	Experiment title: BAG-LEBS-2005-2	Experiment number: MX-441
Beamline: ID23-EH1	**Date of experiment**: from: 18/11/2005 8h to: 19/11/2005 8h	**Date of report**: 24/02/05
Shifts: 3	**Local contact(s)**: Dr E. Micossi.	*Received at ESRF:*

Names and affiliations of applicants (* indicates experimentalists):

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Report:

Hélène Walbott, Béatrice Golinelli (0.5 shift) : Structural study of tRNA methyltransferase from *P. abyssi* (pam5U project)

During this session, we have collected a dataset at 2.5A resolution (space group C2, completion: 100%) from a crystal of the native protein, but we were not able to solve the structure by molecular replacement. So we have decided to produce the SeMet labelled protein in order to conduct MAD experiments next time.

Vincent CHAPTAL, Philippe MEYER (1 shift): *B. subtilis* nitroreducatse in complex with cofactor and inhibitor

Data collected on *B. subtilis* nitroreducatse in complex with cofactor and inhibitor to a resolution of 2.2 Å. The structure has since then been solved by MR with a 22% identity model and is currently under manual rebuilding. Test of various crystal form of molecular chaperon HtpG and a nuleotide kinase with poor resolution.

Mark BROOKS and Nicolas LEULLIOT (1.5 shifts): yeast *Saccharomyces cerevisiae* Structural Genomics project

The systematic names of the genes are used. More details on every orf can be found on http://genomics.eu.org:targets.html

1) Yeast YGL047w or Alg13.

This essential yeast protein is required for the second step of dolichyl-linked oligosaccharide synthesis and is involved in N-linked glycosylation. It presents similarity to bacterial glycosyltransferases. We have tried to co-crystallise this protein with different sugars and crystals appeared in the presence of UDP-galactose. During this session, several crystals have been tested but the resolution was too low to collect any datasets.

2) *P. abyssi Pab1159 protein.*

Spacegroup $P2_12_12$ a=82A; b=90A;c=230A.
Resolution 2.9A
Completion: 100 %.
Rsym=14.8%.

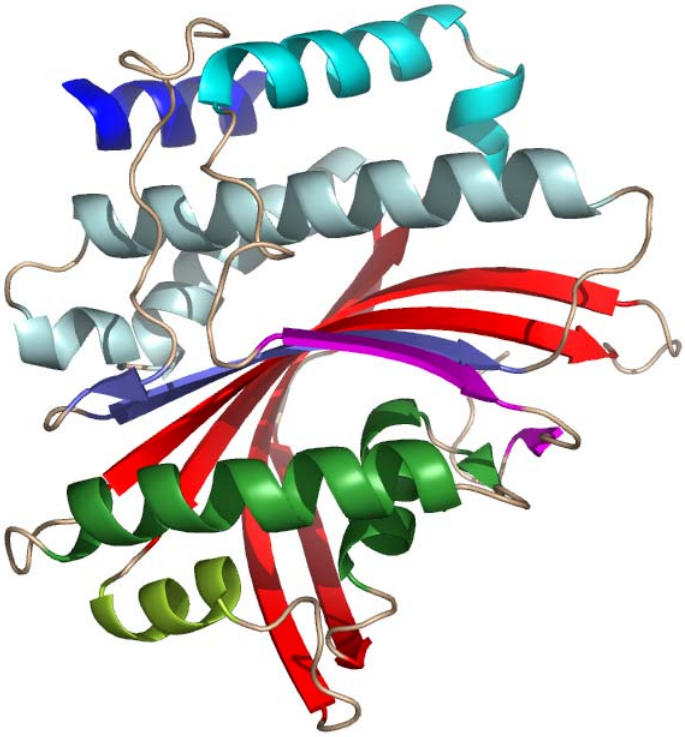
This protein of unknown function is conserved among the three kingdoms of life. From sequence analysis, it is predicted to be an endosialopeptidase. In order to get information on its function, we have purified and crystallized this protein from the archae *Pyrococcus abyssi*. We have previously collected a complete native dataset to 1.5A resolution. During this session, we have collected a MAD dataset (with a 360° highly redundant peak) at the tungstate edge from a crystal obtained by co-crystallization with $NaWO_4$. This allowed us to solve the structure of this protein and to refine it against a 1.5A dataset previously collected on ID14-EH1.

3) EVF protein.

Spacegroup $P2_12_12_1$ a=69A, b=86A; c=91A.
Resolution 2.2A.
Completion 97.8%
Rsym 8.9%

This 31kDa protein from the bacterium *Erwinia carotovora* is a virulence factor that affects *Drosophila melanogaster*. Nothing is known about the virulence mechanism of this protein. In collaboration with B. Lemaitre (CGM, CNRS, Gif/Yvette), we have undertaken the determination of the crystal structure of this protein to try to get insights into the function of this protein. During this run, we have collected a 2.2A MAD dataset at the Se edge as well a 2A native dataset. Although this 277 residues long protein possesses only 2 Met, the collected data allowed us to get experimental phases of excellent quality, which can be used by the program Arp/wArp to rebuild more than 90% of the model. The resolution of this structure has revealed the presence of a lipid covalently linked to a cysteine residue, hence allowing new assumption to be tested to unravel the molecular basis of the virulent effect of this protein.

Ribbon representation of the structure of the EVF protein.



4) Virar9.
Spacegroup $P2_12_12_1$ a=75A, b=129A; c=224A.
Resolution 3 A.
Completion 92
Rsym 13.2%

This small protein (14kDa) of unknown function is encoded by an archaeophage. We have collected a native dataset to 3A during this session. However, experimental phases are needed to solve this structure. SeMet labelling has been done after the diffraction was observed during this run and crystallization of this SeMet