there was no hysteresis. Surprisingly, the SANS profiles of the tetraPEG-sene-DNA/tetraPEG-anti-sense-DNA mixture and tetraPEG-sene-DNA only solution was exactly the same. This might indicate the homogeneity of this hydrogel, but we still need more low-q data to confirm this. The additional experiment is scheduled in the Bio-SANS at NIST at the second half of this year. Now, we are trying to get more quantitative analysis with an empirical equation often used for polyelectrolyte solutions:

$$I(q) = \frac{B}{1 + (|q - q^*|L)^m} + C$$

where q^* is the peak top position, L is the peak width, m relates to the decreasing slope at high-q side, B and C are system dependent constants. Although the physical meaning of these parameters is still in discussion, it is clear that q^* remains constant both in sol and gel state. The correlation length of the peak in real space is around 20 nm, which is much larger than the size of macromonomers (Tetra-PEG/DNA) (R_g ~5 nm from calculation).

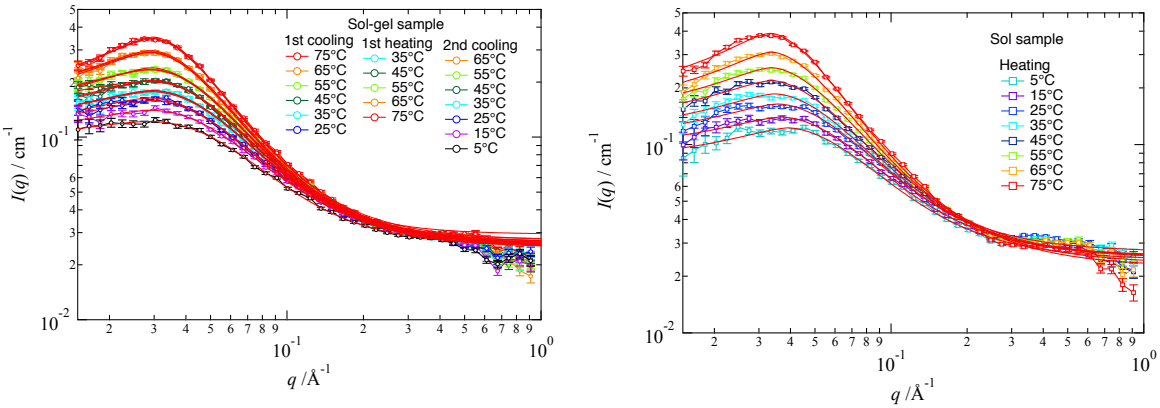

Figure 2. SANS profiles of tetraPEG-sene-DNA/tetraPEG-anti-sense-DNA mixture and tetraPEG-sene-DNA only solution with repetitive heating and cooling cycles. All the scattering profiles at the same temperature exactly overlap each other, indicating no hysteresis in our sample.