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:** Synchrotron-generated microbeam radiation as a possible tool to eliminate seizure generators in the GAERS	**Experiment number**: MD 438
Beamline: ID17	**Date of experiment**: from: 11 Nov 2009 to: 13 Nov 2009	**Date of report**:
Shifts: 12	**Local contact(s)**: Raphaël Serduc	*Received at ESRF:*

Names and affiliations of applicants (* indicates experimentalists):

Elke Brauer* - ESRF
Alberto Bravin - ESRF
Geraldine Le duc -ESRF
Raphael Serduc* -ESRF
Antoine Depaulis* - Inserm U836
Francois Esteve - Inserm U836
Tanguy Chabrol* - Inserm U836
Benoit Pouyatos* - Inserm U836
Isabelle Guillemain - Inserm U836

Report:

Rationale :

The Genetic Absence Epilepsy Rats from Strasbourg (GAERS) is one of the best-described genetic model of absence epilepsy. The GAERS is characterized by spontaneous bilateral and synchronous 7-9Hz spike-wave discharges (SWDs) generated by the thalamocortical loop, concomitant with behavioral arrests. Recent data showed that SWDs are initiated in the somatosensory (S1) cortex - the cortical "focus" - before their diffusion to the rest of the cortex and the ventro-basal thalamus. To date, it is unknown whether the focus is necessary and sufficient for the initiation of absence seizures.
The aim of this study was to investigate the effect of the inactivation of either the cortical focus or the thalamic relay nuclei on SWD initiation and generalization using the novel method of interlaced microbeam radiation therapy (MRT).

Method:

MD438 beamtime allowed us to irradiate bilaterally 20 GAERS rat using the technique described in Serduc et al. (2010) PLoS ONE, and Serduc et al. (2010) JSR (In press).

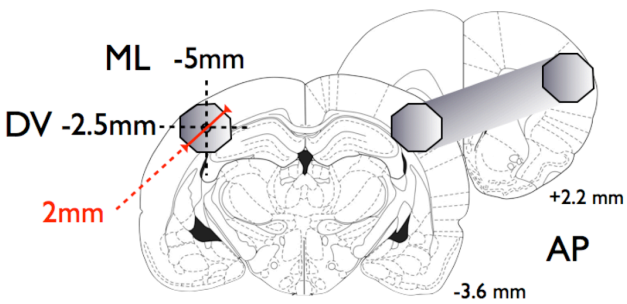
1 - Serduc R, Brauer-Krisch E, Siegbahn EA, Bouchet A, Pouyatos B, et al. (2010) High-Precision Radiosurgical Dose Delivery by Interlaced Microbeam Arrays of High-Flux Low-Energy Synchrotron X-Rays. PLoS ONE 5(2): e9028. doi:10.1371/journal.pone.0009028

2 - Raphaël Serduc, Gilles Berruyer, Michel Renier, and Christian Nemoz (2010) In vivo pink beam imaging and fast alignment procedure for rat brain lesion microbeam radiation therapy (In press).

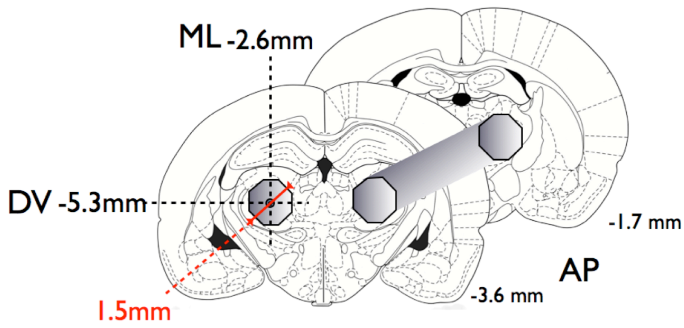
Briefly, animals were anaesthetized, placed in a plexiglas head holder, their Bregma point localized using

pink beam imaging (as described in publication #2). Coordinates of irradiation were calculated relatively to this reference point. Irradiated volumes are described in the figures below.

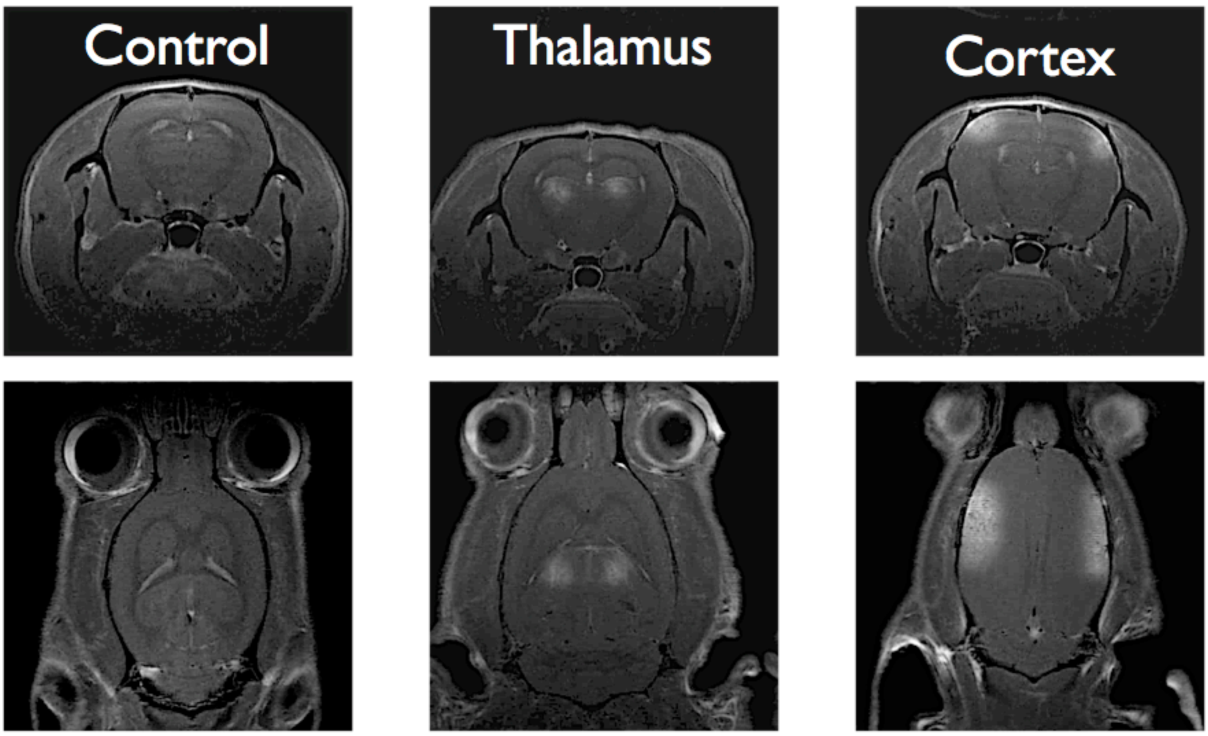
Cortex Irradiation



Thalamus irradiation



Interlaced MRT was obtained by 4 successive microbeam irradiations between which the target, *i.e*. the rat brain, was rotated by 45° (as described in publication #1). The peak entrance dose was 200Gy.
Fifteen days after irradiation, 7T T1-weighed gadolinium MR images were acquired on control (sham), cortex- and thalamus-irradiated animals. Sample images are shown below. They show very delimited irradiated zones at the intended location.

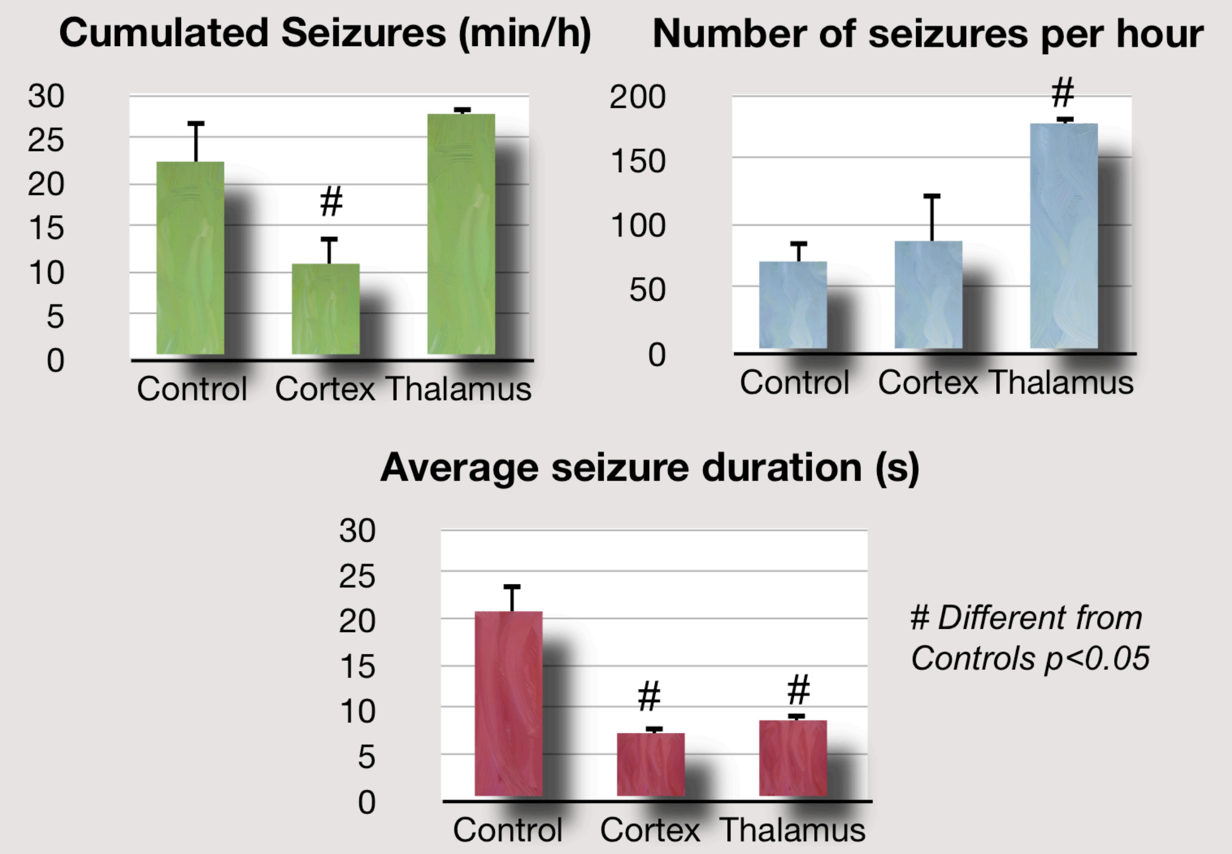


Control and irradiated animals were implanted with three bipolar tungsten electrodes in different brain structures for weekly EEG monitoring.

Results:

A beneficial effect was observed in the cortex-irradiated animals (n=3), as soon as 3 weeks post-irradiation. The average seizure duration was decreased from 20 to 7s. As a result, the cumulated seizure duration was decreased by about 50%.
On the opposite, thalamus-irradiated animals (n=5) displayed increased number of brief seizures that resulted in a slight increase of the cumulated seizure duration. As of now (3 months post irradiation), these effects are still constant.

Below are depicted the effects of cortex and thalamus irradiations on the seizure number and duration, at 2 months post-irradiation.



2-month post-irradiation	number of seizures	seizure duration
S1 Cortex Irradiation	=	- 50 %
Thalamus Irradiation	+ 150 %	- 50 %

Conclusions :

➡ This pilot study supports the hypothesis of a leading role of the S1 cortex in the generation of SWDs.
➡Interlaced MRT offers the possibility to disrupt brain regions involved in SWDs, PRECISELY and NON-INVASIVELY.
➡For the first time, seizures were reduced by radiotherapy in an animal model of epilepsy.

Valorisation :

- Articles :

1 - Serduc R, Braëuer-Krisch E, Siegbahn EA, Bouchet A, Pouyatos B, et al. (2010) High-Precision Radiosurgical Dose Delivery by Interlaced Microbeam Arrays of High-Flux Low-Energy Synchrotron X-Rays. PLoS ONE 5(2): e9028. doi:10.1371/journal.pone.0009028

Microbeam Radiation Therapy (MRT) is a preclinical form of radiosurgery dedicated to brain tumor treatment. It uses micrometer-wide synchrotron-generated X-ray beams on the basis of spatial beam fractionation. Due to the radioresistance of normal brain vasculature to MRT, a continuous blood supply can be maintained which would in part explain the surprising tolerance of normal tissues to very high radiation doses (hundreds of Gy). Based on this well described normal tissue sparing effect of microplanar beams, we developed a new irradiation geometry which allows the delivery of a high uniform dose deposition at a given brain target whereas surrounding normal tissues are irradiated by well tolerated parallel microbeams only. Normal rat brains were exposed to 4

focally interlaced arrays of 10 microplanar beams (52 mm wide, spaced 200 mm on-center, 50 to 350 keV in energy range), targeted from 4 different ports, with a peak entrance dose of 200Gy each, to deliver an homogenous dose to a target volume of 7 mm3 in the caudate nucleus. Magnetic resonance imaging follow-up of rats showed a highly localized increase in blood vessel permeability, starting 1 week after irradiation. Contrast agent diffusion was confined to the target volume and was still observed 1 month after irradiation, along with histopathological changes, including damaged blood vessels. No changes in vessel permeability were detected in the normal brain tissue surrounding the target. The interlacing radiation-induced reduction of spontaneous seizures of epileptic rats illustrated the potential pre-clinical applications of this new irradiation geometry. Finally, Monte Carlo simulations performed on a human-sized head phantom suggested that synchrotron photons can be used for human radiosurgical applications. Our data show that interlaced microbeam irradiation allows a high homogeneous dose deposition in a brain target and leads to a confined tissue necrosis while sparing surrounding tissues. The use of synchrotron-generated X-rays enables delivery of high doses for destruction of small focal regions in human brains, with sharper dose fall-offs than those described in any other conventional radiation therapy.

2 - Raphaël Serduc, Gilles Berruyer, Thierry Brochard, Michel Renier, and Christian Nemoz (2010) In vivo pink beam imaging and fast alignment procedure for rat brain lesion microbeam radiation therapy (In press).

We have developed a fast 50 µm-accuracy alignment procedure for the radiosurgery of brain lesions in rats, using microbeam radiation therapy. *In vivo* imaging was performed using the pink beam (35-60 keV) produced by the ID17 wiggler at the ESRF opened at 120 mm and filtered. A graphical user interface has been developed in order to define the irradiation field size and to position the target with respect to the skull structures observed on X-rays images. The method proposed here allows tremendous time saving by skipping the swap from white beam to monochromatic beam and vice versa. To validate the concept, somatosensory cortex or thalamus of GAERS rats were irradiated under several ports using this alignment procedure. The magnetic resonance images acquired after contrast agent injection showed that the irradiations were selectively performed in these two expected brain regions. Image-guided microbeam irradiations have therefore been realized for the first time ever, and thanks to this new development, the ID17 biomedical beamline provides a major tool allowing brain radiosurgery trials on animal patients.

- Communications :

1 - Pouyatos B. 19th Neuromeeting Beaune 20-23/01/2010 : The focus of absence seizures in the GAERS (Lecture)
2- Pouyatos B. ESRF User Meeting 8-12/02/2010 : Synchrotron-generated microbeam radiation as a possible tool to eliminate seizure generators in the GAERS (Poster).
3- Serduc R. MASR 2010 Melbourne 16-18/02/2010 : High-flux low-energy challenging high-energy conventional radiation therapy for brain lesions? Radiosurgical high dose delivery with synchrotron generated X-rays applied to epileptic foci in a rat genetic model of epilepsy (Lecture).