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S100A13 is a member of the S100 family of EF-hand-containing calcium-binding proteins. S100A13 plays an important role in the secretion of fibroblast growth factor-1 and interleukin-1α, two pro-angiogenic factors released by the non-classical endoplasmic reticulum-Golgi-independent secretory pathway. To gain insight into the structural basis of the interactions with these proteins, we determined the crystal structure of human S100A13 at pH 7.5 at 1.8-Å resolution. Human S100A13 was heterologously expressed in *Escherichia coli* purified and crystallized by the hanging-drop vapor diffusion method using PEG 3350 as the precipitant and at pH 7.5. The crystals diffracted X-rays from a synchrotron-radiation source beyond 1.8-Å resolution. The crystal space group was assigned as primitive orthorhombic $P2_12_12_1$ with unit-cell parameters a= 39.7Å, b= 59.2Å, c = 77.6Å. The structure was solved by molecular replacement and has been refined to a final R factor of 19.0% and a free R of 22.7%. The structure revealed that human S100A13 exists as a homodimer with two calcium ions bound to each protomer. The protomer is composed of 4 α-helices (α1-α4) forming a pair of EF-hand motifs. Dimerization occurs by hydrophobic interactions between helices α1 and α4 and by intermolecular hydrogen bonds between residues from helix α1 and the residues between α2 and α3 of both chains. Comparison between the crystal structures of human S100A13 at pH 7.5 (this study) and pH 6.0 (Li *et al.*, 2007) exhibited recognizable differences in the relative orientation (ca. 2.5°) between the protomers within the dimer and also remarkable differences in the side-chain conformations of several amino-acid residues.

Keywords: calcium-binding proteins, EF-hand proteins, X-ray crystallography of proteins

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X-ray analysis of FliJ, a cytoplasmic component of the flagellar type III protein export apparatus

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The flagellum is a motile organelle composed of the basal body rings and the tubular axial structure. The axial proteins synthesized in the cytoplasm are transferred into the central channel of the flagellum by the flagellar protein-export apparatus, which is classified into the type III protein export system. The apparatus is composed of six transmembrane proteins (FlhA, FlhB, FliO, FliP, FliQ, FliR) and three soluble components (FliH, FliI, FliJ). FliJ is an essential component for protein export. Although FliJ is thought to be a general chaperone, its function is still unclear. Here we report purification, crystallization and X-ray analysis of FliJ. Native FliJ was difficult to handle because of its strong tendency to form insoluble aggregates. Recently, we found that FliJ with extra three residues attached to the N-terminus as a remainder of His-tag is highly soluble. We obtained hexagonal bi-pyramid crystals from this FliJ variant, and determined the structure at 2.2 angstrom resolution using anomalous data from a mercury derivative crystal collected at SPring-8 BL41XU. We will discuss details of the structure and possible function of FliJ.

Keywords: Flagellum, FliJ, type III protein export apparatus

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Crystallographic study of zinc finger domain of Eco1 involved in sister chromatid cohesion

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The sister chromatid cohesion apparatus mediates physical pairing of duplicated chromosomes. This pairing is essential for appropriate distribution of chromosomes into the daughter cells upon cell division. The cohesion apparatus is also involved in gene expression and development. For instance, the Roberts syndrome in humans is caused by mutations in Eco1 homologous protein ESCO2, which is a component of the cohesion apparatus. Eco1 is originally found in budding yeast. Eco1 proteins are composed of variable N-terminal region and conserved C-terminal region composed of PCNA interacting protein box, zinc finger and acetyltransferase domains. Eco1 isn't required for binding of cohesin to chromosomes, but is needed to establish cohesion during S phase in *S. cerevisiae*. Furthermore, it has been reported that Eco1 interacts with PCNA, suggesting that it has a critical role during DNA replication. Although Eco1 has acetyltransferase activity in vitro, its inherent substrates in vivo are still unknown. Recent study shows that acetyltransferase domain of Eco1 is dispensable for S phase cohesion, but required for DSB-induced cohesion in G2/M. The zinc finger domain is alternatively required for chromosome segregation. Here we report crystallographic study of zinc finger domain of Eco1. Zinc finger domain of Eco1 was overexpressed as a GST-fused Protein in *E. coli*. The recombinant protein is purified by an affinity resin, removal of affinity tag by protease, gel-filtration chromatography. The purified protein was successfully crystallized using hanging drop vapor diffusion method. X-ray diffraction studies reveal that the crystal belongs to trigonal/hexagonal system with the cell dimensions of a = b = 73, c = 81Å, γ = 120°.

Keywords: sister chromatid cohesion, crystallization, zinc finger

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Crystallization of carbohydrate oxidase from *Microdochium nivale*

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Carbohydrate oxidase from *Microdochium nivale* is a flavoenzyme containing 475 amino acids with covalently linked flavin. The