SNBL	**Experiment title**: Study of ferryl myoglobin as model for high valent activated oxygen complexes of heme proteins and a cytochrome c with unusual EPR spectrum (+ malat dehydrogenase)	**Experiment number**: 01-02-349 (01-02-350)
Beamline: BM01	**Date of experiment**: from: 08-Mar-02 07:00 to: 10-Mar-02 07:00	**Date of report**: 30-May-02
Shifts: 6	**Local contact(s)**: Jon Are BEUKES	*Received at UNIL:*

Names and affiliations of applicants (* indicates experimentalists):

Prof. K. Kristoffer Andersson, Department of Biochemistry, Univ. of Oslo, Norway

Prof. Carl Henrik Gørbitz, Department of Chemistry, Univ. of Oslo, Norway

Dr. Bjørn Dalhus, Department of Chemistry, Univ. of Oslo, Norway*

Cand.scient Hans-Petter Hersleth, Department of Chemistry, Univ. of Oslo, Norway*

Investigation of myoglobin:

The myoglobin project was this time further developed:

We have previous let myoglobin react with hydrogen peroxide or other organic peroxides at different pH-values. We have in these studies trapped the intermediate called compound II, and looked on the pH dependence. This time we let myoglobin react with hydrogen peroxide making the compound II, and let then this intermediate oxidize other substances.

By making two reactions in the crystals they can very easily be destroyed, but we were able to get two data sets on reaction with glutanthione and L-cysteine. The crystals have not as high quality as we are used to, but very acceptable. We had hoped to be able to see the glutanthione and cysteine in the reaction site, but unfortunately this was not the case (when we solved the structures). The compound II was however reduced, so the reaction had occurred.

Data sets were:

Rection with H_2O_2, and then gluthantione (G) or L-cysteine (C)

(G) Resolution: 25-1.45 Å Rmerge: 4.5% Completeness: 94.2% (scalepack)

(C) Resolution: 25-1.70 Å Rmerge: 5.9% Completeness: 82.2% (scalepack)

Investigation of a double-site mutant of malat dehydrogenase (MDH) from the thermophilic bacteria Chloroflexus Aurantiacus.

A single datasett of a double-mutant (E165Q-T187C) of malate dehydrogenase (MDH) from the green gliding termophilic bacteria Chloroflexus Aurantiacus was collected to approximately 1.9 Å resolution. The crystal mosaicity seems high, resulting in low completeness during data processing with Denzo. Nevertheless, the data are hopefully sufficient to investigate the structural consequences on the tetrameric stabilization by the introduced mutations. The structure has not yet been solved, but should be a routine operation once the processing is finished.

Another crystal of the same protein was also subject to data collection in order to increase the completness of the first data set. Unfortunately there were some problems with the beam collimator at that time and the data collection was aborted without recovery of the crystal.

Related Publications in this periode:

Hans-Petter Hersleth, Bjørn Dalhus, Carl Henrik Görbitz & K. Kristoffer Andersson, (2002). An iron hydroxide moiety in the 1.35 Å resolution structure of hydrogen peroxide derived myoglobin compound II at pH 5.2. *J. Biol. Inorg. Chem.*, **7**, 299-304.

Bjørn Dalhus, Markuu Saarinen, Uwe H. Sauer, Pär Eklund, Kenth Johansson, Andreas Karlsson, S. Ramaswamy, Alexandra Bjørk, Bjørnar Synstad, Kristine Naterstad, Reidun Sirevåg, & Hans Eklund (2002). Structural Basis for Thermophilic Protein Stability: Structures of Thermophilic and Mesophilic Malate Dehydrogenases *J. Mol. Biol.*, **318**, 707-721