binding channel shows conservation of positively charged residues, which are possibly involved in complex formation with the H-protein. These results provide insights into the molecular basis of nonketotic hyperglycinemia.

Keywords: multienzyme complexes, disease-related structures, vitamin B6

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Structure and Inhibition Mode of Protein I^c in Complex with Carboxypeptidase Y

Yasuo Hata[a], Minoru Hayashida[a], Tomomi Fujii[a], Joji Mima[b], Rikimaru Hayashi[b], Mitsuyoshi Ueda[b], [a]*Institute for Chemical Research,* [b]*Graduate School of Agriculture, Kyoto University, Kyoto, Japan.* E-mail: hata@scl.kyoto-u.ac.jp

Carboxypeptidase Y (CPY) inhibitor I^c from the yeast, consisting of 204 amino acid residues, belongs to the phosphatidylethanolamine-binding protein (PEBP) family. The 2.7 Å crystal structure of the I^c-CPY complex has been solved by molecular replacement [1, 2].

The structure of I^c consists of a major β-type domain and an N-terminal helical segment. I^c has two CPY-binding sites: the N-terminal inhibitory reactive site and the secondary CPY-binding site which interact with the S1 substrate-binding site of CPY and the hydrophobic surface flanked by the active site of the enzyme, respectively. I^c also has the ligand-binding site, the putative binding site of the polar head group of phospholipid, which is conserved among PEBPs and accommodates a sulfate ion in the crystal structure.

Along with the complex structure of I^c, its mutational analyses for inhibitory activity and binding to CPY demonstrate that the N-terminal inhibitory reactive site is essential for the complex formation with CPY as well as enzyme inhibition and that the I^c binding to CPY forms a novel mode of the proteinase-protein inhibitor interaction. The unique binding mode of Ic toward CPY gives insights into not only the inhibitory mechanism of PEBPs toward serine proteinases but also the biological functions of I^c belonging to the PEBP family.

[1] Mima J., Hayashida M., Fujii T., Hata Y., Hayashi R., Ueda M., *Acta Crystallog. Sect. D*, 2004, **60**, 1622. [2] Mima J., Hayashida M., Fujii T., Narita Y., Hayashi R., Ueda M., Hata Y., *J. Mol. Biol.*, 2005, **346**, 1323.

Keywords: CPY inhibitor, I^c, PEBP family

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Structures of a Novel *N*-acetyl-L-ornithine Transcarbamylase

Dashuang Shi[a], Xiaolin Yu[a], Lauren Roth[b], Hiroki Morizono[a], Norma M. Allewell[b], Mendel Tuchman[a], [a]*Children's National Medical Center, George Washington University.* [b]*College of Life Science, University of Maryland, USA*. E-mail: dshi@cnmcresearch.org

N-acetyl-L-ornithine transcarbamylase, a new member of the transcarbamylase family, is an essential enzyme to synthesize arginine in a few of eubacteria. Since this enzyme is not present in other bacteria, plants, animals and human, *N*-acetyl-L-ornithine transcarbamylase could provide a potential non-toxic target for specific inhibition to control certain agriculture and human pathogens. We report here the crystal structures of the binary complexes of enzyme from *Xanthomonas campestris* with its substrate carbamoyl phosphate or *N*-acetyl-L-ornithine only and the ternary complex with carbamoyl phosphate and *N*-acetyl-L-norvaline. Comparison of the structures of the enzyme in the different substrate binding states demonstrates that the binding mechanism of this novel transcarbamylase is different from those of aspartate and ornithine transcarbamylases. The enzyme can bind carbamoyl phosphate and *N*-acetyl-L-ornithine independently, and does not require one of substrate binds first in order to bind the second substrate. The main conformational change is the ordering of the 80's loop upon binding the carbamoyl phosphate besides a small domain closure around the active site. The structures of the complexes provide insight into how the enzyme facilitates the carbamoyl group transfer, and provide a starting point for inhibitor design.

Keywords: carbamoyltransferase, acetylornithine, arginine pathway

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Crystal Structure of Spermidine Synthase from *Helicobacter pylori*

Yuh-Ju Sun[a], P.-K. Lu[a], S.-Y. Chien[a], J.-Y. Tsai[a], C.-T. Fong[a], M. J. Lee[b], H. Huang[b], [a]*Institute of Bioinformatics and Structural Biology.* [b]*Institute of Biotechnology, National Tsing Hua University, Hsinchu, Taiwan 300, ROC.* E-mail: yjsun@life.nthu.edu.tw

Spermidine synthase (putrescine aminopropyltransferase, PAPT) catalyzes the transfer of the aminopropyl group from decarboxylated *S*-adenosylmethionine (dcAdoMet) to putrescine in the biosynthesis of spermidine. PAPT of *Helicobacter pylori* (HpPAPT) is encoded by the *speE* gene. HpPAPT has less than 20% of sequence identity with other PAPTs, even containing no signature sequence. The three-dimensional structure of HpPAPT has been determined by multiwavelength anomalous dispersion (MAD) in this study. HpPAPT consists of an N-terminal beta-stranded domain and a C-terminal Rossmann-like domain, with a binding pocket between two domains. The oligomerization of HpPAPT is mostly made by the N-terminal domain and sensitive to the pH values of buffer. Our structure illustrates that HpPAPT has a distinctive binding pocket with a bigger space, a unique electrostatic potential surface of less acidity, and numerous unconserved residues. Due to the lack of the gatekeeping loop, HpPAPT may need to perform a significant conformation change to accommodate the ligand binding.

Keywords: spermidine synthase, putrescine aminopropyl-transferase, *Helicobacter pylori*

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Helix-formation Due to the Binding of α- with β_2-subunit of Tryptophan Synthase

Kazuya Nishio[a], Yukio Morimoto[b], Manabu Ishizuka[c], Kyoko Ogasahara[a], Katsuhide Yutani[d],Tomitake Tsukihara[a], [a]*Inst. of Protein Res., Osaka Univ., Japan.* [b]*RRI, Kyoto Univ., Japan.* [c]*Eng., Univ. Tokushima, Japan.* [d]*RIKEN Harima Inst., Japan.* E-mail: nishi-p@protein.osaka-u.ac.jp

When the tryptophan synthase α- and β_2-subunits combine to form the $\alpha_2\beta_2$ complex, the enzymatic activity of each subunit is stimulated by one to two orders of magnitude. In order to elucidate the structural basis of this mutual activation, it is necessary to determine the structures of the α- and β-subunits alone and together with the $\alpha_2\beta_2$ complex. The crystal structures of the tryptophan synthase $\alpha_2\beta_2$ complex from *S. typhimurium* (*St*$\alpha_2\beta_2$) has been reported. Therefore, we determined the crystal structure of the tryptophan synthase α-subunit alone from *E. coli* (*Ec*α) at 2.3Å resolution. The biggest difference between the structures of the *Ec*α and the α-subunit in the *St*$\alpha_2\beta_2$ (*St*α) was as follows. The helix-2' in the *St*α including an active site residue (Asp60) changed to a flexible loop in the *Ec*α. The conversion of the helix to a loop resulted in collapse of the correct active site conformation. This region is also an important part for the mutual activation in the *St*$\alpha_2\beta_2$ and interaction with the β-subunit. These results suggest that the formation of helix-2' essential for the stimulation of the enzymatic activity of the α-subunit is constructed by the induced-fit mode involved in conformational changes upon interaction between the α- and β-subunits.

[1] Nishio K., et al.. *Biochemistry*, 2005, **44**, 1184

Keywords: protein crystallography, biological structure-activity relationships, protein-protein interactions

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Structure of Statironary Phase Survival Protein SurE from *Thermus thermophilus*

Wakana Iwasaki[a], Kunio Miki[a,b], [a]*RIKEN Harima Institute/SPring-8.* [b]*Graduate School of Science, Kyoto University, Japan.* E-mail: wiwasaki @spring8.or.jp

Stationary-phase survival protein SurE is a metal ion-dependent