

we have determined the crystal structure of YlqF from *Thermotoga maritima* in complex with GDP, GTP, and a non-hydrolyzable GTP analog. YlqF is a circularly permuted GTPase. It is composed of two domains: an N-terminal G domain and a C-terminal basic alpha-helical bundle domain. The structures of Tm YlqF bound with different ligands reveal a significant difference in the relative orientation of the two domains, when we compare the GDP-bound structure with either the GTP- or GNP-bound structure. Our study thus provides a glimpse of a possible conformational change of YlqF upon GTP hydrolysis.

Keywords: GTPase, circular permutation, YlqF

## P04.02.79

*Acta Cryst.* (2008). A64, C255

### Crystal structure of L-sorbose reductase from *Gluconobacter frateurii* at 2.4 Å resolution

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L-Sorbose reductase (SR) from *Gluconobacter frateurii* is an NADPH-dependent reductase that catalyzes the reduction of L-sorbose to D-sorbitol with a high substrate specificity. To understand the structural bases of substrate specificity and catalytic mechanism of SR, we have determined the first crystal structure of SR in complex with L-sorbose at 2.4-Å resolution. SR adopts the Rossmann fold and belongs to the short-chain dehydrogenase/reductase family. SR takes a tetrameric assembly in crystal and solution at optimal pH's, pH 6.2 (for the conversion of L-sorbose to D-sorbitol) and pH 9.0 (for the conversion of D-sorbitol to L-sorbose). Although the crystal structure does not contain the co-factor NADPH, the NADPH-binding site can be predicted by comparing the structures of the SDR family members. The bound L-sorbose was located in the active-site pocket near the putative NADPH-binding site and recognized by  $\eta$ 1,  $\alpha$ 5 and  $\alpha$ 6 helices and loops.

Keywords: crystal structures, enzyme structure, enzyme ligand complexes

## P04.02.80

*Acta Cryst.* (2008). A64, C255

### Structural studies by X-ray on enzymes involved in propionate metabolism from membrane integrated protein leukotriene C4 synthase

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Propionate is the second most abundant organic compound in the soil. The metabolism of propionate has been investigated in *Salmonella enterica serovar typhimurium* and *Escherichia coli*. The first

reaction in the anaerobic breakdown of L-threonine to propionate is catalyzed by the biodegradative threonine deaminase (TdcB), which catalyzes the deamination of L-threonine to  $\alpha$ -ketobutyrate. The X-ray structure of TdcB from *S. typhimurium* was determined in two forms to resolutions of 1.7 Å and 2.2 Å, respectively. The dimeric protein displays an interesting variation in the quaternary association of subunits in one of the crystal forms. CMP binding alters the oligomeric state of TdcB from a dimeric to a tetrameric form. The structure of the TdcB-CMP determined at 2.5 Å resolution suggests that the changes induced at the dimer interface by ligand binding are essential for tetramerization. The structural alterations also appear to account for enzyme activation and increased affinity for L-threonine. Propionate kinase (TdcD) catalyzes the last step of non-oxidative degradation of threonine to propionate by enabling the conversion of propionyl phosphate and ADP to propionate and ATP. The crystal structures of unliganded TdcD from *Salmonella typhimurium* and its complexes with ADP and AMPPNP have been determined to resolutions of 2.6, 2.2 and 2.3 Å, respectively. Examination of active site pocket revealed the plausible structural rationale for the higher specificity of the enzyme towards propionate than acetate. The structure of TdcD in complex with ATP determined at 1.98 Å resolution revealed a novel reaction catalyzed by the enzyme, i.e., synthesis of 5', 5'''-P1, P4-tetraphosphate (Ap4A). This was further confirmed by determination of the structure of TdcD - Ap4A complex.

Keywords: propionate, threonine deaminase, propionate kinase

## P04.02.81

*Acta Cryst.* (2008). A64, C255-256

### Crystal structure of tetrameric malate dehydrogenase from *Antarctic psychrophile*

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The psychrophilic and thermolabile malate dehydrogenase was isolated from *Flavobacterium frigidimarum* KUC-1 (FfMDH) living in Antarctic seawater. The enzyme is a homotetramer with a molecular weight of 123 kDa and consists of 311 amino acid residues per subunit. To understand structural features of psychrophilic enzymes adapting to cold environment, the structure of FfMDH has been solved by X-ray analysis. FfMDH was crystallized by a hanging-drop vapor diffusion method at 288 K. The crystal has space group  $P3_221$  with unit cell dimensions of  $a=b=147.8$  Å and  $c=165.1$  Å, and contains four subunits in the asymmetric unit. Diffraction data were collected at 100 K to 1.8 Å resolution at Beamline BL-5A, PF, Tsukuba. The crystal structure was solved by molecular replacement as the search model of a hybrid MDH (PDB code 1GUZ) using the program MOLREP. It was refined for 1.8 Å diffraction data to  $R_{work}=0.146$  and  $R_{free}=0.164$  with the programs CNS and Refmac5. The structure of FfMDH is a dimer of dimers and similar to those of MDHs from other bacteria. A structural comparison of four MDHs from the present psychrophilic, mesophilic, moderate thermophilic and thermophilic bacteria, however, reveals several differences in molecular architecture, especially intersubunit interaction. The number of ion pairs in the FfMDH tetramer is smallest among the MDH molecules compared. FfMDH has no intersubunit ion-pairs, although three other MDHs have two to six ion-pairs between every two subunits in each molecule. The lack of intersubunit ion-pair in the oligomeric molecule results in the loose contact among the subunits