DbeA and DbeA1 was initiated to understand the structure-function relationships of the wild type and the insertion mutant.

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**Keywords: haloalkane dehalogenase; catalytic peptide; halide-binding residues**

## FA1-MS03-P12

**1,3-Propanediol Dehydrogenase from Klebsiella Pneumoniae: Decameric Quaternary Structure and Possible Subunit Cooperativity.** Maria Arménia Carrondo[a], David Marçal[a,b], Ana Toste Rêgo[a], Francisco J. Enguita[b]. [a]*Instituto de Tecnologia Química e Biológica, Universidade Nova de Lisboa, 2781-901 Oeiras, Portugal.* [b]*Instituto de Medicina Molecular, Universidade de Lisboa, 1649-028 Lisbon, Portugal.*
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Alcohol dehydrogenases are a rather old subject. They play a central role in the most ancient business of biotechnology: alcoholic fermentation. As a consequence, they also play an important role in our liver and stomach, providing a line of defense against a potentially dangerous molecule, ethanol. It is therefore not strange that they were subject of early attention, with the first alcohol dehydrogenase purified and crystallized in 1937 [1]. However, there are many different enzymes that interconvert alcohols, aldehydes and ketones. [2]. The enzyme 1,3-propanodiol dehydrogenase from *Klebsiella pneumoniae* is a type III iron-dependent dehydrogenase, a not so well studied group of enzymes, with very few known structures [3]. *Klebsiella pneumoniae* is a nosocomial pathogen frequently isolated from opportunistic infections, especially in clinical environments [4]. In spite of its potential pathogenicity, this microorganism has several metabolic potentials that could be used in biotechnology applications. *K. pneumoniae* is able to metabolize glycerol as a sole source of carbon and energy [5]. 1,3-Propanediol dehydrogenase is the core of the metabolic pathway for the use of glycerol [6]. We have determined the crystallographic structure of 1,3-propanediol dehydrogenase, a type III Fe-NAD-dependent alcohol dehydrogenase, at 2.7-Å resolution. The structure of the enzyme monomer is closely related to that of other alcohol dehydrogenases. The overall arrangement of the enzyme showed a decameric structure, formed by a pentamer of dimers, which is the catalytic form of the enzyme. Dimers are associated by strong ionic interactions that are responsible for the highly stable in vivo packing of the enzyme. Kinetic properties of the enzyme as determined in the article would suggest that this decameric arrangement is related to the cooperativity between monomers.

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**Keywords: glycerol metabolism; type III alcohol dehydrogenase; 1,3-propanediol**

## FA1-MS03-P13

**Crystal Structure of Shikimate Dehydrogenase from *Helicobacter Pylori.*** Wen-Chi Cheng[a], Shuang-Chih Lin[a], Hung-Jung Wang[a], Jinn-Moon Yang[b], Jong-Yih Lin[c], Wen-Ching Wang[a]. [a]*Institute of Molecular and Cellular Biology and Department of Life Sciences, National Tsing Hua University, Hsinchu, Taiwan.* [b]*Institute of Bioinformatics and Department of Biological Science and Technology, National Chiao Tung University, Hsinchu, Taiwan.* [c]*National Chung Hsin University, Taichung, Taiwan.*
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Shikimate dehydrogenase (EC 1.1.1.25) catalyzes the NADPH-dependent reduction of 3-dehydroshikimate to shikimate, as well as its reverse, and has been developed as a promising target for the discovery of new antimicrobial agent, herbicides, and antiparasitic agents. It is the fourth enzyme in the shikimate pathway for aromatic amino acid biosynthesis in bacteria, fungi, and plants, but not mammals. The crystal structure of native shikimate dehydrogenase form *Helicobacter pylori* (HpSDH) was solved to 1.6Å resolution using single-wavelength anomalous dispersion methods, showing an N-terminal α/β domain and a C-terminal Rossmann domain. We have also determined the binary HpSDH·shikimate structure (1.4 Å) and the ternary HpSDH·shikimate·NADPH (2.0 Å) structure, respectively. These structures demonstrate that shikimate binds to the N-terminal domain, while NADPH binds to the Rossmann-fold domain. Furthermore, the apo-form adopts an open-state conformation, while the complex structures have a closed-form conformation. Crucial shikimate binding residues (Ser16, Ser18, Tyr21, Thr65, Lys69, Asn90,Asp105 and Gln237) are identified, which provide a basis for the structure-guided design of SDH inhibitors.

**Keywords:crystal structure; shikimate dehydrogenase; shikimate pathway; *helicobacter pylori***

## FA1-MS03-P14

**Crystallization of Bifunctional Catalase-phenol Oxidase(CATPO)from *Scytalidium Thermophilum.*** Yonca Yuzugullu[a], Chi Trinh[b], Arwen R. Pearson[b], Mark A. Smith[b], Simon Phillips[b], Ufuk Bakır[a], Michael J.McPherson[b], Zumrut B. Ogel[a]. [a]*Food Engineering Department, Middle East Technical University, Turkey.* [b]*Astbury Centre for Structural Molecular Biology, University of Leeds, UK.*
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Catalase is a common enzyme in all aerobic and many

anaerobic organisms. It decomposes hydrogen peroxide ($H_2O_2$) into one dioxygen ($O_2$) and two water molecules. Catalases can be divided into three subgroups based on structural and functional similarities: monofunctional heme (typical) catalases, catalase-peroxidases and manganese catalases. The first member of a novel fourth group of catalases, the catalase-phenol oxidases (CATPO), has been recently purified and crystallized by Sutay *et al.* (2008) from *Scytalidium*. It comprises 717 amino acids with a 19 amino acid signal sequence, and a 17 amino acid prosequence. It is a homotetrameric protein of molecular mass 320 kDa and subunit molecular mass 80 kDa. As well as peroxidase activity, CATPO is also able to oxidize various phenolic compounds in the absence of hydrogen peroxide. The goals of our current studies are to solve the three dimensional structure of CATPO and to clarify its catalytic mechanism(s) by studying mutations in the active site. We have obtained crystals of native CATPO that diffract to 2.8 Å and are pursuing crystals of CATPO mutants.

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**Keywords: catalase; phenol oxidase; *scytalidium thermophilum***

## FA1-MS03-P15

**Structural Studies of RNases H and Their Complexes with RNA/DNA Hybrids.** Marcin Nowotny[a,b], Sergei Gaidamakov[c], Robert J. Crouch[c], Wei Yang[a]. [a]*Laboratory of Molecular Biology, National Institute of Diabetes and Digestive and Kidney Diseases, NIH, Bethesda, MD, USA.* [b]*International Institute of Molecular and Cell Biology, Warsaw, Poland.* [c]*Laboratory of Molecular Genetics, National Institute of Child Health and Human Development, NIH, Bethesda, MD, USA.*
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RNases H are nucleases that bind RNA/DNA hybrids and degrade the RNA strand. We solved crystal structures of B. halodurans RNase H1 in complex with a 12-mer RNA/DNA hybrid [1]. They showed that the RNA strand of the hybrid is recognized by protein through extensive contacts with 2'-OH groups. The RNA strand adopts A-form conformation, but the DNA strand is B-form and only this form is complementary with the surface of the protein. Since RNA cannot adopt B-form conformation, the protein can only bind RNA/DNA hybrids and not dsRNA. Two $Mg^{2+}$ ions were observed at the active site, which suggest that the catalysis occurs through a two-metal ion mechanism. We also solved crystal structures of human RNase H1 in complex with various RNA/DNA hybrids. They confirmed that the substrate recognition and enzymatic mechanism for human protein is the same as for bacterial RNases H.

[1] Nowotny, M et al. (2005). Crystal structures of RNase H bound to an RNA/DNA hybrid: substrate specificity and metal-dependent catalysis. Cell 121(7): 1005-16

**Keywords: RNase H; RNA/DNA hybrid**