
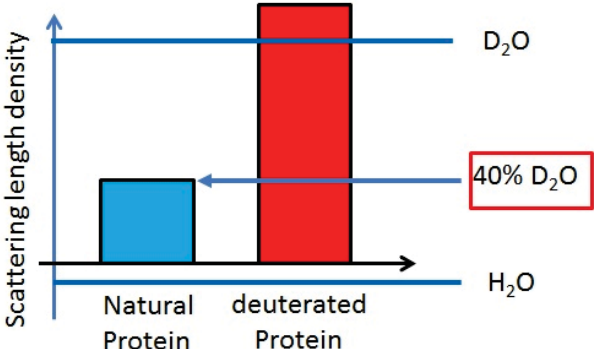


(※本報告書は英語で記述してください。ただし、産業利用課題として採択されている方は日本語で記述していただいても結構です。)

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|  | 承認日 Date of Approval 2015/1/4 承認者 Approver Jun-ichi Suzuki 提出日 Date of Report 2014/5/13 |
| 課題番号 Project No. 2013B0069 実験課題名 Title of experiment Study on Interaction between Transcription factor domain and Genral Transcription Factor by Small-Angle Neutron Scattering 実験責任者名 Name of principal investigator Masaaki Sugiyama 所属 Affiliation Kyoto University | 装置責任者 Name of Instrument scientist Shin-ichi Takata 装置名 Name of Instrument/(BL No.) BL-15 実施日 Date of Experiment 2014/04/04-2014/04/07 |

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.

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| 1. 試料 Name of sample(s) and chemical formula, or compositions including physical form. |
| We observed two protein systems concerning about RNA polymerase. 1. hydrated-Qb (protein) in 100% D ₂ O 2. deuterated-Qb + hydrated TAF complex in 40% D ₂ O. |

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| 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. | |
| Sample protein, Qb, is an intrinsically disordered protein (IDP) and is considered to make a complex with TAF. However, there is no experimental result concerning about the structural modulation of Qb on the formation of the complex with TAF. The neutron scattering length density of a natural (=hydrogenated) protein is matched to that of the 40% D ₂ O solvent but that of deuterated protein is larger (Figure 1). Therefore, we can observe only a deuterated protein of the complex in 40% D ₂ O solution with small-angle neutron scattering. |  <p>Figure 1. Contrast map of natural (hydrogenated), deuterated proteins and solvents with the mixture of H₂O/D₂O.</p> |

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

Figure 2 shows a scattering profile of hydrogenated Qb in 100% D₂O and its Guinier plot. As mentioned before, Qb is an IDP and likely makes aggregation. Therefore, it is very difficult to observe the scattering profile in the monodispersed state. In this experiment, the scattering profile from well-monodispersed samples was observed (Figure 2(a)). From the Guinier plot (Figure 2(b)), the gyration radius of Qb was found to be $21.3 \pm 0.9 \text{ \AA}$. This value is relatively smaller than 37 \AA , which was observed with small-angle x-ray scattering in our previous study. One possible reason to explain this discrepancy is that Qb slightly made aggregation. However, it was also reported that the gyration radius of non-folding protein, such as IDP, is given by the following empirical equation [1],

$$R_g = R_0 N^\nu,$$

Where R_g , R_0 , N , ν are the observed gyration radius, the constant (1.330), chemically denatured residues length, and exponent constant (0.598). Considering the length of Qb, the SANS result indicates that Qb is more globule protein in our experimental condition. Now, we are examining the secondary structure of Qb with circular dichroic spectroscopy and also analyzing the structural modulation of Qb in the complex.

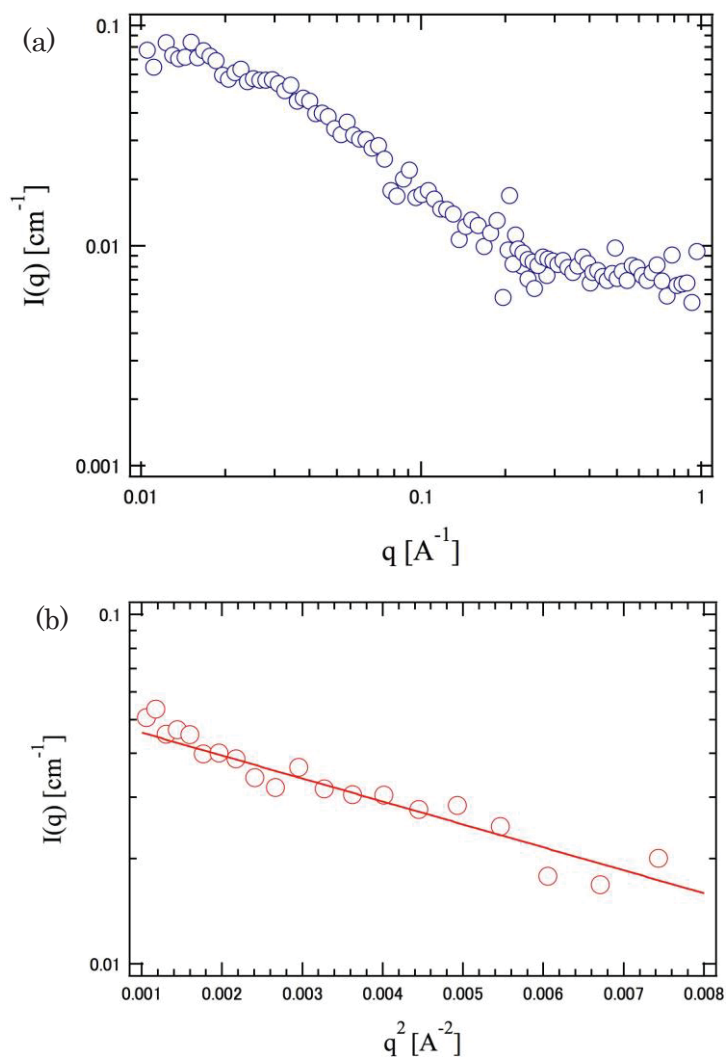


Figure 2. (a) SANS profile of hydrogenated Qb in 100% D₂O and (b) Its Guinier plot.