Ornithine aminotransferase (OAT) catalyses exclusively the transfer of the delta-amino group of ornithine to alpha-ketoglutarate, forming glutamate-r-semialdehyde and glutamic acid. It is one of the major metabolic enzymes that modulate nitrogen fixation in the urea cycle as well as the synthesis of arginine, proline, and spermidine.

Inhibition of OAT has recently been explored as means of detoxification in animal experiments. In view of its diversified roles, it is of interest to compare the structure of OAT with other subgroups of the transaminase superfamily in order to understand the basis of its extraordinary specificity.

The structure of OAT is presented at 2.5 Å resolution. The recombinant enzyme crystallized in the trigonal space group P3(2)121 with one and a half dimers per asymmetric unit. The refined model has a R-factor of 17.5% (free-R = 23.3%) and excellent stereochemistry. Each subunit of the dimeric enzyme contains three distinct regions: an extended N-terminal segment, a cofactor binding large domain, and a relatively flexible small domain. The OAT dimer is stabilized by a large contact surface between the large domains of the two subunits and between the N-terminal segment of one subunit with the large domain of the other. The pyridoxal-5-phosphate (PLP) cofactor is bound to the large domain close to the subunit interface and is stabilized through multiple hydrophobic and electrostatic interactions to both subunits. Glu235 and Arg413, which form a salt bridge at the entrance to the active site pocket, are the most probable residues for the binding of ornithine. The location of Glu235 explains the selectivity of OAT for the delta-amino group despite the presence of the more reactive alpha-amino groups of ornithine and glutamate.

Branched-chain amino acid aminotransferase from Escherichia coli (EC 2.6.1.42) (BCAT) is one of vitamin B6 enzyme having pyridoxal-5'-phosphate (PLP) as a co-factor. BCAT forms a hexamer (M.W. 204,000 Da) of six identical subunits each consisting of 308 residues (M.W. 34,000 Da). Crystallization of BCAT was performed by hanging-drop vapor diffusion method. Two polymorphic crystals with good quality for X-ray crystallography were obtained and diffracted to 2.2 Å resolution with a conventional X-ray source. The crystals belong to the orthorhombic space group C2221 with unit cell dimensions of a=156.0, b=101.2, c=141.3 Å, and the monoclinic space group C2 with unit cell dimensions of a=135.3, b=144.1, c=102.9 Å, β=136.1°, respectively. Each Matthews’ Vm value is 2.7 (C2221) and 3.4 (C2), which indicates three subunits related by a three-fold noncrystallographic axis in the asymmetric unit. Multiple isomorphous replacement (MIR) techniques were used to determine the three-dimensional structure of BCAT. We succeeded in preparing two kinds of Hg derivatives (EMTs and CH3CH2HgCl), and initial phasing at 3.0 Å resolution.

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