The crystal structure of *Pichia pastoris* lysyl oxidase at 1.23Å reveals a lysine-lysine covalent cross-link, dehydrolysinonorleucine

A.P. Duff[1], A.E. Cohen[2], P.J. Ellis[2], D.B. Langley[1], D.M. Dooley[3], H.C. Freeman[1] and J.M. Guss[1], [1]School of MMB, University of Sydney, NSW 2006, Australia, [2]Stanford Synchrotron Radiation Laboratory, CA, USA and [3]Chemistry and Biochemistry, Montana State University, Bozeman MT, USA.

We have refined the structure of *Pichia pastoris* lysyl oxidase (PPLO) in a new crystal form at 1.23Å resolution with R = 0.112 and Rfree = 0.146. PPLO is a copper amine oxidase (CuAO) containing a trihydroxyphenylalanine quinone (TPQ) cofactor. PPLO is unusual in being able to oxidise the side chain of lysine residues in a polypeptide. In this respect, it is functionally related to another class of CuAOs of unrelated sequence, which contain the related quinone cofactor, lysine tyrosylquinone (LTQ). The asymmetric unit comprises residues 43 to 779 of the polypeptide, 7 carbohydrate residues, the active-site Cu atom, an imidazole molecule bound in the substrate-binding site, 2 buried Ca^{2+} ions, 5 surface Mg^{2+} ions, 5 surface Cl^{-} ions, and 1045 water molecules. The cofactor, TPQ, and some other active site residues are poorly ordered, in contrast to the high degree of order of their other neighbours. A covalent cross-link between two lysine residues, Lys 778 and Lys 66, is observed. The cross-link, dehydrolysinonorleucine, is formed by the oxidation of Lys 778, followed by spontaneous reaction with Lys 66.