



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.



	Experiment title: Mapping Ion Concentrations in the Airway Epithelium and Surface Liquid of Normal and Cystic Fibrosis Mice	Experiment number: LS-2128
Beamline: ID21	Date of experiment: from: 03 July 2002 to: 07 July 2002	Date of report: 26/8/2002
Shifts: 15	Local contact(s): Jean Susini	<i>Received at ESRF:</i>
Names and affiliations of applicants (* indicates experimentalists): Sam BAYAT MD PhD. Medical Research ID17, ESRF BP 220 6, rue Jules Horowitz, F-38043 Grenoble Cedex, France. Francis Grimbert MD. TIMC, PRETA, UMR CNRS 5525, Université, Joseph Fourier, Faculté de Médecine, Laboratoire de Physiologie, Domaine de La Merci, 38700 La Tronche, France.		

Report:

Cystic Fibrosis (CF) is an autosomal recessively inherited disease caused by a mutation of the CF transmembrane conductance regulator (CFTR) gene, which encodes for the cAMP-regulated Cl^- channel in the apical membranes of epithelial cells. The pathophysiology of CF remains controversial, and an accurate description of Airway Surface Liquid (ASL) properties is critical to the rational design of therapies for cystic fibrosis and other airway diseases. Comparison of wild type and CF mice in terms of absolute ion concentrations in these compartments would provide a valuable experimental model to explore the pathophysiology of CF. The aim of the present experiment was to test the feasibility of mapping ion concentrations, particularly Na and Cl in blood, epithelial intracellular compartments, and in ASL. Male C57BL/6-j mice were anesthetized with inhaled Isoflurane. The lungs and heart were removed and immediately frozen in isopentane cooled with liquid nitrogen. Tissue sections of 6 μm were prepared using a cryostat microtome, and maintained at -70°C on Kapton foils. Tissue sections were slowly freeze-dried, and studied at ambient temperature under vacuum. Ion concentration mapping was performed using the scanning synchrotron radiation X-ray microscope on ID21 with a beam at 2.9 keV. The focused beam size was $1 \mu\text{m} \times 1 \mu\text{m}$ at the focal plane. X-ray fluorescence maps were obtained for S, P, Cl, and Na (figure 1), in regions of interest centered on the tracheal epithelium. Since the samples were lyophilized, the ASL could not be evaluated. The Na x-ray fluorescence signal was weak; therefore prolonged integration times were required for mapping.

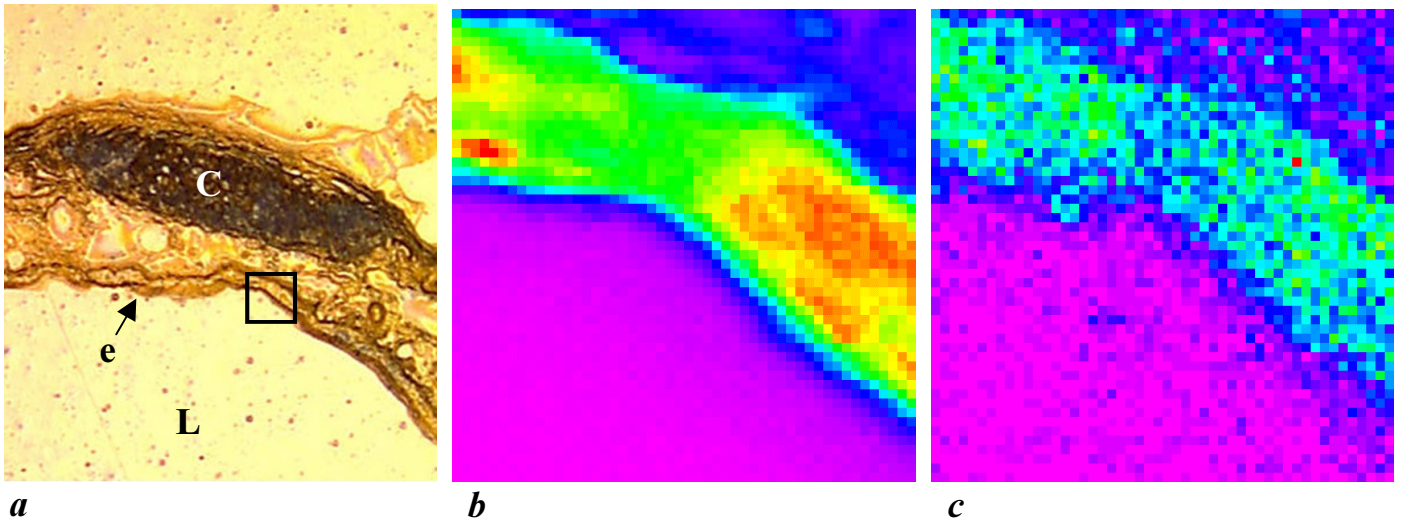


Figure 1: *a*; lyophilized 6 μm unstained slice of trachea, optical microscope ($\times 10$ magnification) showing the lumen (L), the epithelium (e), and tracheal cartilage (C), a $50 \times 50 \mu\text{m}$ region of interest (\square) centered on the tracheal epithelium (thickness $\sim 18 \mu\text{m}$) was scanned, *b*; elemental distribution of Cl in the region of interest, integration time: 3 sec/pixel *c*; elemental distribution of Na in the region of interest, integration time: 3 sec/pixel.

These preliminary results demonstrate the feasibility of Cl and Na mapping of the airway epithelium, in tissue slices. Further work will be necessary to obtain data on ion concentrations in the ASL. For this purpose a low temperature sample environment is currently under study. An enhancement of the incident photon flux will be necessary for quantitative mapping of Na concentrations. The experimental setup could be improved in the near future for this reason, by the design and use of multilayers instead of Si(1,1,1) crystals in the monochromator.

