



ESRF

**Experiment title:**  
**Crystallographic studies of pyruvate:ferredoxin  
oxidoreductase (PFOR) from *Desulfovibrio africanus***

**Experiment  
number:**  
LS-748

**Beamline:**  
BM14

**Date of experiment:**  
f r o m : 23Sept97 t o : 25Sept.97

**Date of report:**  
5Nov.97

**Shifts:**  
9

**Local contact(s):**  
Andy Thompson

*Received at ESRF:*  
13/11/97

**Names and affiliations of applicants** (\* indicates experimentalists):

\*Charon Marie-Helene      IBS/LCCP - CEA  
\*Chabriere Eric            IBS/LCCP - PhD student  
\*Volbeda Anne             IBS/LCCP - CEA  
Fontecilla Juan             IBSLCCP - CEA

---

### **Preliminary report:**

During the beam time allocated at the end of September 97 for our project we have been able to collect 4 data sets :

2 data sets concern crystals of the native enzyme, 1 data set concerns a complex between the PFOR and pyruvate which is one of the reactional substrates, and the fourth data set concerns a complexe between PFOR and acetylcoenzymeA, the latter becoming substrate of the enzyme in the inverse reaction. As the four crystal were prepared in the presence of air, the enzyme is always in an oxidized state.

As the analyses of the data sets are underway, we can mention today only preliminary results

- the first native cristal diffracted up to 2.3Å. The cell parameters are a=86.6, b=146.5 and c=203.9, and correspond to the crystal form 1 of POR (space group P212121). We hope to obtain a good completeness with this data set. The mosaicity is 0.3 but the Rfactor obtained with the integration of the first 30 degrees was 12.6%.

- the second native crystal diffracted at high resolution (up to 1.95Å) but was very mosaic (around 1.0). We have been able to find the cell parameters ( P212121, a=86.5, b=149.1, c=212.4, crystal form 2) but we do not know yet if we can integrate the data and the set should be only 50% complete.

- the pyruvate-PFOR crystal gave diffraction up to 2.6Å with a=85.6 b=146.0 and c= 210.3 (probably crystal form 1). Integration of the first 20 degrees gave Rfactor=7.6%.
- the acetylcoA-PFOR crystal diffracted up to 3.2Å with a=85.6, b=146.3 and c=204.2Å (crystal form 1). Integration of the first 25degrees gave Rfactor=9.8%.

After complete treatment of the data we hope to find out the binding mode of the substrates pyruvate and acetylcoA to the enzyme.

Secondly, we intend to collect new data with crystals obtained under reducing and anaerobic conditions ( in a glove box) in order to obtain more information on the catalytic mechanism.