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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 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:** Unravelling the kinetics pathways of ovine prion protein oligomerization	**Experiment number:** SC - 2395
Beamline: ID02	**Date of experiment:** **from:** 14 May 2008 **to:** 16 May 2008	**Date of report:** 15-08-2009
Shifts: 6	**Local contact(s):** Anuj Shukla	*Received at ESRF:*

Names and affiliations of applicants (* indicates experimentalists):

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Report:

Our work focuses on the prion protein (PrP), linked to Creutzfeldt-Jacob and bovine spongiform encephalopathy (BSE). We have studied the molecular mechanisms and kinetics of ovine PrP (OvPrP) oligomerization over the past few years [1-4] using a combination of biophysical techniques. These studies have shown that OvPrP partially unfolds into 3 intermediate states, which aggregate independently into 3 distinct oligomers O1, O2 (very instable) and O3. The same pattern of oligomerization is obtained with the C-terminal globular domain of the PrP, namely ΔOvPrP.

In this study, SAXS has been used to determine the low resolution structure of each of the monomeric species (OvPrP and ΔOvPrP) and the different oligomers obtained from their oligomerization.

The following figures show selected data obtained for the monomeric species OvPrP using Svergun's ATSAS suite programs (GNOM, DAMMIN) [5].

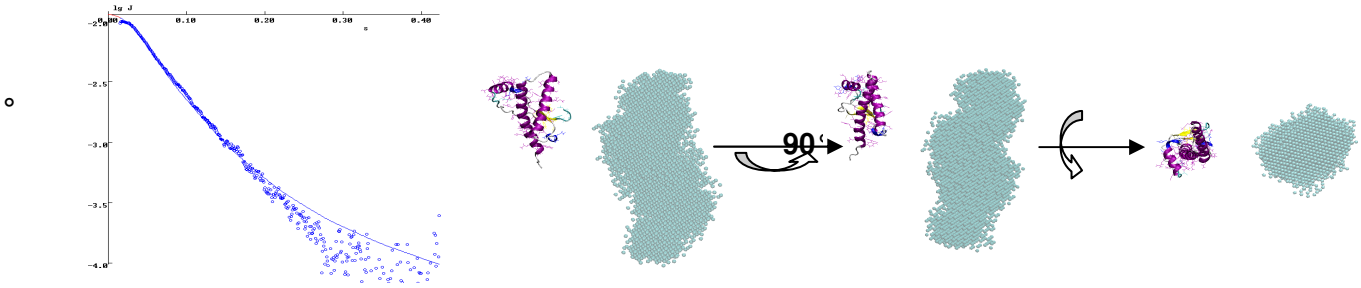

Fig1. *Scattering intensity I(q) from SAXS experiments for OvPrP monomer at 200μM and low-resolution shape compared to the crystal structure of ΔOvPrP domain (PDB 1UW3). Fitting the data using GNOM (Svergun's ATSAS suite) enabled the determination of a Radius of gyration Rg=2.75nm. DAMMIN (Svergun's ATSAS suite) was used for the sturcture determination by averaging 10 independent runs.*

SAXS data for the the monomer OvPrP, ΔOvPrP.and each oligomer have been successfully collected and analysed. These data will help to construct an architectural model of monomer assembly into the different oligomers.

These SAXS experiments at the ID02 have enabled us to set up and optimize the best experimental conditions for OvPrP monomer and oligoners structure analysis.

1. Eghiaian, F., et al., *Diversity in prion protein oligomerization pathways results from domain expansion as revealed by hydrogen/deuterium exchange and disulfide linkage.* Proc Natl Acad Sci U S A, 2007. **104**(18): p. 7414-9.
2. Rezaei, H., et al., *Amyloidogenic unfolding intermediates differentiates sheep prion protein variants.* J Mol Biol, 2002. **322**(4): p. 799-814.
3. Rezaei, H., et al., *Sequential generation of two structurally distinct ovine prion protein soluble oligomers displaying different biochemical reactivities.* J Mol Biol, 2005. **347**(3): p. 665-79.
4. Rezaei, H., et al., *High yield purification and physico-chemical properties of full-length recombinant allelic variants of sheep prion protein linked to scrapie susceptibility.* Eur J Biochem, 2000. **267**(10): p. 2833-9.
5. http://www.embl-hamburg.de/ExternalInfo/Research/Sax/software.html