ESRF	**Experiment title:** *E. coli* orotidine 5′-monophosphate decarboxylase	**Experiment number:** LS1389
Beamline: BM14	**Date of experiment:** from: 02/04-99 to: 04/04-99	**Date of report:** 24/1-00
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Report:

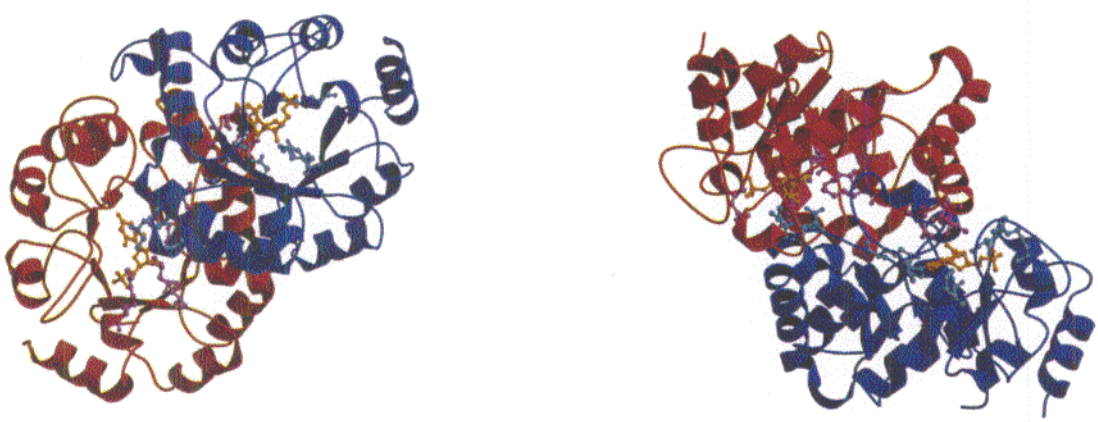
Orotidine 5'-monophosphate decarboxylase (ODCase) catalyses the decarboxylation of orotidine 5'-monophosphate (OMP) to uridine 5'-monophosphate (UMP). ODCase is one of the most proficient enzymes known. At ambient temperatures it enhances the rate of reaction by a factor of 10^{17} [1]. Considering the extreme proficiency, the mechanism of ODCase is unique, because no cofactor is required for the decarboxylation.

The data collection on the cryo cooled ODCase:BMP crystals was performed at beam-line BL I711 at MAX-II. The crystals diffracted to 2.5 Å. A MAD data collection on the selenomethionine substituted crystals was performed at beam-line BM14 at the ESRF. These crystals diffracted to 3.0 Å. A change of the unit cell of the selenomethionine substituted crystal form was observed as compared to the crystals of the native protein. This change was seen as the appearance of rather weak diffraction spots between the diffraction spots corresponding to the unit cell and space group of the native protein complex.

The diffraction pattern of the native ODCase:BMP complex was indexed in space group $P2_12_12_1$. The diffraction pattern from the selenomethionine substituted ODCase:BMP

complex could be indexed in space group $C222_1$ or in space group $P2_1$. By looking at the merging statistics, we determined the latter of these to be correct - meaning that the asymmetric unit of the selenomethionine substituted complex is actually four times larger than of the native complex. The cell change of the selenomethionine substituted crystal form was ignored and only reflections corresponding to the small unit cell were included - hence assuming space group $P2_12_12_1$. SOLVE [2] was able to localise 24 of 32 selenium atoms in the "small" asymmetric unit. The non-crystallographic symmetry between the four subunits (two dimers) was found manually and the maps were averaged using the CCP4 program DM [3]. From this map about 2/3 of the structure was built. After a few cycles of simulated annealing using CNS [4], we were able to refine the structure to the native dataset, hereby overcoming the problem of pseudo-symmetry and getting a final structure resolution of 2.5 Å (R=0.217; R_{free}=0.261). Residues 12 to 242 are included in the model, indicating that the N-terminal and the last few residues of the C-terminal are disordered.

The asymmetric unit of the native ODCase:BMP complex contains four subunits arranged as two dimers [5]. Each subunit folds up as an α/β-barrel with ten α-helices surrounding the eight central β-strands. The homodimer of the ODCase:BMP complex is very closely packed with a BMP molecule in the C-terminal end of each barrel. The residues that interact with BMP come from both subunits, so the two active sites are effectively shared by the two subunits shown in the figure.



Homodimer of E. coli ODCase. One subunit is coloured red, the other blue. Completely conserved residues are emphasised with ball-and-sticks. The inhibitor, BMP, is drawn in yellow.

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