ESRF	**Experiment title:** Fumarate reductase, a membrane protein complex from *Wolinella succinogenes*	**Experiment number:** LS-1137
Beamline: BM14	**Date of experiment:** from: 10-Nov-98 15:00 to: 13-Nov-98 15:00	**Date of report:** 23-Feb-99
Shifts: 9	**Local contact(s):** Gordon Leonard	*Received at ESRF:* 26 FEV. 1999

Names and affiliations of applicants (* indicates experimentalists):

C. Roy D. Lancaster* and Hartmut Michel,

Max-Planck-Institut für Biophysik,
Abteilung Molekulare Membranbiologie,
Heinrich-Hoffmann-Str. 7,
D-60528 Frankfurt am Main, Germany

Report:

Fumarate reductase (menaquinol:fumarate oxidoreductase) from *Wolinella 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 and carries the site of fumarate reduction. FrdB (27 kDa) contains a binuclear [2Fe-2S], a trinuclear [3Fe-4S], and a tetranuclear [4Fe-4S] iron sulfur centre. This enzyme is currently the best investigated system involved in anaerobic respiration.

Crystals of this bioenergetically important 130 kDa membrane protein have two different unit cells, both of the monoclinic space group $P2_1$. The unit cell of crystal form "A" is a = 85 Å, b = 190 Å, c = 119 Å, with β =104.6°, the unit cell of form "B" is a = 119 Å, b = 85 Å, c = 190 Å, with β = 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 units and single crystals of both unit cells are not distinguishable morphologically.

During the beam time allocated to LS-1137, nine data sets could be collected, requiring only one crystal per data set (cf. Table 1).

Table 1. Data collection and processing statistics of LS-1137 at ESRF BM14

Xtal form	data set	WT/mutant	wavelength	resol	hkl_{meas}	hkl_{unique}	complete	R_{sym}
A	Hg1	mut1	0.9999 Å	3.2 Å	387,708	58,864	99.9%	7.0%
B	Hg2	mut1	0.9999 Å	3.2 Å	226,488	61,688	99.9%	8.1%
A	Hg3	mut2	0.9999 Å	3.2 Å	329,636	58,621	99.8%	8.6%
A	Ir1	WT	1.105 Å	3.2 Å	220,988	58,879	99.9%	7.3%
A	Ir2	WT	1.105 Å	3.2 Å	233,083	58,392	99.8%	6.6%
A	U1	WT	0.722 Å	3.2 Å	199,052	58,463	99.8%	5.4%
A	U2	WT	1.000 Å	2.6 Å	389,519	109,204	99.9%	6.9%
A	Hg4	WT	1.000 Å	2.6 Å	384,736	108,731	99.1%	6.5%
A	Pb1	WT	0.9464 Å	2.9 Å	262,397	77,793	97.8%	8.6%

For this project, this was the first time that we were able to collect complete data sets on individual crystals. This is attributed to both improvements in crystal quality and optimum measurement conditions at the beamline. The latter included the availability of a marCCD detector, an FTS cooling system, and operation of the ring in 16-bunch mode. Unfortunately, the collected data sets are only partly useful for MIRAS phasing, due to lack of isomorphism and occasional low phasing power. A drawback of the rapid data collection procedure was that we were only able to process 35-40% of the data before data collection was complete, so *ad hoc* decision making was limited.