ESRF	**Experiment title:** MAD - experiment on electron transport protein flavin reductase from *E.Coli*.	**Experiment number:** LS - 742
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Report:

The reduction of free flavins by NADPH or NADH is not an efficient reaction. As a consequence living organisms have evolved enzymes that catalyze the reduction of riboflavin, FMN and FAD by NADPH or NADH. The NADP(H):flavin oxidoreductase from *E. coli* belong to a group of flavin reductases that utilize flavin only as a substrate and not as a coenzyme. It is part of an enzyme system that converts an inactive form of ribonucleotide reductase (RNR) into an active enzyme by convertion of the Fe(III) center of RNR to Fe(II). The flavin reductase has also been suggested to be generally involved in iron metabolism systems.

Flavin reductase has been crystallised in several different forms and we have experienced some problems in getting isomorphous crystals also within a given crystal type. A crystal from the protein that belong to the space group $P2_12_12_1$ with cell dimensions a = 51.56 b = 99.60 and c = 216.17 was used to collect data sets that could be used to solve the structure with the MAD technique. One crystal was soaked in 4 mM $KAu(CN)_2$ for 12 h and three data sets were collected at 1.3096 A (inflection point) 1.3092 A (peak) and 0.9536 A (remote). Anomalous difference Pattersons were calculated using the peak data set. This indicated two heavy atom positions that could be further refined with the program SHARP. In addition SHARP was able to detect another 8 heavy atom positions. These 10 positions were refined and the resulting phases were used to calculate a map that was fully interpretable. The position of the molecules could be defined and each asymmetric unit was shown to contain 4 molecules as could be expected from the relatively large cell volume. Solvent flattening and averaging with the program dm gave a map where about 70 % of the residues could be traced. Subsequent refinement of the structure has helped us to trace most of the sequence except for some of the loop regions and the C-terminal residue.

The structure of the flavin reductase reveals two domains one β-sandwiched domain that is suitable for the binding of flavins and one α/β NAD(P) domain. This fold supports the suggestion that this protein belongs to the same structural family as the ferredoxin/flavodoxin reductases.