



Experiment title:
STRUCTURE *Haemophilus influenzae* NadN

Experiment number:
MX394

Beamline:
ID29

Date of experiment:
from:14/07/05 to:15/07/05

Date of report:
22th July 2005

Shifts:
3

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REPORT

Background – *Haemophilus influenzae* has an absolute requirement of NAD for growth, because it lacks all but one of the NAD biosynthetic enzymes. Since NAD cannot pass through the membrane, a set of periplasmic enzymes transform NAD into either NMN or nicotinamide riboside which are then assumed to diffuse across the inner membrane. NadN is a periplasmic enzyme endowed with both NAD pyrophosphatase and NMN 5'-nucleotidase activities and has been regarded as a sensible target for novel specific antibiotics. Moreover since NadN is antigenically highly conserved among typeable and nontypeable *H. influenzae*, it is of great interest as a promising candidate for a new vaccine against both *H. influenzae* strains. We recently obtained crystals suitable for X-ray structure determination which belong to the C2221 orthorhombic space group and diffract to 1.5 Å resolution at ID14 EH2.

Results – We have been so far unable to successfully produce the Se-Met form of the enzyme, due to the severe formation of inclusion bodies and to our failure in standard refolding procedures. Therefore we are

attempting structure determination by MIR and by SAD at the Sulfur edge. 20 different data sets have been collected at resolution around 2.0 Å at the anomalous peak of the respective heavy atom. A complete data set, with very high redundancy has been collected up to 2.7 Å resolution at a wavelength of 1.77 Å, for maximizing the Sulfur anomalous signal. A difference Patterson has been solved in the case of the Xe derivative and the subsequent calculated phases clearly allowed to start tracing. We expect the structure to be completed in the near future.