# EUROPEAN SYNCHROTRON RADIATION FACILITY

INSTALLATION EUROPEENNE DE RAYONNEMENT SYNCHROTRON



# **Experiment Report Form**

The double page inside this form is to be filled in by all users or groups of users who have had access to beam time for measurements at the ESRF.

Once completed, the report should be submitted electronically to the User Office using the **Electronic Report Submission Application:** 

http://193.49.43.2:8080/smis/servlet/UserUtils?start

# Reports supporting requests for additional beam time

Reports can now be submitted independently of new proposals – it is necessary simply to indicate the number of the report(s) supporting a new proposal on the proposal form.

The Review Committees reserve the right to reject new proposals from groups who have not reported on the use of beam time allocated previously.

### Reports on experiments relating to long term projects

Proposers awarded beam time for a long term project are required to submit an interim report at the end of each year, irrespective of the number of shifts of beam time they have used.

#### Published papers

All users must give proper credit to ESRF staff members and proper mention to ESRF facilities which were essential for the results described in any ensuing publication. Further, they are obliged to send to the Joint ESRF/ ILL library the complete reference and the abstract of all papers appearing in print, and resulting from the use of the ESRF.

Should you wish to make more general comments on the experiment, please note them on the User Evaluation Form, and send both the Report and the Evaluation Form to the User Office.

# **Deadlines for submission of Experimental Reports**

- 1st March for experiments carried out up until June of the previous year;
- 1st September for experiments carried out up until January of the same year.

# **Instructions for preparing your Report**

- fill in a separate form for each project or series of measurements.
- type your report, in English.
- include the reference number of the proposal to which the report refers.
- make sure that the text, tables and figures fit into the space available.
- if your work is published or is in press, you may prefer to paste in the abstract, and add full reference details. If the abstract is in a language other than English, please include an English translation.

| ESRF   | Experiment title:  Archaeal signal recognition particle | Experiment<br>number:<br>MX-230 |
|--|---|---------------------------------|
| Beamline: ID23-1   | Date of experiment: from: 070504 to: 080504             | Date of report: 010305          |
| Shifts:  | Local contact(s): Edward Mitchell                       | Received at ESRF:               |
| Names and affiliations of applicants (* indicates experimentalists):  Tobias Hainzl Shenghua Huang |   |                                 |
| UCMP, Um   | neå University<br>eå, Sweden                            |                                 |

# **Report:**

How proteins become properly localised within cells is one of the fundamental questions in biology. Protein sorting is the principal process by which order is achieved and maintained in all living cells. A key player in this process is a ribonucleoprotein, the signal recognition particle (SRP), which functions as a molecular adapter coupling protein synthesis and membrane translocation. The signal recognition particle (SRP) is an evolutionary conserved ribonucleoprotein (RNP) that associates with ribosomes to mediate co-translational targeting of secretory proteins to biological membranes.

In mammalian cells, the SRP consists of a 7S (or SRP) RNA, and six protein components (SRP9/14, SRP19, SRP54 and SRP68/72). SRP can be divided into two structurally and functionally distinct RNP domains: Alu- and S-domain. The S-domain of SRP comprises the 7S.S part of RNA bound to SRP19, SRP54 and the SRP68/72 heterodimer. SRP54 is playing the key role in recognising signal sequences of nascent polypeptide chains and docking SRP to its receptor at the membrane. In archaeal cells like *Methanococcus jannaschii*, the SRP is simpler and consists of 7S RNA and two homologues of the eukaryotic SRP proteins, namely SRP19

and SRP54. Insight into the mechanism by which SRP functions in signal-sequence binding and release requires the knowledge of the protein-RNA and RNA-RNA interactions within the S-domain of SRP. Therefore, we are aiming for a high-resolution structure of the *M.jannaschii* ternary complex comprising 7S.S RNA, SRP19 and SRP54.

We analysed cryo-cooled crystals (0.05 x 0.1 x 0.2 mm<sup>3</sup>) of the complete S-domain of M.jannaschii - consisting of 7S.S RNA, SRP19 and full-length SRP54, on beam line ID23-1, ESRF, Grenoble in experiments conducted 07-May-04 to 08-May-04. These crystals diffracted to 2.5 Å. Diffraction data were processed using the program MOSFLM. The crystals belong to space group P212121 with cell parameters a = 70 Å, b = 129 Å, c = 165 Å. Our previously solved structure of the binary complex comprising the 7S.S RNA of M.jannaschii bound to SRP19 was used in molecular replacement searches with the program CNS and X-ray data from 10.0-4.0 Å resolution. The model was built in O and refined by CNS and REFMAC against all data from spacings between 20-2.5 Å. The  $R_{work}$  and  $R_{free}$  for the current model are 24.5% and 29.3%, respectively. A full structural analysis of the 7S.S RNA-SRP19-SRP54 complex is currently underway.