ESRF	**Experiment title:** Fumarate reductase, a membrane protein complex from *Wollinella succinogenes*	**Experiment number:** LS-802
Beamline: BM14	**Date of experiment:** from: 16-Dec-97 7:00 to: 18-Dec-97 7:00	**Date of report:** 23-Feb-98
Shifts: 6	**Local contact(s):** Gordon Leonard	*Received at ESRF:* 02 MAR. 1998

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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 dihaem 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.

Crystals of this bioenergetically important 130 kDa membrane protein complex diffract to beyond 2 Å resolution and have two different unit cells, both of the monoclinic space group $P2_1$. The unit cell of crystal form "A" is a = 87 A, b = 190 A, c = 119 A, with $\beta = 104.6°$, the unit cell of form "B" is a = 119 A, b = 85 A, c = 190 A, with $\beta = 96.4°$. Assuming a solvent content of 65%, there are four complexes per unit cell and thus two complexes in the asymmetric units of both unit cells. However, many crystals contain both unit cells, and single crystals of both unit cells are not distinguishable morphologically. This makes data collection from several crystals (beam-)time-consuming, but still feasible.

The first nine hours of beam time allocated to LS-802 could not be used due to problems with the calibration of the CCD detector and subsequent setting up of the MAR345 detector as an alternative. Even in the small diameter (180 mm) larger pixel (150 μm) mode, reading-out time was 53 seconds compared to exposure times of 15-30 seconds. This caused us to abandon the original plan of a two-wavelength experiment on a putative derivative. Despite attempting to collect derivative data from a number of crystals, the only potentially useful data were collected from a single crystal (grown with an improved procedure) which displayed an effective mosaicity of 0.9° (which is comparatively low in this system), and which remained intact in the beam for an exceptionally long time, thus allowing 184 images to be merged to a data set with 288,091 measured reflections between 30.0 and 2.80 Å. The number of unique reflections was 160,053, which corresponds to a completeness of 90.0% with R_{sym} for the intensities of symmetry-related reflections at 7.3% ($I/\sigma(I) = 10.9$). Although this was a crystal of a mutant protein which was designed to have an additional heavy atom binding site, no additional binding site could be deduced from the isomorphous difference Patterson maps.