EUROPEAN SYNCHROTRON RADIATION FACILITY

INSTALLATION EUROPEENNE DE RAYONNEMENT SYNCHROTRON

ESRF

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 via the User Portal:

https://wwws.esrf.fr/misapps/SMISWebClient/protected/welcome.do

Reports supporting requests for additional beam time

Reports can 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:** Mechanisms underlying kainate receptor functions	**Experiment number**: MX2208
Beamline: CM01	**Date of experiment**: from: 02nd September 2019 to: 05th Septemeber 2019	**Date of report**:
Shifts: 9	**Local contact(s)**: Michael Hons	*Received at ESRF:*

Names and affiliations of applicants (* indicates experimentalists):

Janesh Kumar*

Ananth P. Burada*

***National Centre for Cell Science, S. P. Pune University, Pune-411007, India**

Report:

For the allocated beamtime at CM01 beamline, we were able to collect a decent dataset (~4120 movies) for ligand bound form of Delta receptor using the Krios Cryo-Electron Microscope. With the help of the local contact (HONS Michael), we loaded 7 grids in the grid loader containing ligand bound protein. First we screened the prepared grids and selected the best grids with good ice thickness and particle distribution. We chose grid no.7 and marked good squares with intermittent ice thickness imaged. The following parameters were adjusted using grid hole of each square:

Magnification: 130,000; pixel size: 1.067; spot size: 6; with a total dose of 40.38e-/Å2; fractions (# frames): 40; exposure time: 5s; images per hole: 3; amplitude contrast: 10%; drift correction was set once per grid square and autodefocus was set once every 3rd grid hole; Energy filter was set to 20eV and the data was collected in super resolution counting mode. The data collection was monitored using the ExiMX interface; data processing (motion correction and CTF estimation) was done simultaneously as the data was collected as shown in Figure 1. In addition, data collection statistics was performed by the system to measure the resolution distribution across movies, average motion per frame and astigmatism. Unfortunately, due to Gain reference issues with Gatan detector a few hours of data collection were lost. In total, 4120 movies were collected, out of which 3800 were good and 320 were bad containing gain reference issues and drift in movies.