MLF Experimental Report	提出日 Date of Report
課題番号 Project No. 2017B0130 実験課題名 Title of experiment Investigation of specific protein aggregation depended on meso-cavity sizes 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 11-15/Dec/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_2O/H_2O mixtures.

Myoglobin in D_2O/H_2O mixtures.

Mesoporous silica containing myoglobin dispersed in D_2O/H_2O 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.

SANS experiments for above samples were performed by TAIKAN. The mesoporous silica is one of inoragnic nanoporous material with uniform pore structure. The main purpose of our research is observation of myoglobin (Mb) molecules confined inside the pore of mesoporous silica by contrast-variation SANS experiments. Until now, we have performed SANS experiments for Mb within mesoporous silica. The main purpose of this project is to obtain reliable SANS profiles to analyze the Mb tertiary structure depended on the pore size. In addition, the possibility of the specific molecular aggregation of Mb was examined.

Result 1: SANS experiments for Mb within mesoporous silica (62.1% D_2O)

Herein, we prepared mesoporous silica powders with different pore sizes. The conjugates of mesoporous silica (MPS) and Mb (Mb/MPS conjugate) were dispersed in the contrast-matching solvent (62.1% D_2O), and the slurry samples were applied for SANS experiments.

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

In the present study, we prepared a series of Mb/MPS conjugates with different MPS pore sizes. The measurement time for each slurry samples was 8 to 10 h to obtain reliable scattering signal due to the Mb within MPS.

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 D_2O. This result indicates that the tertiary structure of Mb is almost same with that of free Mb with spherical structure.

On the other hand, SANS profile of Mb within specific MPS sample was not well fitted to the hard-sphere model. From other spectroscopic studies, complete unfolding of Mb within the specific MPS was ruled out. We hence analyzed the SANS profile by other models (ellipsoidal and core-shell models). As the results, it was concluded that partial unfolding of Mb took place within the specific MPS pores. In addition, it was also concluded that the pore size did not affect the Mb tertiary structure.

Article containing above results is now preparing.

Result 2: Specific Mb assembly within mesoporous silica

Herein, we also found specific Mb assembly within MPS with specific pore sizes. This specific Mb assembly could be explained by the results in previous SANS study.