s8a.m4.p14.la **Crystal Structure of Human Placental Alkaline Phosphatase.** M.H. le Du*, T. Stigbrand?, M. Taussig,#, A. Ménez*, and E. Stura*. *Département d'Ingénierie et d'Etudes des Protéines (DIEP), CEA, C.E. Saclay, 91191 Gif-sur-Yvette Cedex, France. ?Department of Immunology, Umea University Sweden; and #Laboratory of Molecular Recognition, The Babraham Institute, Babraham, Cambridge CB2 4AT, UK.
Keywords: alkaline-phosphatase, tissue-specificity, extra-domains.

Placental alkaline phosphatase (PLAP) is one of three tissue specific alkaline phosphatases (AP) and probably a late development in the evolution of mammals. It is also an important cancer marker as its ectopic expression has been observed in a number of cancers. There are large differences in enzymatic activity between PLAP and other AP, probably connected to their specific function in each tissue. Under optimal conditions, PLAP is ten times less active than the intestinal enzyme, but its activity is further reduced in its physiological environment. The X-ray structure of PLAP at 1.8Å resolution enables us to overcome the lack of information between the mammalian APs and the only known Escherichia coli AP structure. It reveals three extra domains which are probably responsible for tissue specificity since more than half of the substitutions between the human enzymes are found within these domains. The first domain is a long N-terminal a-helix which embraces the second monomer. The second is interfacial, crown shaped, and mainly composed of insertion loops. The third includes a b-sheet flanked by two a-helices, a fourth metal site, and one of the two carbohydrate chains. The position of the two latter domains, on either side of the active site, might be functionally relevant. The structure brings insights concerning both the allostery observed in PLAP, and the specific uncompetitive inhibition by L-amino acids. The knowledge of the surface of PLAP will enable to refine new diagnostic tools to specifically detect this marker.

s8a.m5.p5.la **Adrenodoxin/Adrenodoxin-Reductase: X-ray Structure of a Complex of two Components of the Steroidogenic Electron Transport System in Adrenal Cortex Mitochondria at 2.3 Å Resolution.** J.J. Müller[1], A. Lapko[1,2], G. Bourenkov[3], E.-Ch. Müller[4], A. Otto[4], K. Ruckpaul[5], U. Heinemann[1,#]. [1]*Forschungsgruppe Kristallographie, Max-Delbrück-Centrum für Molekulare Medizin, Robert-Rössle-Str. 10, D-13122 Berlin, Germany,* [#]*Freie Universität Berlin, Takustr. 6, D-14195 Berlin, Germany,* [2]*International Sakharow Institute of Radioecology, 2220009 Minsk, Belarus,* [3]*MPG-ASMB c/o DESY, Notkestrasse 85, D 22603 Hamburg, Germany,* [4]*Forschungsgruppe Proteinchemie, Max-Delbrück-Centrum für Molekulare Medizin, Robert-Rössle-Str. 10, D-13122 Berlin, Germany,* [5]*Max-Delbrück-Centrum für Molekulare Medizin, Robert-Rössle-Str. 10, D-13122 Berlin, Germany*
Keywords: adrenodoxin/adrenodoxin reductase complex, electron transfer, steroid biosynthesis.

The adrenal cortex mitochondrial steroid hydroxylating system consists of two soluble proteins, the NADPH-dependent flavoprotein adrenodoxin reductase (ADR), and the iron-sulfur protein adrenodoxin (ADX), and three different membrane bound cytochromes P450, which are involved in cholesterol side chain cleavage and the biosynthesis of cortisol and aldosterone. Here we present the structure of a covalently crosslinked complex of ADR and ADX with full electron transfer activity in a cytochrome c assay. The data were measured at the MPG BW6 beamline at the DORIS storage ring, DESY, Hamburg. The hexagonal crystals with cell parameters A=B=92.21Å, C=607.85Å contain two complexes in the asymmetric unit. The structure was solved by molecular replacement using the coordinates of the both components which have been determined recently [1, 2]. The final R-factor is 22.3% for 55,229 independent reflections, with rmsd of 0.007Å and 1.33 degrees from ideal bond lengths and angles, respectively. The final model of ADX contains residues 5-117 and a new C-terminal helix could be detected due to crystal contacts. Residues 5 to 460 as well as the FAD moiety have been localized for ADR. Additional to earlier biochemical characterised polar interactions between the complex components, half of the buried interacting surface is from hydrophobic side chains. Furthermore, the crosslink between ADR and ADX, induced by carbodiimide, is different from expectations and confirms analogue protein interactions as found for bacterial and plant ferredoxins. ADR undergoes conformational changes within the complex to adapt the ADX and to make possible very short electron transfer pathways between the [2FE-2S] cluster and FAD. Secondary pathways render possible electron transfer from FAD to the heme group of cytochrome c via ADX in electron transfer assays.

[1] Müller, A. *et al., Structure*, **6** (1998) 269.
[2] Ziegler, G. A. *et al., J. Mol. Biol,* **289** (1999) 981.