

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.

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

[1] Sutay Kocabas, D., Bakir, Ufuk., Phillips, Simon E. V., McPherson, Michael J. and Ogel, Zumrut B. *Appl Microbiol Biotechnol* **2008**, 79, 407–415. [2] Zamocky, M., Koller, F. *Progress in Biophysics & Molecular Biology* **1999**, 72, 19-66

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**Structural Studies of RNases H and Their Complexes with RNA/DNA Hybrids.** Marcin Nowotny<sup>a,b</sup>, Sergei Gaidamakov<sup>c</sup>, Robert J. Crouch<sup>c</sup>, Wei Yang<sup>a</sup>. <sup>a</sup>Laboratory of Molecular Biology, National Institute of Diabetes and Digestive and Kidney Diseases, NIH, Bethesda, MD, USA. <sup>b</sup>International Institute of Molecular and Cell Biology, Warsaw, Poland. <sup>c</sup>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