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 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:** Synchrotron studies of manganese distribution in brain structure using quantitative x-ray fluorescence at cellular and tissular level to improve manganese enhanced magnetic resonance imaging (MEMRI) in vivo technique	**Experiment number**: MD-458
Beamline: ID21 & ID18F	**Date of experiment:** **from: 23/11/2009 to: 26/11/2009 (ID21)** **from: 20/01/2010 to: 25/01/2010 (ID18F)**	**Date of report**: 2/04/2010
Shifts: 9 & 15	**Local contact(s)**: M. Salomé (ID21) ; S. Bohic (ID18F)	*Received at ESRF:*

Names and affiliations of applicants (* indicates experimentalists):

*A. Daoust (Thesis student) Laboratory INSERM U836 Team 5; Grenoble Institute of Neuroscience

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Report:

It has been discovered that manganese enhanced magnetic resonance imaging (MEMRI) can be used to trace neuronal connections activated following specific stimulation. These studies used manganese (Mn) as a contrast agent. The biological basis for the movement of Mn^{2+} into tissues and its cellular distribution are still unclear. A major drawback to the use of Mn^{2+} as a contrast agent is its cellular toxicity. Indeed, non specific enhancement contributes to MEMRI. This raise the need to better depict the quantitative Mn distribution at cellular scale a critical issue for the development of MEMRI, particularly to identify the brain cells targeted with Mn from which results increaseMRI contrast, in order to increase sensitivity so that lower doses of Mn^{2+} can be used and to drive the development of new Mn-based contrast agents.

November 2009: 9 shifts on ID21. 1shift ½ lost due to general ESRF file server failure.

The ID21 KirkPatrick-Baez focusing sytems was not available (problems on mirors) and Zone plate was used at 7.2 keV; (spot size achieved 0.2 x 0.9 mm) with a flux of 3.10^{+9} e.g a factor 10 or 20 less than wit h KB optic. All animals that receive $MnCl_2$ via various route of administration were imaged at MRI platform prior brains cryosectioning were analysed on synchrotror. Rat brain sections were 20 microns in thickness . Olfactory bulb cryosections for animals that receive intranasal exposition to $MnCl_2$ were possible to analysed first with scanning larger region with 50 microns pinhole and then on smaller region of interest using the zone plate. Integration time wasextremely long due to combined low flux available and to the very low content of Mn estimated of few ppm only despite injection with $MnCl_2$; and few tenth ppm in the olfactory bulb.

January 2010: 15 shifts on ID18F

The set-up used was at 14 keV excitation with the 3 undulators of ID18 beamline and Aluminium compound refractive lenses used for ID18F. This result in a flux of around 2. 10^{+10}. The integration was 10 s per pixel

and 20 micron step was used with the objective of mapping most of the hippocampus. This was achieved for only 2 control and 2 intraperitoneal; 1 intraveneous and intracranial injected animals.

The work carried out by the PhD student is already important on the side of in vivo Mn-MRI protocol for imaging and immunohistological processing. But Mn concentration despite exgeneous injection of $MnCl_2$ and clear MRI signal, Mn is fairly low in hippocampus (2-3 ppm.) and olfactory bulb. Consequently, the synchrotron results due to the low photon flux achieve on ID21 and ID18F and some technical problems encountered could not bring the necessary results in order to make correlative images (MRI, histology, synchrotron XRF). As mention, best results were obtained at low spatial resolution on large zone at ID18F. These studies are very relevant and informative so far the hippocampus was scarcely investigated for trace element content and for first time here for Mn as neuronal contrast agent.This allow us to choose one injection modality for a final study that would required another much higher flux beamline ID22.Indeed, in order to identify the cells implicated in Mn storage compared to control, ID22 seems to us the only beamline possible to achieve simultaneously 1-5 micrometer beamsize with high flux (5.10+11 ph/s) that would allow to reach for Mn ppm sensitivity within 1-2 sec counting time. That are the reason of re-applying for beamtime on this project to allow the PhD student Alexia Daoust to reach the goal of the project (correlative imaging and cellular Mn location compared to MRI contrasted areas) and to publish it (This is not possible actually from the data we already processed**).**

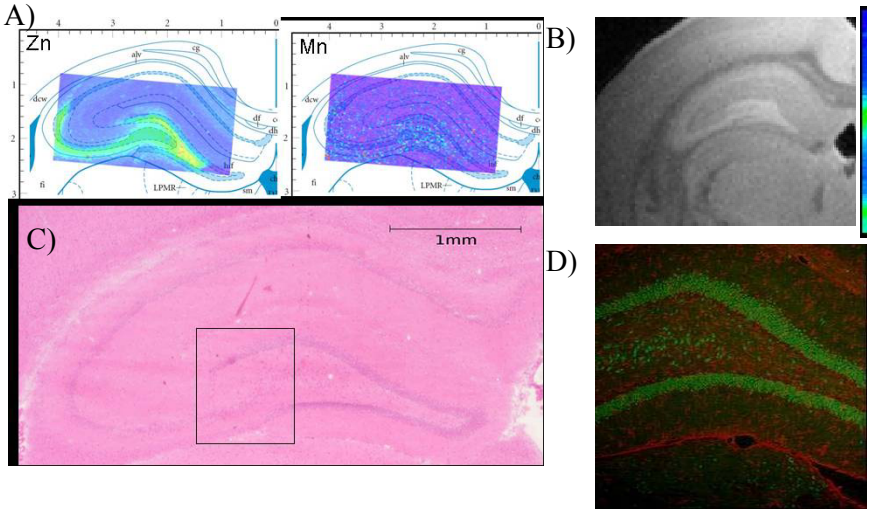

Figure: A Image of Zn and Mn distribution in rat brain hippocampus (animal injected with $MnCl_2$ and imaged at MRI facility prior brain cryosectioning and XRF studies) obtained at ID18F with 10s/pixel and surperimposed on a rat brain atlas. B) Mn MRI coronal section of rat brain after $MnCl_2$ intraperitoneal injection. C) Hematoxylin eosin staining rat brain cryosection adjacent to the one used unstained for XRF. D) Immunohistofluorescence staining of dentate girus for neurone (green) and glial cells (red)