Crystallographic study of IBDV virus-like particle

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Infectious bursal disease virus (IBDV) causes a highly contagious disease in young chicks and leads to significant economic losses in poultry industry. VP2 protein, which consists of 452 amino acid residues, is the primary immunogen of IBDV and contains the epitopes responsible for eliciting neutralizing antibodies. When the chimeric VP2 protein (rVP2H) of a local IBDV strain P3009 was expressed alone using baculovirus system, dodecahedral virus-like particles of approximately 23 nm diameter formed spontaneously. Highly pure rVP2H particles were successfully crystallized using vapor-diffusion method. The crystals had a maximum size of 0.4 mm and diffracted X-rays to 6 Å resolution Preliminary analysis of the diffraction data showed that the rVP2H crystals belong to the cubic space group $P2_13$ with unit-cell dimension of 324.8 Å. The crystal was soaking with Ta6Br14 cluster **(Fig.1)** and the XAFS scanning can measure the significant signal of Ta atoms, and the SAD data was collected at the energy of peak (fig.2).

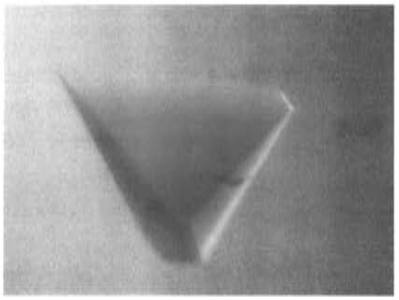

Fig.1

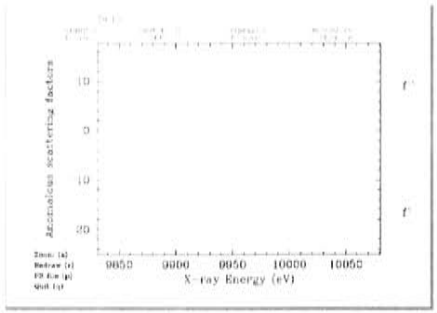

Fig.2

Finally, 630 frames are collected, and the further analysis is in progress now.

Diffraction data collection for several endonucleases bound with DNA

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Vivrio Vulnificus nuclease / 16mer double strand DNA complex

Vibrio Vunificus nuclease (Vvn) is a non-specific periplasmic nuclease capable of digesting nucleic acid. There is an Mg ion located in the domain of nuclease activity. Removing of Mg ion by EDTA was able to inactivate the Dnase activity of Vvn was proved pervious assay. The crystal structure of Vvn bound with 8mer DNA was solved in 2003. For obtaining the further information of interaction between Vvn and DNA, the oligo nucleotides of 16mer DNA were used as a material for Vvn (EDTA treaded) DNA complex crystallization and diffraction datasets were collected in BL12B. However, due to the poor diffracted feature crystal composed with long DNA, The diffraction shell was restricted around 2.9Å.

Preliminary stats table of diffraction data of Vvn 16mer

Cell constant A,B,C α,β,ρ	 65.2489 65.4025 82.0873 75.0007 75.2907 76.0579
Space group	P1
Rsym liner (20~2.9) Å	7.8%
Completeness (20~2.9) Å	77.4%

Colicin E7 Dnase Domain T2A /12mer DNA complex

High resolution X-ray diffraction data around 1.8 Å of 26 k Dnase Domain of Colicin E7 was collection in Spring-8 Synchrotron Radiation. The 3D structure shows the "ββα" feature of Dnase activity domain and Zn metal serves as a catalytic role near the key residues. Different divalent metals ion was soaking for further locating the position of Zn ion and anomalous diffraction data was collected for obtaining the stronger signal for the electron density for divalent metal.

For the characterization of bound between T2A and DNA, the crystals of EDTA treaded T2A combine with 8mer double strand oligo nucleotide were obtained. The x-ray data that diffracted up to 2.5Å was collected by BL12B2 of Spring-8 and structure was solved, but the coordinate of divalent ion was missing caused by the treatment of EDTA. Thus, a batch of crystals composed by mutated T2A bound with 12mer DNA was crystallized and we expected to localize the divalent metal around active site by solving the structure.

Preliminary stats table of diffraction data of T2A 12mer

Cell constant A,B,C α,β,ρ	 44.0304 44.0304 201.7112 90.0000
Space group	$P4_12_12$
Rsym liner (50~3.5) Å	12.6%
Completeness (50~3.5) Å	100%