Magnetic Field Effects on Crystallization of Thaumatin by LLIP Method Ⅱ

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Many crystallization techniques have been studied to produce a huge protein crystal with high quality for XRD analysis in the field of drug development. Liquid-liquid interfacial precipitation (LLIP) method is one of new crystallization techniques and it is available to control the crystal size and polymorph[1]. When two kinds of solvent are stacked to make their liquid-liquid interface, the 2-dimensional interface gets supersaturated due to a mutual diffusion. Then a crystal is formed and precipitated at around the interface. A large number of studies using LLIP method have been reported to make a protein crystal in space. Meanwhile a vertical high-gradient magnetic field is able to provide a reduced or hyper-gravity environment. In this paper, thaumatin protein was crystalized by LLIP method under the influence of vertical magnetic field with gradient, and its magnetic field effects on size of crystal was studied.

Thaumatin was crystallized and grown at various positions under the vertical magnetic fields of up to 10 T for 20 days at 20℃. The bipyramidal shaped crystals were observed on the bottom of well plate rector. The angle between the direction of *c*-axis and the magnetic flux was measured. The *c*-axis was aligned parallel to the flux under the influence of vertical magnetic fields above 5.35 T. On the other hand, the lengths of *a* and *c*-axes were also measured. The ratio between the two axes was mostly constant for crystals under all position in the magnetic field and even in no magnetic fields. The growth behavior was observed in no magnetic fields *in situ*. The ratio was independent on time.

Fig. 1 showed the distribution of the length of *a*-axis on behalf of the crystal size. The distribution for the crystals observed in the center position(b) shifted a little small size compared with that in no magnetic fields. The timing to start to sedimentation was thought to be advanced due to the effect of alignment. When the thaumatin was crystalized in lower position of the magnet bore, where corresponded to hyper-gravity environment, the distribution got smaller compared with (b). Stokes speed was accelerated due to downward magnetic force and the growth duration got shorter.

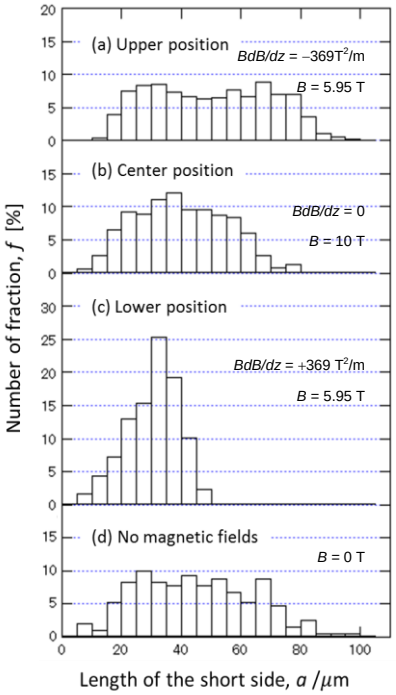

Fig. 1. Size distribution of thaumatin crystal grown at upper(B = 5.35 T, *BdB/dz* = −369 T^2/m), center (10 T), lower positions of the bore of the vertical magnet, and no magnetic fields by using LLIP method.

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Reference: [1] M. Tatara *et al*, Abs. 78th JPSJ Autumn Meeting (2017) p. 09-014.