

**P04.03.201***Acta Cryst.* (2008). A64, C294**The asymmetric architecture of 2Fe-2S IscU, a scaffold protein for iron-sulfur cluster biosynthesis**Keiichi Fukuyama<sup>1</sup>, Kei Wada<sup>1</sup>, Yoshimitsu Shimomura<sup>1</sup>, Yasuhiro Takahashi<sup>2</sup><sup>1</sup>Osaka University, Department of Biological Sciences, 1-1 Machikaneyama-cho, Toyonaka, Osaka, 560-0043, Japan, <sup>2</sup>Saitama University, Saitama, Saitama, 338-8570, Japan, E-mail : fukuyama@bio.sci.osaka-u.ac.jp

Iron-sulfur (Fe-S) clusters are ubiquitous prosthetic groups that are required to maintain fundamental life processes. The assembly of Fe-S clusters in several bacteria as well as eukaryotic mitochondria is achieved by a multicomponent system, called ISC machinery. In this machinery, IscU serves as a scaffold for assembly of a nascent Fe-S cluster, prior to its delivery to an apo-protein. The Hsp70-type molecular chaperone HscA and the J-type cochaperone HscB comprise a specialized chaperone system that selectively binds IscU and facilitate the cluster transfer process. We have determined the crystal structure of holo-IscU from the hyperthermophilic bacterium *Aquifex aeolicus* at 2.3 Å resolution, using multi-wavelength anomalous diffraction of the [2Fe-2S] cluster. IscU formed an asymmetric homotrimeric structure harboring only one [2Fe-2S] cluster that was coordinated by three cysteines and one histidine (Cys36, Cys63, His106, and Cys107) on the surface of just one of the protomers. The cluster was buried inside the homotrimer by the neighboring protomers. The three protomers were conformationally distinct from each other, and associated around a non-crystallographic pseudo-three-fold axis. This unique trimeric holo-IscU architecture was clearly distinct from other known monomeric apo-IscU/SufU structures, indicating that asymmetric trimer organization would be involved in the scaffolding function of IscU. This oligomeric holo-IscU structure provides mechanistic implications concerning the initial event in the assembly of Fe-S cluster, as well as the event in the transfer to target apo-proteins.

Keywords: iron sulfur clusters, iron sulfur proteins, crystal structure analysis

**P04.03.202***Acta Cryst.* (2008). A64, C294**Crystallographic study of Cu, Zn superoxide dismutase in extreme pressure conditions**Isabella Ascone<sup>1</sup>, Carmelinda Savino<sup>2</sup>, Eric Girard<sup>3</sup>, Anne-Claire Dhaussy<sup>4</sup>, Richard Kahn<sup>3</sup>, Roger Fourme<sup>1</sup><sup>1</sup>synchrotron-soleil, BP48 Saint Aubin, Gif sur Yvette, IDF, 91192, France, <sup>2</sup>Department of Biochemical Sciences and CNR, University of Rome La Sapienza, P.le A. Moro 5, 00185 Rome, Italy, <sup>3</sup>IBS, 41 rue Jules Horowitz, 38027 Grenoble, France, <sup>4</sup>CRISMAT Ensicaen, 6 bd Marechal Juin, 14704 Caen, France, E-mail : isabella.ascone@synchrotron-soleil.fr

High-pressure macromolecular crystallography is now a mature and powerful technique (Girard et al., 2007). We have investigated the effect of high pressure perturbation on crystal and molecular structures of oxidized bovine Cu,Zn superoxide dismutase (Cu,Zn SOD). This enzyme belongs to a family of metalloenzymes that catalyze the dismutation of the superoxide anion into dioxygen and hydrogen peroxide. Cu,Zn SOD has a key role in the cellular defence against oxidative damage. X-ray absorption spectroscopy had previously revealed (Ascone et al., 2000) that the metal site local structure is preserved beyond 1 GPa. X-ray crystallography was performed at ID27 beamline (ESRF, Grenoble, France) at room

temperature. In the crystalline state, the secondary and tertiary structures are preserved up to at least 1 GPa in spite of the fact that the enzyme is a dimer. The structure of oxidized Cu,Zn SOD at 0.57 GPa has been refined at 2.0 Å resolution, using a highly-complete data set. The effects of pressure on different regions of the molecule, including the active site, have been analyzed with reference to the oxidized and reduced PDB structures at room and liquid nitrogen temperature respectively. Moreover, advantages and limitations in coupling macromolecular crystallography with X-ray absorption spectroscopy for high-pressure studies, in non-crystalline and crystalline states, will be discussed.

## References

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Keywords: superoxide dismutase, dimer, high-pressure

**P04.03.203***Acta Cryst.* (2008). A64, C294**Insights into iron transport in *Helicobacter pylori* ferritin**Ji-Hye Lee<sup>1</sup>, Ki Joon Cho<sup>1</sup>, Hye Jeong Shin<sup>2</sup>, In Seok Yang<sup>1</sup>, Seung-Taik Lim<sup>1</sup>, Kyung Hyun Kim<sup>2,3</sup><sup>1</sup>Korea University, Food Technology, Anam dong, Sungbuk gu, Seoul 136-701, Seoul, Seoul, 136-701, Korea (S), <sup>2</sup>College of Engineering, Korea University, 5-1 Anam dong, Sungbuk gu, Seoul, Seoul, 136-701, Korea (S), <sup>3</sup>Dept. of Biotechnology & Bioinformatics, College of Science & Technology, Korea University, Chungnam 339-700, Korea (S), E-mail : jihyelee@korea.ac.kr

Ferritin, a biomineralizing protein found in organisms from all kingdoms, catalyzes the oxidation of iron into a ferric oxide core within the protein shell. Ferritin-mediated iron storage is required for the colonization and survival of *Helicobacter pylori* in human gastric mucosa, protecting the bacteria against acid-amplified iron toxicity. Here we report the crystal structures of *H. pylori* ferritin (Hpf) in four distinct forms: apo, low-iron bound, high-iron bound, and acidic pH. Despite high degree of sequence and structural homology with other ferritins, Hpf may use a pair of distinct histidine residues His93 and His96 located in proximity to the ferroxidase center as a novel metal binding site. At the high-iron bound form, a quartet of His149 at the entrance of the 4-fold symmetry channels undergoes marked conformational changes upon binding of Fe ions, which was found to be tightly coupled to Fe translocation. Hpf may have evolved to possess the dual transport mechanism for the control of irons.

Keywords: *Helicobacter pylori*, ferritin crystal structure, iron transport

**P04.03.204***Acta Cryst.* (2008). A64, C294-295**Structure of a member of glycoside hydrolase family 61: Are these true glycoside hydrolases?**Leila Lo Leggio<sup>1</sup>, Ditte H Welner<sup>1</sup>, Keith McFarland<sup>2</sup>, Jens-Christian N Poulsen<sup>1</sup>, Rune Salbo<sup>1</sup>, Sine Larsen<sup>1,3</sup>, Esben Friis<sup>4</sup>, Paul Harris<sup>2</sup><sup>1</sup>University of Copenhagen, Department of Chemistry, Universitetsparken 5, Copenhagen Oe, -, 2100, Denmark, <sup>2</sup>Novozymes, Inc., 1445 Drew Ave