Report on the use of ID 14:3, 9-10 october, 3 shifts

5-fluorouracil is one of the five-most used drugs in the chemical combat against cancer, in spite of its severe side-effects. These side effects are due to the rapid breakdown of the drug in the pyrimidine catabolic pathway. The rate limiting enzyme in this pathway is dihydropyrimine dehydrogenase (DPD) and the enzyme is a target for improvement of cancer therapy. This enzyme consists of more than 1000 amino acids per subunit. We have determined the structure to 1.9 Å resolution based on MAD phasing using the anomolous signal of Fe due to the presence of four Fe-S clusters, with data collected at the ESRF (Dobritzsch et al., 2001). The experiment performed during this visit extended the study to the analysis of an enzyme-inhibitor complex in order to provide mechanistic insights.

* ternary complex of DPD / 5-iodouracil / NADPH obtained by cocrystallization

pH of the solution was changed from 4.7 (citrate) to 7.5 (HEPES) by 25 min soaking in mother liquor with appropriate buffer system;
wavelength: 0.93106 Å, distance: 180 mm, $\Delta\varphi$: 0.5°, images: 1-330, detector: MAR-CCD

resolution	30-2.25 Å	(2.37-2.25 Å)	
completeness	98.3 %	(97.4 %)	
R_{sym}	0.064	(0.250)	
I/σ	8.3	(2.3)	SCALA
multiplicity	3.6	(3.4)	

results:

- change in unit cell dimensions, most likely due to pH-change;
- 5-iodouracil is bound in active site, but there is very weak density for the iodine atom: cleaved off?
- active site loop is closed and density is present for all loop-residues;
- NADPH is bound close to FAD in a conformation suitable for hydride transfer