

Structure of the Mannitol dehydrogenase from *Agaricus bisporus*

Mannitol, an acyclic six-carbon polyol, is one of the most abundant sugar alcohols occurring in nature. In the button mushroom, *Agaricus bisporus*, it is synthesized from fructose by the enzyme mannitol dehydrogenase[§] (MtDH) (EC 1.1.1.138) using NADPH as a cofactor. Mannitol serves as the main storage carbon (up to 50% of the fruit body dry weight) and plays a critical role in growth, fruit body development, osmoregulation and salt tolerance (1).

Crystals of MtDH generally belonged to the space group C2, with cell parameters $a = 227$, $b = 125$, $c = 133$ Å and $\beta = 118^\circ$ and diffracted to 1.5 Å resolution at **BM01A**. The structure was solved by molecular replacement with mouse lung carbonyl reductase (MLCR) as probe. The sequence identity between MtDH and MLCR is 31%. MtDH is a functional tetramer in solution. The asymmetric unit of the crystal lattice is occupied by three tetramers, giving a total of 3180 amino acid residues. Initial phases from molecular replacement were automatically improved with the program ARP/wARP and the model was completed within the program O. The difference in anisotropic movement between the individual tetramers was accounted for by refining independent parameters T, L and S for each subunit within REFMAC. The final model consists of 3132 amino acids, 12 molecules of the cofactor NADP⁺ and 3000 water molecules. The final *R* factor was 19.3% (*R* free 20.9%) (2).

MtDH is a member of the short chain dehydrogenase/reductase superfamily of proteins. The NADP binding rossman fold is extended by a seventh β -strand with a left handed connection between strand F and G. The insertion loops between strand E, F and G are responsible for the binding of the sugar substrate.

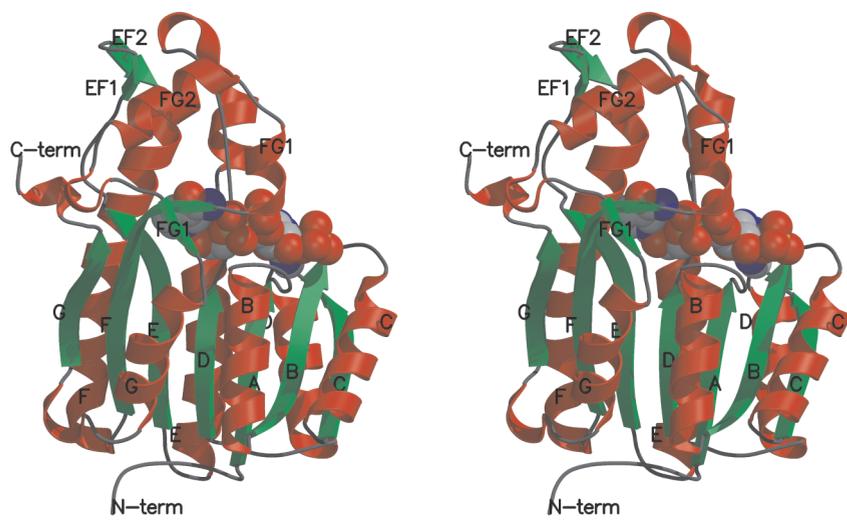
Literature:

1. Jennings, D. H. (1984) *Adv. Microb. Physiol.* **25**, 149-93
2. Sassoon, J., Hörer, S., Stoop, J., Mooibroek, H., and Baumann, U. (in press) *Acta Crystallogr. D Biol. Crystallogr.*

Figure 1: Stereo ribbon representation of MtDH from *A. bisporus*. 1a: Representation of a single subunit of MtDH and the cofactor NADP⁺. 1b: Quaternary structure of MtDH showing one tetramer. The C-termini coordinate two nickel ions (blue).

Figure 1

a



b

