



	Experiment title: <b>A number of Proteins from Bacteria to Eukarya and from Antarctic to Volcanic Areas</b>	<b>Experiment number:</b> LS-2199
<b>Beamline :</b> ID 14-4	<b>Date of experiment:</b> from: 6 April to: 8 April. 2002	<b>Date of report:</b>  26/02/03 <i>Received at ESRF:</i>
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### Report:

Four complete data sets for the *Sulfolobus solfataricus* alcohol dehydrogenase system (SsADH) have been registered. In particular, X-ray data has been collected on a tetragonal crystal of (N249Y)SsADH. This mutant exhibits a higher specific activity compared to the wt-enzyme as well as a higher dissociation constant for the NAD(H) cofactor.

X-ray data has also been registered on orthorhombic forms of putative ternary complexes of (N249Y)SsADH-NADH and substrate analogues (DMSO dimethyl sulfoxide; IBA isobutiramide). Due to the low resolution of the in-house datasets collected on these crystals it was not possible to check the cofactor binding. However, the analysis of the electron density maps derived from the synchrotron data showed that the crystals contain the apoform of the molecule.

Finally, diffraction data were registered for a monoclinic form of wild-type SsADH complexed with NAD(H) and 3-fluoroethanol (TFE). Although this crystal turned out to be non-merohedrally twinned, data from the dominant lattice could be reduced and processed. The solution of the structure revealed that the substrate site contains a 2-ethoxyethanol molecule used as precipitant instead of a 3-fluoroethanol molecule.

## Data collection report

Mutant (N249Y)SsADH apoform

Space group	I4 <sub>1</sub> 22
Cell dimensions (Å, °)	a=b=125.40; c=117.19
Resolution (Å)	25.-2.5 (2.59-2.50)
Total No. of reflections	104158
No.of independent reflections	15757
Completeness (%)	95.0 (96.9)
Rsym(I) (%)	3.6 (8.8)
Mosaicity (°)	0.7

Putative complex (N249Y)SsADH-NADH-DMSO

Space group	P4 <sub>2</sub> 2 <sub>1</sub> 2
Cell dimensions (Å, °)	a=b=88.28 c=86.12
Resolution (Å)	25.-2.5 (2.59-2.50)
Total No. of reflections	83462
No.of independent reflections	12361
Completeness (%)	99.7 (100.)
Rsym(I) (%)	5.1 (12.4)
Mosaicity (°)	0.7

Putative complex (N249Y)SsADH-NADH-IBA

Space group	P4 <sub>2</sub> 2 <sub>1</sub> 2
Cell dimensions (Å, °)	a=b=88.43 c=86.87
Resolution (Å)	25.-2.85 (2.95-2.85)
Total No. of reflections	117688
No.of independent reflections	8521
Completeness (%)	100. (100.)
Rsym(I) (%)	6.0 (11.8)
Mosaicity (°)	0.4

Complex wt-SsADH-NAD(H)-2ethoxyethanol

Space group	C2
Cell dimensions (Å, °)	a=131.06 b=83.01 c=68.67, β=96.5
Resolution (Å)	25.-2.3 (2.3-2.38)
Total No. of reflections	70105
No.of independent reflections	29287
Completeness (%)	89.5 (60.8)
Rsym(I) (%)	5.8 (7.1)

