



Experiment title: X-Ray Structure Analysis of the Membrane Protein Complex Fumarate Reductase from *Wollinella succinogenes*

Experiment number:
LS-426

Beamline:
D14-BL19

Date of experiment:

from: 06-FEB-96 to: 08-FEB-96

Date of report:
23 February 1996

Shifts:
6

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Received at ESRF:
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Report:

Fumarate reductase (menaquinol:fumarate oxidoreductase) from *Wollinella succinogenes* consists of three subunits, FrdA, FrdB, and FrdC, with a total molecular weight of 130 kDa. FrdC (30 kDa) is a diheme cytochrome *b*, which anchors the enzyme in the membrane and contains the site for menaquinol oxidation. FrdA (73 kDa) contains covalently bound FAD, a trinuclear [3Fe-4S] iron sulfur centre, and carries the site of fumarate reduction. FrdB (27 kDa) contains binuclear [2Fe-2S] and tetranuclear [4Fe-4S] iron-sulfur centres. This enzyme is currently the best investigated system involved in anaerobic respiration.

The protein has been crystallized in the monoclinic space group P2₁, with most probably two complexes in the asymmetric unit. The cell parameters were determined to be $a = 87 \text{ \AA}$, $b = 190 \text{ \AA}$, $c = 119 \text{ \AA}$, with $\alpha = \gamma = 90^\circ$, and $\beta = 104.6^\circ$. These crystals diffract X-rays from a rotating Cu anode source to beyond 2.3 \AA resolution.

During the beam time allocated to LS-426, approximately 5000.5° oscillation images (including test exposures) to high resolution limits of 3.2 Å (top edge of detector) and 2.7 Å (bottom edge of detector) were collected from 40 crystals. The acquisition of this large number of exposures was only possible because of the availability of the CCD detector at D14/BL19, thus reducing the reading-out time to a minimum. Whenever possible, crystals were translated up to two times after the recording of 10-15 images each, because of deterioration of the diffraction pattern due to radiation damage.

Preliminary processing of 151 of the 208 images recorded on native crystals using the HKL processing package (Otwinowski and Minor, to be published) has resulted in a partial data set of 63,646 measured reflections between 99 Å and 3.1 Å (38,423 unique, 55.2% complete), with R_{sym} for the intensities of symmetry-related reflections at 9.7%. Processing of some of the images is complicatedly the fact that some of the crystals were not single.

Processing of 153 images from putative uranyl derivatives and 100 images from possible lead derivatives is currently in progress.