	Experiment title: BAG-LEBS-2007-1	Experiment number: 30-01-796
Beamline: BM30A	**Date of experiment**: from: Saturday 01/12/2007 at 8:30 to: Monday 03/12/2007 at 8:00	**Date of report**: 28/2/07
Shifts: 6	**Local contact(s)**: Dr. L. Jacquamet	*Received at ESRF:*

Names and affiliations of applicants (* indicates experimentalists):

Jenny Keller* (PhD Student) and Julien Henri* (PhD Student): Herman Van Tilbeurgh's Group, Institut de Biochimie et de Biophysique Moléculaire et Cellulaire (IBBMC), Equipe Génomique Structurale, CNRS UMR8619, Université Paris-Sud, Orsay, France

Jean-Christophe Zeeh*, LEBS, Bat 34, CNRS UPR9063, 1 av. de la Terrasse, Gif-Sur-Yvette, France

Report:

Jenny Keller* and Julien Henri* (3 shifts) : yeast multi-protein complexes involved in DNA replication, ribosome biogenesis, mRNA quality control pathway and cell signalling and archeophage structural genomics project

1) Tpa1

Spacegroup: $P2_12_12_1$ with a= 105 Å, b=160 Å and c=210Å.
Resolution: 3.50A.
Completion= 98.4%
Rsym=17%

The deletion of the gene encoding for yeast Tpa1 protein strongly affects translation termination, deadenylation and mRNA stability, suggesting a role in the control of gene expression at the level of translation. The Tpa1 protein is a component of a ribonucleotidic complex bound to the 3'-end of mRNAs. The knowledge of its 3D structure might help to decipher the precise function of Tpa1. Crystals of native Tpa1 have been grown and diffract to 3.2 Å. During this session, we have tested a 3-wavelength MAD dataset from crystals grown from SeMet-labelled protein. The resolution reached 3.5A but the crystal suffer from serious diffraction anisotropy. The anomalous signal was too weak to allow experimental phases to be obtained.

J.C. Zeeh (3 shifts): structural studies of complexes between guanine exchange factors and Arf small GTP-binding proteins with inhibitors

1) EKLM11

ARF1 is a small G protein and involved in vesicular transport in cell. In 2003, we obtained the structure of Arf1 complexed with its regulator ARNO (a guanine exchange factor). By in silico screening, we found a new inhibitor of exchange reaction of Arf1 catalysed by ARNO, called LM11. The mechanism of this inhibitor is known and LM11 binds to the complex Arf1/ARNO, so we try to obtain a crystal of ARF1/ARNO with LM11 by soaking ARF1/ARNO in LM11 solution.
During the session, I tested 1 crystal but the crystal gave no diffraction maybe due to a bad cryoprotection step.

2) SECIN

ARNO is a protein which regulate the function of Arf1 in cell. M.Famulok, by screening small molecules,found a new inhibitor of ARNO called SecinH3. He showed that this inhibitor is specific to ARNO in vitro and in vivo binds ARNO. So we try to obtain a crystal of ARNO with SecinH3 by soaking ARNO in SecinH3 solution, or by cocrystallization of ARNO and SecinH3.

During this session, 23 crystals were tested. But just two gave a good diffraction pattern for molecular remplacement.

First crystal :

Spacegroup: P213 with a= 88 Å, b=88 Å and c=88 Å.
Resolution: 2.5 A.
Completion= 99.8%
Rsym=24%

Second crystal :

Spacegroup: P213 with a= 88 Å, b=88 Å and c=88 Å.
Resolution: 2.5 A.
Completion= 99.9%
Rsym=22%

Unfortunately, The structure analysis showed no density which could be correlated with the inhibitor SecinH3.