ESRF	**Experiment title:** Frankfurt BAG subproject: Fumarate reductase, a membrane protein complex from *Wolinella succinogenes*	**Experiment number:** LS-1514
Beamline: BM14	**Date of experiment:** from: 1-Oct-99 8:00 to: 2-Oct-99 7:00	**Date of report:** 29-Feb-2000
Shifts: 3	**Local contact(s):** Edward Mitchell	*Received at ESRF:*

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Report:

Fumarate reductase, a bacterial version of complex II from the respiratory chain, catalyses the reduction of fumarate to succinate, in a reaction opposite to that catalysed by complex II (succinate dehydrogenase). Fumarate reductase (menaquinol:fumarate oxidoreductase) from the anaerobic bacterium *Wolinella succinogenes* consists of three protein 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 andd contains the site for menaquinol oxidation. FrdA (73kDa) 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-sulphur centre. This enzyme is currently the best investigated system involved in anaerobic respiration.

Crystals of this bioenergetically important 130 kDa membrane protein complex diffract up to at least 2.1 Å and have two different unit cells, both of the monoclinic space group $P2_1$. The unit cell of crystal form "A" is a = 85.2 Å, b = 189.0 Å, c = 117.9 Å, and $\beta = 104.5°$. Crystal form "B" has the unit cell dimensions a = 118.4 Å, b = 85.1 Å, c = 188.9 Å, $\beta = 96.5°$. 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. Using data collected earlier at ESRF BM14 (cf. experimental reports for LS-1369), the structure of crystal form A has been solved by multiple isomorphous replacement and anomalous scattering (MIRAS) and refined to 2.2 Å resolution, and that of crystal form B has been solved by molecular replacement (MR) and refined to 2.33 Å resolution.

During the beam time allocated to the fumarate reductase project of LS-1514 in October, only one medium quality data set (for crystal form B) could be collected from a single crystal:

Table: Diffraction data collected at ESRF BM14 (1-2 October 1999)

	resol. range [Å]	measured reflections	unique reflections	complete [%]	I/σ(I)	>2σ [%]	R_{sym}* [%]
substrate complex							
proj6/306a503_1	15.0-2.75	250,787	86,864	89.2	11.3	67.7	8.0
	2.81-2.75	*16,350*	*5888*	*91.9*	*2.3*	*43.1*	*38.8*

This disappointing result was due to the insufficient quality and quantity of the crystals at the time. The situation has since improved (cf. LS1514@BM14 report for Dec 1999), although still a large number of crystals have to be screened prior to the collection of full data sets.