

Crystal Structure Analyses of Human High-affinity Receptor for IgE

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The high-affinity receptor for IgE (FcεRI) exclusively expressed on mast cells and basophils is the key molecule in triggering the IgE-mediated allergic reaction. Multivalent antigens bridge the receptor-bound IgE and induce the aggregation of FcεRI on the cell surface, and in turn trigger the release of mediators responsible for allergic symptoms. To prevent the IgE-mediated allergic reaction specifically, one of the best strategies is the usage of the antagonist to FcεRI molecule. We started the crystallographic study of soluble domain of FcεRI for the efficient design of the antagonist.

The soluble domain of the FcεRI α subunit was expressed in Chinese hamster ovary (CHO) cells and was shown to bind IgE with an affinity similar to that of the cell surface FcεRI. We use this soluble domain of the FcεRI α subunit (soluble α) for crystallization. The crystallization was done by the hanging-drop vapour diffusion method using sodium acetate as a precipitant. Prism crystals were grown to size in the range of 20 to 100 μm in length and 5 to 20 μm in diameter. They belong to the tetragonal crystal system with

unit cell dimensions of $a = b = 81.3 \text{ \AA}$ and $c = 115.3 \text{ \AA}$. The crystals are unstable towards X-ray exposure and diffract up to only 6 Å resolution using a laboratory-use X-ray generator as a source. To bring out the diffraction power of the crystals to the limit, the usage of the X-ray from synchrotron radiation is required.

The diffraction study at SPring-8 was 8 hour long and it was long enough for the preliminary check of the diffraction limit and the X-ray damage of the crystal towards synchrotron radiation. In the case of the crystals normally packed into grass capillaries, the diffraction limit was about 3.5 Å resolution which is much better than using laboratory-use X-ray generator as a source. But the radiation damage was so serious that the second shot from the same crystal showed a few spots at the low resolution range. This result suggests that the cryogenic experiment is necessary for the collection of full data set. The screening for the condition of cryo-protection is in progress.