

Beamline: 19	Date of experiment: from: 27/04/2003, 20/07/03 21/07/03	Date of report: 1/09,
Shifts: 9	Local contact(s): P. Cloetens	<i>Received at ESRF:</i>
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Report:

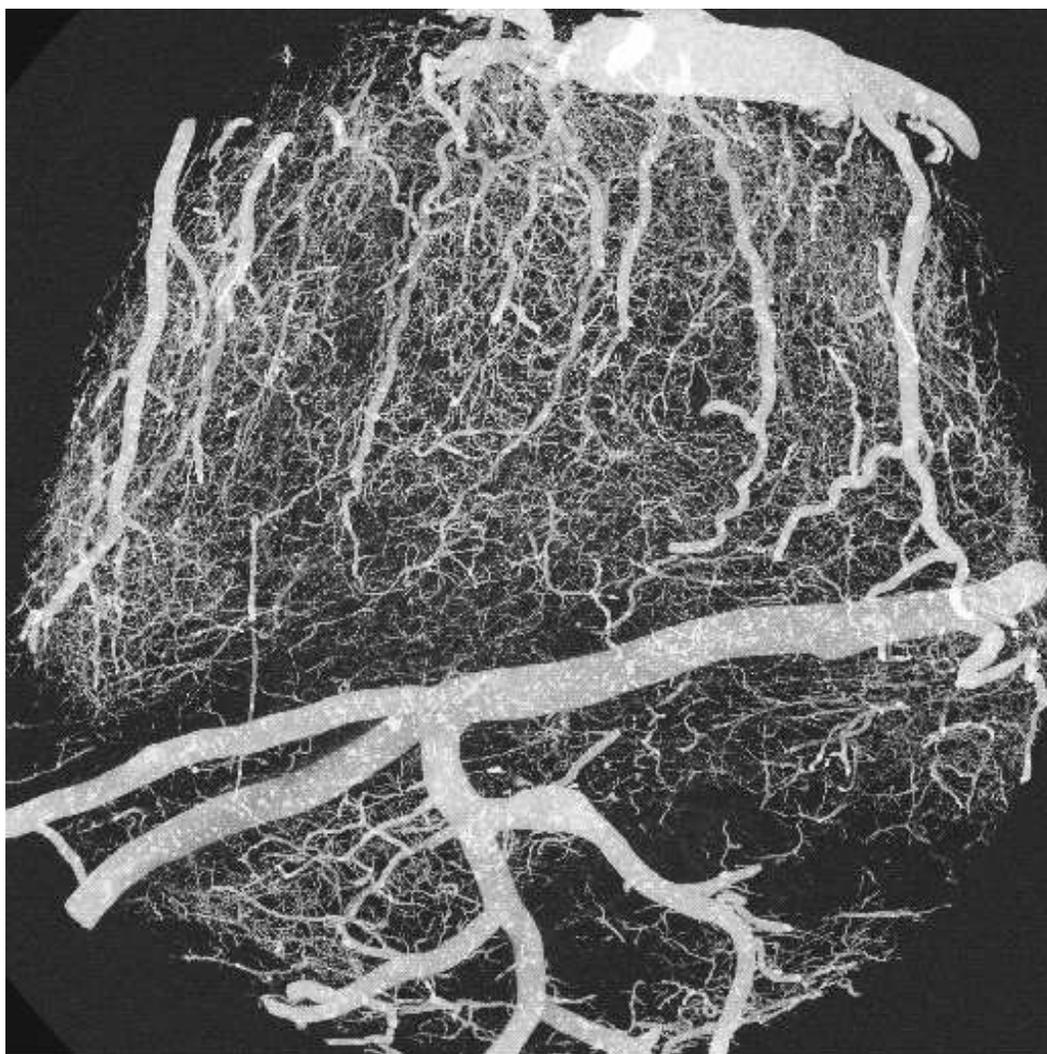
The objective of our present research being, in the first place, the improvement of sample preparation protocol, we first thanks the help of line 19 direction which has accepted our request to use allocated shift on a short beam line access (one day).

Albeit our previous report has shown repeated progress on the obtained images and the sample preparation, much was still to be done in order to better analyse the impact of different aspects of sample preparation protocol on the image quality. Focusing on this crucial matter, our new efforts have provided important footsteps :

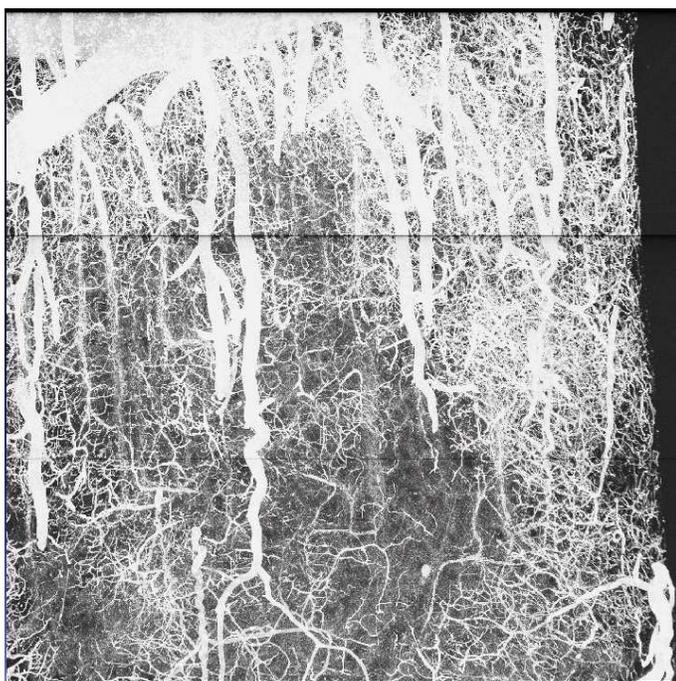
- The sample preparation has been done with adults laboratory albinos rats, that are easier (and cheaper) to obtain, to inject and to dealt with.
- Two contrast agents (Iron-dextran, and sulfate Baryum) have been compared. The later, use at the highest possible concentration (400mg/ml) has given far better results for X-ray imaging.
- Each injection has been associated with a post-mortem optical microscopy inspection on the collateral hemisphere's vascular bed. This inspection permits to estimate the spatial as well as the individual variability of the injection protocol. It is also a useful systematic observation to select the more interesting sample to image.
- Some intriguing experimental observation showing the presence of small bubbles in some regions of the vascular capillary bed have been elucidated as being associated with the evaporation of the dehydration agent (propylene oxide) during the resin cast preparation.

The figure represented on the next page, are projection views obtained from different sample extracted from different cortical regions of the same rat. Image (a) is a bottom view (x/y plane projection) of a few millimetre cube volume of fronto-central cortex, whereas (b) —resp. (c)— are y/z —resp. x/z — projection views of frontal cortex. One clearly distinguishes the cortical grey matter region from its dense vascularisation, as well as its typical arterial columnar structures, that appears as being sub-orthogonal to the original cortical surface. To our knowledge, these results shows the first three-dimensional images of the all grey matter vascular structure, over its entire thickness.

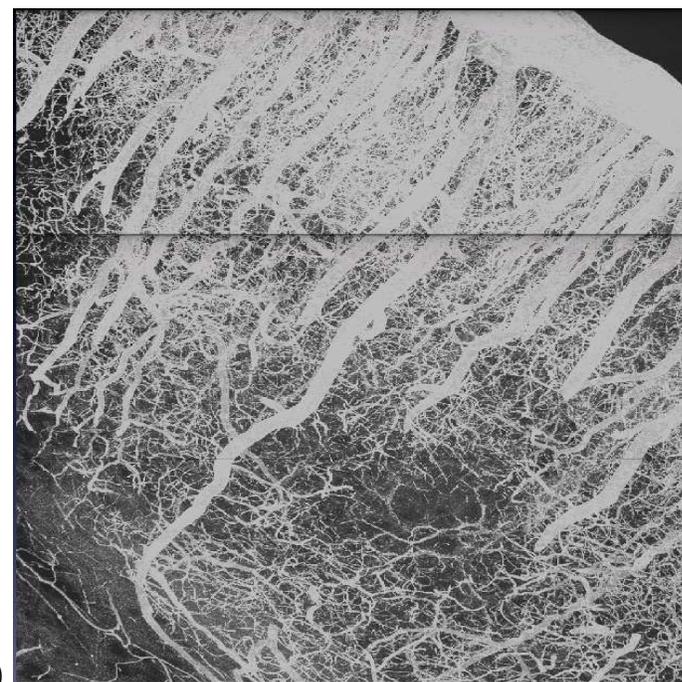
In conclusion high-resolution X-ray is a very interesting tool to analyse micro-vascular network structures. The obtained results demonstrate the ability of this experimental technique for giving important structural informations on those networks, over an impressive range of length-scales (from micron to millimetre) in a full three-dimensional volume. This research has yet given rise to a first article related to the development of this new experimental imaging protocol being submitted to *J. of Microscopy*, the abstract of which is hereby attached.



(a)



(b)



(c)

X-Ray high resolution vascular network imaging : sample preparation and imaging technique

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Key words : microtomography, X-ray, synchrotron, sample preparation, micro-vascular, brain, contrast agent

Abstract :

This paper presents the first application of high resolution X-ray synchrotron tomography to the imaging of micro-vascular networks in biological tissue samples. This technique offers the opportunity of screening the full three dimensional vascular structural organisation from the micron to the millimeter scale. This paper presents a specific sample preparation along with the X-ray imaging principles as well as specific technical aspects of the imaging procedure. Either Baryum or Iron were injected in the vascular network to be used as contrast agents. Their relative concentration, as well as the composition of the injected solution were investigated. The impact of the sample preparation on the X-ray synchrotron tomography images has been studied. Synchrotron high-resolution computed tomography offers new prospects in the three-dimensional imaging of biological tissu vascular networks.