### 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: Structure determination of Human γD crystallin | Experiment<br>number:<br>LS1941 |  |
|---|--|---------------------------------|--|
| Beamline:   | Date of experiment:  | Date of report:                 |  |
| ID14 EH1  | from: 21.04.01 to: 23.4.01                                       | 20.08.01                        |  |
| Shifts:   | Local contact(s):  | Received at ESRF:               |  |
| 1   | Dr. Hassan   |                                 |  |
| Names and affiliations of applicants (* indicates experimentalists):  Ajit K. Basak*, Christine Slingsby. |  |                                 |  |
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| UK  |  |                                 |  |

## **Report:**

Gamma-crystallins are a family of closely related, long-lived proteins that occur at high protein concentrations inside lens fibre cells. In the human lens the gammaD member is expressed during foetal life. Several human genetic cataracts have been linked to point mutations in the gammaD-crystallin gene. Some of these mutations cause congenital cataract by altering the protein solubility and leading to crystallization in the lens. Protein crystallography is an excellent method to assess both the intramolecular and intermolecular structural consequences of these kinds of mutations. In a previous trip we collected diffraction data from crystals of recombinant human gamma-D crystallin that carried the mutation for aculeiform cataract.

In this trip we collected diffraction data from crystals of recombinant native protein that diffracted to 1.25A resolution. The structures for both the native and the mutant (which was collected in a previous trip) was solved by molecular replacement using bovine gammaD-crystallin as search molecule. The molecular and lattice structures of the native and mutant human gammaD crystals are currently being analysed.

|                       | Native                 | Mutant                |  |  |
|-----------------------|------------------------|-----------------------|--|--|
| Space group           | $P2_12_12_1$           | $P2_12_12_1$          |  |  |
| Diffraction Limit (Å) | 1.25 Å                 | 1.92Å                 |  |  |
| Cell parameters 33.22 | 2, 52.97, 90.47Å       | 33.40, 53.18, 89.59Å  |  |  |
| Rmerge (%)            | 4.8(18.9in 1.32-1.25Å) | 7.9 (10.7 2.03-1.92Å) |  |  |
| I/sd                  | 7.4(3.7)               | 6.4(5.5)              |  |  |
| Comp (%)              | 86.8% (45.5%)          | 99.2(97.6)            |  |  |
| Mult (%)              | 2.9 (1.8)              | 7.1(6.8)              |  |  |
| Refinement statics:   |                        |                       |  |  |
|                       | Native                 |                       |  |  |
| R-factor              | 16.36%                 | 15.57%                |  |  |
| Free R-factor         | 21.10%                 | 21.02%                |  |  |

16.00%

15.98%

17.00%

16.74%

Overall R-factor

R (work+test)