SN BL	Experiment title : Studies of the R2 subunit with Co(II) and Mn(II) of Ribonucleotide reductase (RNR) from mouse	Experiment number: 01-02-350 (01-02-349)
Beamline: BM01	Date of experiment: from: 10-Mar-02 07:00 to: 11-Mar-02 07:00	Date of report: 30-May-02
Shifts: 3	Local contact(s): Jon Are BEUKES	Received at UNIL:

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Investigation of the ribonucleotide reductase (RNR) R2 subunit from mouse:

The RNR R2 project was this time further developed:

In previous experiments we have investigated R2 from mouse substituted with Co(II) (1) and Mn(II) and have got the first crystal structures of fully occupied dinuclear sites of R2 RNR at pH 6. This time we have been working on R2 substituted with Fe(II), the natural metal ion in the dinuclear cluster of R2. The apo-R2 crystals were treated (soaking) with 5 mM Fe²⁺ and 0.5 % sodiumdithionite for about 4 hours. Phenosafranine was added in order to monitor reducing (yellow coloured crystals) or oxidising (red coloured crystals) conditions. One data set at cryo temperature was collected on a reduced Fe-R2 crystal (yellow colour, data set 1) and another data set was collected on a Fe-R2 crystal oxidised in air (red colour, data set 2). We have also collected data on Fe-R2 crystals oxidised with 44 mM H₂O₂ (data set 3) and treated with methanol (data set 4).

The data sets analysis:

Treatment with Fe²⁺, dithionite and phenosafranine (reduced complex):
Resolution: 30-2.0 Å Rmerge: 5.2% Completeness: 92.1.2% (scalepack)
Treatment with Fe²⁺, dithionite, phenosafranine, and then oxidation in air:
Resolution: 30-2.1 Å Rmerge: 6.9% Completeness: 87.9.2% (scalepack)
Treatment with Fe²⁺ and ascorbate, and then oxidation with H₂O₂:
Resolution: 30-2.4 Å Rmerge: 10.1% Completeness: 95.3% (scalepack)
Treatment with Fe²⁺, dithionite, phenosafranine, and then air and 10% methanol
Resolution: 30-2.1 Å Rmerge: 5.6% Completeness: 93.3% (scalepack)

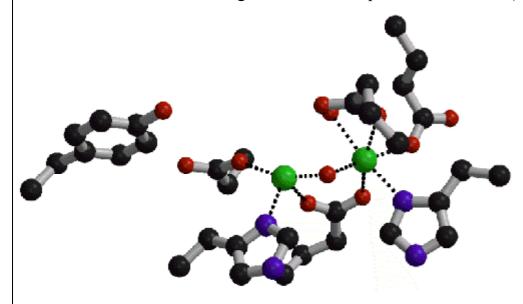


Figure 1: The diferric cluster in R2 RNR from mouse. The iron ions are seen in green.

Published in this period:

- 1. Strand, K. R., Karlsen, S. & Andersson, K.K. Spectroscopic and structural evidence for a ferromagnetic coupled dinuclear cobalt cluster in a cobalt substituted R2 from mouse ribonucleotide reductase, *J. Biol. Chem.* (submitted) (2002)
- 2. Karlsen, S., K.R. Strand, C.H. Gørbitz and K. K. Andersson (2002). Structural investigation of reactions in the dinuclear R2 subunit from mouse Ribonucleotide Reductase. *The 38th Norwegian Biochemical Society Meeting at Rørås*, January 17-20, p52