Analysis of small heat shock protein and insulin B chain complex by small-angle neutron scattering

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Amyloid fibers, which are deeply involved in many serious diseases such as Alzheimer's disease and Huntington's disease, are formed by abnormal protein aggregation. Elucidating the mechanism of abnormal aggregation, which triggers the onset of the aforementioned diseases, is essential for future drug development. We have been aiming to elucidate the mechanism of amyloid fiber formation by applying techniques such as small-angle X-ray scattering (SAXS), using insulin B-chain as a model protein. SAXS measurements of insulin B-chain fibers with and without αB-crystallin were performed. Both samples showed an $I(Q)\sim Q^{-1}$ relationship, suggesting a rod-like shape at low Q. Cross-sectional Guinier analysis calculated the radii of the two samples to be 22.0±1.0 Å and 31.0±3.0 Å, respectively, for insulin B-chain fibers and insulin B-chain fibers in the presence of αB-crystallin, respectively. To reveal the spatial distributin of insulin B-chain and αB-crystallin in the complex, we performed small-angle neutron scattering measurements on protein complex comprised of hydrogenated insulin B-chain and partially deuterated αB-crystallin. Prior to detaield strcutural analyzes of complex, we checked the matching point of partially deuterated αB-crystallin by changing the D_2O ratio of solvent. Figure 1 shows the $(I(0)/c)^{0.5}$ as a function of D_2O ratio of solvent. It was calculated that matching point of partially deuterated αB-crystallin was 100% D_2O. Figure 2 shows the SANS profiles from the complex comprised of hydrogenated insulin B-chain and hydrogenated αB-crystallin (pink circle) and the complex comprised of hydrogenated insulin B-chain and partially deuterated αB-crystallin (light blue circle), respectvely. Both SANS profiles showed an $I(Q)\sim Q^{-1}$ relationship, however the slope of complex comprised of hydrogenated insulin B-chain and partially deuterated αB-crystallin seems to be samller than that of complex comprised of hydrogenated insulin B-chain and hydrogenated αB-crystallin. This finding qualittatvely means that αB-crystallin existed at outer region of complex. We are perfroming the detailed analyzes of complex.

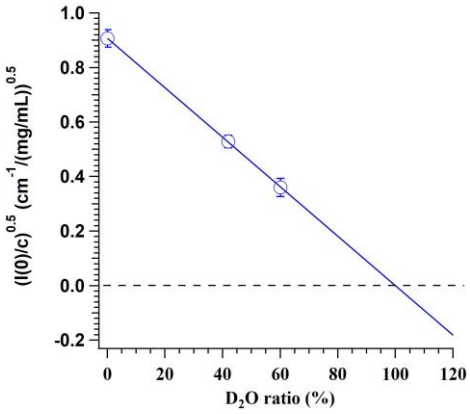

Fig. 1. $(I(0)/c)^{0.5}$ of partially deuterated αB-crystallin as a function of D_2O ratio of solvent.

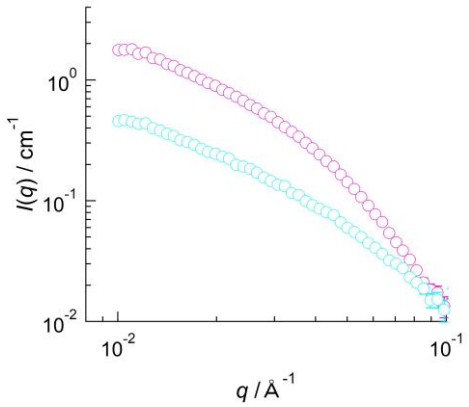

Fig. 2. SANS profiles from complex comprised of hydrogenated insulin B-chain and hydrogenated αB-crystallin (pink circle) and the complex comprised of hydrogenated insulin B-chain and partially deuterated αB-crystallin (light blue circle), respectvely.