PS04.02.45 CRYSTAL STRUCTURE OF THE GOWER II HUMAN EMBRYONIC HEMOGLOBIN ($\alpha_2\varepsilon_2$). A.J. Sutherland-Smith, H.M. Baker, and E.N. Baker, Department of Biochemistry, Massey University, Palmerston North, New Zealand and R.M. Mould, O.M Hofmann and T. Brittain, School of Biological Sciences, University of Auckland, New Zealand

Three hemoglobin molecules (Gower I, Gower II and Portland) are synthesised by the human embryo between weeks two and twelve of gestation. These hemoglobins appear to function as scavengers of O_2 from the mother's interstitial fluid before the placenta has developed. Functional Gower II hemoglobin is a tetramer comprising the adult α chain and the embryonic ε, which has 79% sequence identity to the adult β. Binding studies have indicated that Gower II hemoglobin binds O_2 cooperatively, with a higher affinity than the adult molecule, and displays similar allosteric behaviour towards H^+, Cl^- and 2,3 DPG.

Gower II hemoglobin with carbon monoxide bound was crystallised. The crystals proved to be tetragonal (spacegroup $P4_32_12$) a=b=62.8, c=320.8Å with one $\alpha_2\varepsilon_2$ tetramer in the asymmetric unit. The 3D structure has been solved at 2.9Å resolution by molecular replacement and refined to a crystallographic R-factor of 0.204 (R_{free} of 0.279) with good geometry.

The quaternary structure is very similar to that of the adult molecule. Within the ε subunit the main difference from the adult β is a small shift of the N terminal helix over the central cavity of the tetramer. The environment of the heme pocket is like that of the adult with the major variation being a closer packing of Ser 70 (Ala in β). Clear density is visible for the bound CO ligand.

PS04.02.46 CRYSTAL STRUCTURE OF A BACTERIAL COPPER-CONTAINING AMINE OXIDASE FROM *ARTHROBACTER GLOBOFORMIS* AT 2.8Å RESOLUTION. Matthew C.J. Wilce, Hans C. Freeman, J. Mitchell Guss, Vinay Kumar (University of Sydney, NSW 2006, Australia) and William S. McIntire, (Department of Veterans Affairs Medical Center, San Francisco, CA 94143, USA)

We present the crystal structure and structural analysis of a bacterial copper-containing amine oxidase (AO). AOs are homodimeric proteins with molecular weights of between 70-90 kDa per subunit. Their function is the oxidative metabolism of amines in the presence of molecular dioxygen. They are involved in many fundamental cellular processes including: tissue differentiation, tissue development, wound healing, cancer and possibly programmed cell death. AOs are of particular importance in gram-positive methylotrophs, including *Arthrobacter* spp., as these organisms are able to utilise methylamine as their sole carbon and energy source.

A. globoformis AO crystallises in a number of forms with and without the presence of ammonium salts that are known to inactivate the enzyme. The structure of the crystal form known as type II is reported here. The crystals were grown from $LiSO_4$. The space group is *C*2 (*a*=157.6, *b*=64.3, *c*=92.6 Å, β=112.6°) with one subunit per asymmetric unit. Molecular replacement was used to determine an initial phase set. Both the pea seedling and *E. coli* AO [Parsons, M.R. *et al.* (1995). *Structure*, **3**, 1171-1184] structures were successfully used as search models. The structure has been refined at 2.8Å resolution. The structure of *A. globoformis* AO is compared with both the *E. coli* AO and pea seedling AO structures with particular reference to the active site.

PS04.02.47 CRYSTAL STRUCTURE OF AN INHIBITOR COMPLEX WITH THE QUINOENZYME AMINE OXIDASE IDENTIFIES THE ACTIVE SITE BASE. Carrie M. Wilmot, Mark R Parsons, Maire A. Convery, Veronica Blakeley, Adam S. Corner, Kapil D.S. Yadav*, Mike J. McPherson, Peter F. Knowles & Simon E.V. Phillips, Department of Biochemistry and Molecular Biology, The University of Leeds, Leeds, LS2 DJT, United Kingdom* Present address: Department of Chemistry, University of Gorakhpur, Gorakhpur 273009, India

The crystal structure of the complex between the copper amine oxidase from *Escherichia coli* (ECAO) and a covalently bound inhibitor, 2-hydrazinopyridine, has been determined to a resolution of 2.5Å. The enzyme contains a cofactor, 2,4,5-trihydroxyphenylalanine quinone (TPQ), which is formed by the post-translational modification of a tyrosine residue. The inhibitor covalently binds at the 5 position of the quinone ring. This complex is analogous to the substrate Schiffs base formed during the reaction. The inhibitor structure has a nitrogen in place of a carbon in the aromatic primary amine substrate, and this prevents the abstraction of a proton by the catalytic base, which would allow the reaction to proceed. The electron density of the Schiffs base moeity in the complex is clear. The inhibitor nitrogen is hydrogen bonded to the sidechain of Asp383, a conserved residue among the known amine oxidase sequences, identifying it as the probable base. The positioning of Asp383 is such that the pro-**S** proton of a susbtrate would be abstracted. Th TPQ/inhibitor moeity is not directly interacting with the copper. The O4 position on the quinone ring is hydrogen bonded to the axial water ligand of the copper. The O4 position on the quinone ring is involved in a symmetrical hydrogen bond with the hydroxyl of the conserved residue Tyr369. The distance between the oxygens is less thsn 2.5Å, consistent with a shared proton, and suggesting ionisation at the O4 position of the quinone ring. The Tyr369 residue would appear to play an important role in stabilising the position of the quinone/inhibitor complex.