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 MLF Experimental Report	提出日 Date of Report
課題番号 Project No. 2017A0078 実験課題名 Title of experiment SANS study for hemoglobin confined inside silica nanocavity 実験責任者名 Name of principal investigator Akira Yamaguchi 所属 Affiliation Ibaraki University	装置責任者 Name of responsible person Hiroki Iwase 装置名 Name of Instrument/(BL No.) Taikan/BL15 実施日 Date of Experiment 1-3/June/2017

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.
Mesoporous silica (SiO ₂) powder dispersed in D ₂ O/H ₂ O mixtures. Myoglobin and hemoglobin in D ₂ O/H ₂ O mixtures. Mesoporous silica containing myoglobin dispersed in D ₂ O/H ₂ O mixtures. Mesoporous silica containing hemoglobin dispersed in D ₂ O/H ₂ O mixtures.

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.
<p>SANS experiments for above samples were performed by TAIKAN. The mesoporous silica is one of inorganic nanoporous material with uniform pore structure. The main purpose of our research is observation of hemoglobin (Hb) molecules confined inside the pore of mesoporous silica by contrast-variation SANS experiments. For that purpose, we performed SANS experiments for mesoporous silica with myoglobin (Mb) to determine the experimental conditions. In addition, the SANS experiments were applied to observe tertiary structure of Hb within mesoporous silica.</p> <p>Result 1: SANS experiments for Mb within mesoporous silica (D₂O)</p> <p>Herein, we prepared conjugates of mesoporous silica (MPS) and Mb (Mb/MPS conjugate). The Mb/MPS conjugates with different amount of Mb adsorbed were dispersed in D₂O, and the slurry samples were applied for SANS experiments. The present SANS profiles were discussed by taking into account contrast-variation SANS results (2016B0051).</p>

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

Herein, area of Bragg diffraction peaks due to hexagonal pore arrangement of MPS were estimated from the SANS profiles, and their dependency on Mb amount within MPS was analyzed. The analyzed data were compared to peak intensities simulated by core-shell cylinder model. The simulation predicts that the peak intensities monotonically decrease with increasing the Mb amount within MPS. This tendency is somewhat different with the experimental data, suggesting inadequacy of the core-shell cylinder model or specific molecular assembly of Mb within MPS.

Result 2: SANS experiments for Mb within mesoporous silica (62.1% D₂O)

Herein, we prepared conjugates of mesoporous silica (MPS) and Mb (Mb/MPS conjugate). The conjugates were dispersed in the contrast-matching solvent (62.1% D₂O), and the slurry samples were applied for SANS experiments. As the results, we obtained scattering data for Mb without interference of MPS. The scattering data could be well fitted to hard-sphere model, and the radius of Mb within MPS obtained by the fitting analysis was well agreed with that of free Mb in bulk D₂O. This result indicates that the tertiary structure of Mb is almost same with that of free Mb with spherical structure.

Result 3: SANS experiments for Hb within mesoporous silica (62.1% D₂O)

Herein, we prepared conjugates of mesoporous silica (MPS) and Hb (Hb/MPS conjugate). The conjugates were dispersed in the contrast-matching solvent (62.1% D₂O), and the slurry samples were applied for SANS experiments. As the results, we could not obtain clear scattering signal due to Hb within MPS. This is due to low amount of Hb adsorbed at MPS. In SANS study of Hb in D₂O, it was revealed that Hb molecules were aggregated in concentrated Hb solution. This aggregation of Hb prevents the adsorption of Hb within the MPS pores (pore diameter = 75 Å). Suppression of the Hb aggregation in water should be required to introduce Hb within MPS.