



	Experiment title: Structural studies on NMN adenylyltransferase from <i>Methanococcus jannaschii</i>	Experiment number: LS-1664
Beamline: ID14-1	Date of experiment: from: 16.06.2000 to:19.06.2000	Date of report: 1 August 2000
Shifts: 9 to BAG	Local contact(s): B. Rasmussen	<i>Received at ESRF:</i>
Names and affiliations of applicants (* indicates experimentalists): Igor D'Angelo, Menico Rizzi Dept. Genetics and Microbiology University of Pavia Via Abbiategrasso 207 27100 Pavia Italy		

Within a framework aimed to the structural characterization of enzymes involved in NAD biosynthesis we have recently solved the structure of NMN adenylyltransferase (NMN-AT) from *M. jannaschii* in complex with ATP (D'Angelo et al. 2000, in press). In order to shed light on the reaction mechanism adopted by NMN-AT the structure determination of NMN-AT in complex with nicotinamide mononucleotide and NAD have been carried out. Moreover, a data collection on a complex with tiazofurin (a substrate analogue) has also been performed. Crystals of *M. jannaschii* NMN-AT belong to space group $P2_1$ with unit cell parameters $a=79.5 \text{ \AA}$ $b=112.4 \text{ \AA}$ $c=82.1 \text{ \AA}$ and $\beta=117.8^\circ$. The asymmetric unit contains an homoexamer having 32 point symmetry. We have measured three data sets (complexes with NMN, NAD and tiazofurin) at 2.5 \AA resolution with the following statistics:

NMN complex

N° measurements=103,115

N° unique reflection=41,246

Completeness=96%

Multiplicity=2.5

Rsym=5.5 (highest resolution shell 24.4%).

NAD complex

N° measurements=83,550

N° unique reflection=41,775

Completeness=97.1%

Multiplicity=2.0

Rsym=6.5 (highest resolution shell 21

Tiazofurin complex

N° measurements=98,880

N° unique reflection=39,552

Completeness=95.1%

Multiplicity=2.5

Rsym=4.5 (highest resolution shell 28.4%).

The ligands (NMN, NAD and tiazofurin) have been located through difference Fourier and the structures refined. We are now able to describe the entire active site and to assess the role played by several protein residues in catalysis and substrate/product recognition.

Reference

1.) D'Angelo, N. Raffaelli, V. Dabusti, T. Lorenzi, G. Magni and M. Rizzi.

Structure of nicotinamide mononucleotide adenylyltransferase: a key enzyme in NAD⁺ biosynthesis

Structure with Fold. Des., 2000, in press