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X-ray Crystallographic Analysis of the N-terminal Domain of XpsE from *Xanthomonas campestris*

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In the type II secretion pathway, extracellular proteins are secreted in two steps. In the first step, the proteins are exported from cytoplasm to periplasm through the Sec apparatus. Subsequeatly, they are secreted through a secretion apparatus composed of twelve to fourteen proteins to reach the extracellular space. In *Xanthomas campestris*, the secretion apparatus are constituted of the protein products of twelve genes. Among them, XpsE protein is predicted to be a cytoplasmic protein for its lack of membrance spanning sequence. It possesses a nucleotide binding motif, and has been suggested to have either ATPase or kinase activity. And it has been hypothesized to serve as an energy generating component for the secretion apparatus. To understand the structural basis of type II secretion pathway, we have crystallized the 21 kDa N-terminal domain of XpsE protein ($XpsE_N$) and its selenomethionine derivative in a PEG400-based crystallization condition. The $XpsE_N$ crystals belong to tetragonal space group $P4_32_12$ with cell parameters of a=b=56.1, c=102.7Å. And the crystal structure of $XpsE_N$ has been determined by Se-MAD approach by collecting diffraction data at three different wavelengths at Taiwan beamline SP12B2 (SPring8, Japan). The structure of $XpsE_N$ is bilobal: the N-subdomain of $XpsE_N$ resembles a distorted four-helix bundle, while the C-subdomain forms a α/β sandwich. Packing analysis revealed the presence of a crystallographic $XpsE_N$ dimer. As the dimerization of $XpsE_N$ has been implicated by results obtained from chemical cross-linking and gel-filtration analysis, the crystallographic $XpsE_N$ dimer may be of functional importance. In consistent with this prediction, multiple sequence alignment also shows that this interface is composed of highly conserved residues. The functional relevance of this interface will be further addressed by mutagenesis studies.

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X-ray Diffraction of Agglutinin and Cardiotoxin III

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Agglutinin

The seeds of the plant, *Abrus precatorius* contains two kinds of class II ribosome inactivating proteins (RIPs), the cytotoxic abrins and the low-*toxic Abrus precatorius* agglutinin (AAG). The molecular functional roles of RIP activity and toxicity are called for to elucidate their structures as tentative first steps.

The diffraction data of AAG crystal were collected in cold nitrogen gas stream using 15% Glycerol as cryoprotectant and recorded at BL12B2 Taiwan beamline at Spring-8, Japan. Diffraction data were integrated by HKL processing package. AAG crystals belonged to the primitive tetragonal crystal system, with cell parameters a = b = 137.050, c = 214.424Å Diffraction data were collected up to 2.47 Å resolution with 577355 total reflections and 141248 unique reflections, 99.1% completeness, linear R factor 0.084, and square R factor 0.072.

CardiotoxinIII

The lethal action of the snake venoms is mainly due to the presence of two types of highly homologous toxins, namely, the cardiotoxin and the neurotoxins. Their mode of action is still controversial as the existence of specific receptors is unclear. In order to understand these biochemical functions and an anticipation of potential the determinations of the 3D structures of cardiotoxins have attracted much interest.

The diffraction data of CTX3 crystal were collected in cold nitrogen gas stream using 26% Glycerol plus 7.4% water as cryoprotectant and recorded at BL12B2 Taiwan beamline at Spring-8, Japan. The X-ray wavelength was 1.00Å, with the oscillation range 1.0°. Diffraction data were integrated and scaled by HKL processing package. CTX3 crystals belonged to the primitive hexagonal crystal system, with cell parameters a = b = 83.300, c = 232.155 Å. Diffraction data were collected up to 3.50 Å resolution with 71772 total reflections and 11440 unique reflections, 99.5% completeness, linear R factor 0.118, and square R factor 0.120.

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