EUROPEAN **S**YNCHROTRON **R**ADIATION **F**ACILITY

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:** Functional imaging of the effects of inhaled drugs and air pollution particles on regional ventilation in healthy and asthmatic animals	**Experiment number**: MD238
Beamline: ID17	**Date of experiment**: from: I/2008 to: II/2008	**Date of report**: 15.1.2009
Shifts: 15	**Local contact(s)**: Christian Nemoz	*Received at ESRF:*

Names and affiliations of applicants (* indicates experimentalists):

Anssi Sovijärvi	Helsinki University Central Hospital, Finland
*** Sam Bayat**	University of Amiens, France
* **Skander Layachi**	**-"-**
*** Liisa Porra**	University of Helsinki, Finland
*** Satu Strengell**	**-"-**
*** Heikki Suhonen**	**-"-**
Pekka Suortti	**-"-**
Walid Habre	Geneva University Central Hospital, Switzerland
*** Tibor Janosi**	**-"-**
Zoltan Hantos	University of Szeged, Hungary
Ferenc Petak	**-"-**
*** Gergely Albu**	**-"-**

Report:

This report is the second year report of Long Term Project MD238, where 90 shifts were granted for 2 years. The proposal was accepted originally for the beamtime allocation periods 2006/II – 2008/I. Due to reconstruction of ID17, the use of beamtime was delayed by one year, and so far 62 shifts have been used: 30 in 2006/II, 17 in 2007/I, 15 in 2008/II. Accordingly, 2 experiments of 15 and 13 shifts remain to be carried out in 2009.

In the first and second year total 4 of the 6 experiments were performed and 62 of the 90 shifts were used.

Beamline	Shifts		Start Date	Finish Date	Local Contact
ID17	15	MD238/1	07 December 2006	12 December 2006	Dr. Christian NEMOZ
ID17	15	MD238/2	05 February 2007	12 February 2007	Dr. Christian NEMOZ
ID17	17	MD238/3	11 July 2007	16 July 2007	Dr. Christian NEMOZ
ID17	15	MD238/4	04 December 2008	09 December 2008	Dr. Christian NEMOZ

Background and aim of the study:

Traditional measurements of lung function such as spirometry can at best provide overall assessments and do not give any insight into the localization and heterogeneity of airway response. We have introduced a novel CT imaging technique that uses synchrotron radiation to quantitatively image inhaled stable xenon gas within the airways with a high spatial resolution (Bayat et al 2006). Using this method, regional lung volume, ventilation, and airway luminal diameters down to 2 mm can be measured. The spatial resolution of this technique is the best available for regional ventilation imaging in small animals, and the structure of the lungs can be studied simultaneously.

K-edge subtraction (KES) imaging and Forced Oscillation Technique (FOT) were combined in all MD238 experiments. KES provides high-resolution images of lung structure and ventilation distributions, and the frequency-dependent impedance of lung, as measured by FOT, provides physiological parameters of lung biomechanics. Simultaneous observations by KES and FOT yield a comprehensive picture of lung function. Imaging setup is presented in figure 1.

Methacholine challenge (Mch) is routinely used as non-specific challenge to diagnose bronchial hyper-reactivity, although its effect in the lungs and main airways as compared to a specific allergen challenge are not precisely known. Mch provocation was studied in first 2 experiments of the LTP project MD238/1 and 2. The aim of these studies was to use new asthma model using sensitized rabbits, and compare images obtained with synchrotron radiation with results on overall lung mechanics obtained with the forced oscillation technique (FOT) (Petak et al, 2006). This animal model was used to study first the effect of Mch in the lungs, and compare the results to the effects of allergen provocation, and the results were compared to the results from healthy animals (Strengell et al, 2008).

Tobacco smoke is an increasing health problem in the world, and more information about the acute effects of tobacco smoke in the lungs is needed. The effect of acute tobacco smoke was studied in the third experiment MD238/3, where the effects of previous smoke provocation on the subsequent Mch provocation were studied (Janosi et al, 2009). More detailed summary can be found in first year report of MD238 (2007).

Studies of small animals are important lines of future research. In addition to development of whole-lung and 3D imaging, studies of transgenic mice are important for separation of expressions of hereditary and induced diseases. The methods of mechanical ventilation and physiological monitoring in KES+FOT experiments were developed for rabbits, and first trials of applying the methods in rats and mice have been carried out in the 4th experiment MD238/4.

Experiment MD238/4:

This experiment concentrated mainly on technical development using small animals (3 rats, 7 mice) and high-resolution FReLoN detector (50μm pixel size). Ventilator setup and animal holder were modified to be suitable for smaller animals, and images were acquired at baseline and after Mch aerosol provocation (figure 2a and 2b). All results will be fully analyzed and published later.

Offline experiments:

2 offline experiments were performed in Geneva University Central hospital to complete the results obtained from cigarette exposure study (MD238/3). Cigarette smoke is a complex mixture of a particle phase, containing multiple components including nicotine, and a gas phase containing NO and CO, both of which are bronchodilators.The combined KES+FOT experiments were completed by off-line FOT experiments where the animals were exposed to the gas phase of cigarette smoke, NO, CO, IV nicotine, among other mediators. Although some bronchodilatation was observed with NO, and CO, the delayed inhibitory effect of cigarette smoke exposure on airway reactivity in this species could only partially be reproduced by the smoke gas phase. Inter-species differences in the response to cigarette smoke hold great promise in the understanding of the machanisms of the toxic effects of cigarette smoke on airway function. Experiments were performed with the same ventilator setup used at the ESRF, and lung mechanics were measured with FOT system. Rabbits were exposed with HEPA -filtered cigarette smoke, alpha-chymotrypsine, nicotine, NO and CO gases, and the results were compared with the earlier results with non-filtered smoke. (Janosi et al., manuscript in preparation).

References:

Bayat S, Porra L, Suhonen H, Nemoz C, Suortti P, Sovijärvi AR. Differences in the time course of proximal and distal airway response to inhaled histamine studied by synchrotron radiation CT. *J Appl Physiol*. 2006 Jun;100(6):1964-73.

Peták F, Hantos Z, Adamicza A, Gálity H, Habre W. Development of bronchoconstriction after administration of muscle relaxants in rabbits with normal or hyperreactive airways. *Anesth Analg*. 2006 Jul;103(1):103-9.

Strengell S, Porra L, Janosi T, Petak F, Suhonen H, Suortti P, Hantos Z, Sovijärvi A, Habre W, and Bayat S. Metchacholine and ovalbumin challenges assessed by forced oscillations and synchrotron lung imaging. Submitted to *AJCCRM* (2008).

T. Janosi et al. Bronchodilation following acute cigarette smoke exposure in rabbits assessed by forced oscillation technique and K-edge subtraction imaging (*manuscript*) (2009).

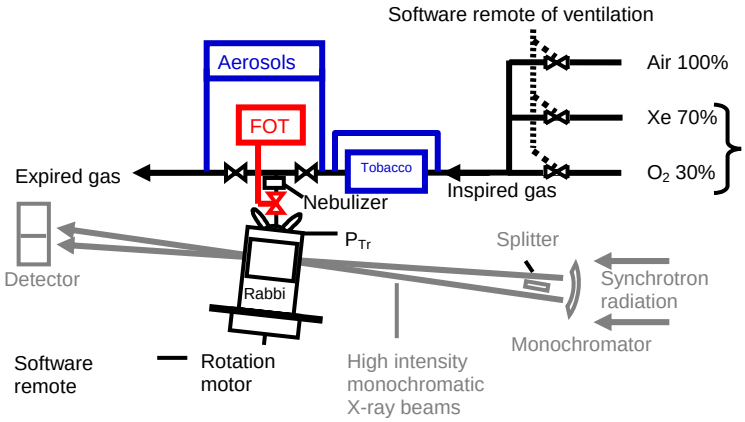

Figure 1: Imaging setup with new modifications; FOT measurement system used in all experiments is shown in red, and tobacco chamber and aerosol measurement system, used in MD238/3 experiment is shown in blue.

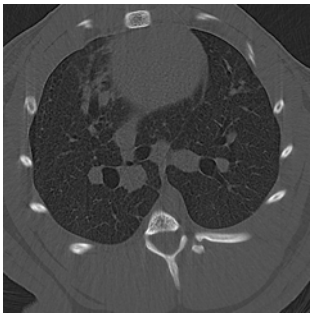

Figure 2a: Single-energy CT image from rat lungs obtained by 50µm FReLoN detector. Image is 4cm wide.

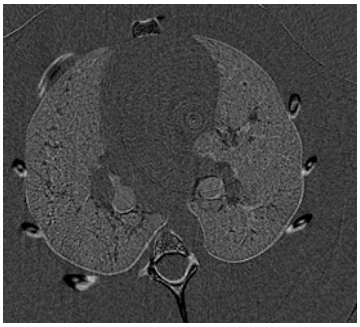

Figure 2b: KES image from rat lungs obtained by 50µm FReLoN detector. Image is 4cm wide.

<u>Conference presentations 2008:</u>

1. T. Janosi et al. Bronchodilation following acute cigarette smoke exposure in rabbits assessed by forced oscillation technique and K-edge subtraction imaging. **European Respiratory Society annual congress 2008**, 4-8.10.2008, Berlin, Germany. Presentation 4432.
2. S. Bayat, L. Porra, H. Suhonen, P. Suortti, A. R. A. Sovijärvi. Paradoxical conducting airway response and heterogeneous regional ventilation after histamine inhalation in healthy rabbit studied by synchrotron radiation CT. **European Respiratory Society annual congress 2008**, 4-8.10.2008, Berlin, Germany. Poster 1138.
3. L. Porra, S. Bayat, H. Suhonen, S. Strengell, T. Janosi, P. Suortti, ARA. Sovijärvi. Histamine-Induced Changes of Regional Lung Tissue and Airway Distensibility in Rabbit studied by Synchrotron Radiation CT. **American Thoracic Society international conference 2008**, 16-21.5.2008, Toronto, Canada. Poster.
4. F. Peták et al. Funkcionális és strukturális elváltozások a tüd?ben metakolin provokáció és allergiás reakció során szenzitizált nyulakban (Functional and structural changes in the lungs following methacholine provocations and allergic reactions in sensitized rabbits). **Meeting of the Hungarian Synchrotron Committee**, Hungarian Academy of Sciences, 7.5.2008. Hungary. Presentation.
5. S. Strengell et al. Functional Imaging of Regional Airway and Ventilation Response to Intravenous Allergen and Methacholine Studied by Synchrotron Radiation CT in a Rabbit Model of Asthma. **14th Congress SSCPNM-2008.** 13-15.2.2008, Lillehammer, Norway. Presentation.