***E. Coli* Dihydroorotase: Loop Movement and Cooperativity**

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Dihydroorotase (DHOase) is a zinc metalloenzyme that catalyses the reversible cyclization of *N*-carbamyl-L-aspartate (CA-asp) to dihydroorotate (DHO) in the *de novo* pyrimidine biosynthesis. The first structure of a DHOase (from *E. coli*) has been reported to a resolution of 1.7 Å with one homodimer per asymmetric unit [1].

We have collected data from crystals of *E. coli* DHOase grown in the presence of product, DHO and refined the structure to 1.9 Å resolution [2]. As in the original report [1], we find the product DHO bound in the active site of one subunit and CA-asp in the active site of the other. Importantly, we have resolved the conformations of residues B109-112, which were disordered in the reported structure. These residues comprise a loop which takes on different orientations in the two subunits, depending on whether DHO or CA-asp is present in the active site. This is accompanied by movements of residues A/B256-258 which seem to 'communicate' between subunits the respective contents of the active sites. Subsequent kinetic analysis at low DHO concentrations shows positive cooperativity [2].

Aspects of this structure of DHOase will be discussed. In addition, the structures of inhibitor complexes and site-directed mutagenesis that allow us to understand more about this loop movement will be described in relation to the enzyme mechanism.

[1] Thoden J. B. et al., *Biochemistry*, 2001, **40**, 6989. [2] Lee M. et al., *J. Mol. Biol.*, 2005, *in press.*

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