

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 via the User Portal:
<https://www.esrf.fr/misapps/SMISWebClient/protected/welcome.do>

Deadlines for submission of Experimental Reports

Experimental reports must be submitted within the period of 3 months after the end of the experiment.

Experiment Report supporting a new proposal (“relevant report”)

If you are submitting a proposal for a new project, or to continue a project for which you have previously been allocated beam time, you must submit a report on each of your previous measurement(s):

- even on those carried out close to the proposal submission deadline (it can be a “*preliminary report*”),
- even for experiments whose scientific area is different from the scientific area of the new proposal,
- carried out on CRG beamlines.

You must then register the report(s) as “relevant report(s)” in the new application form for beam time.

Deadlines for submitting a report supporting a new proposal

- 1st March Proposal Round - **5th March**
- 10th September Proposal Round - **13th September**

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.

Instructions for preparing your Report

- fill in a separate form for each project or series of measurements.
- type your report in English.
- include the experiment number 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: Virtual histology of human brains with Alzheimer's disease	Experiment number: MD-1018 (& MD-1106)
Beamline: ID-19	Date of experiment: from: 5 Feb 2017 to: 7 Feb 2017 (MD-1018) from: 13 Mar 2018 to: 14 Mar 2018 (MD-1106)	Date of report: 01/2022
Shifts: 6 (+ 3)	Local contact(s): Lukas Helfen – Elodie Boller	<i>Received at ESRF:</i>
Names and affiliations of applicants (* indicates experimentalists): Marlène Wiart, CarMeN Laboratory* Fabien Chauveau, Lyon Neuroscience Research Center* Coralie Gislard, Lyon Neuroscience Research Center* Hugo Rositi, CREATIS* Elisa Cuccione, CREATIS* Françoise Peyrin, CREATIS* Cécile Olivier, CREATIS* Max Langer, CREATIS Louise Van der Weerd, Molecular Imaging Laboratories Leiden (MILL)		

Report:

Beamline allocation allowed to validate an XPCT protocol and use it as a fast virtual histology tool (ca 3 minutes acquisition per mouse brain or human cortical sample), relevant for various neurological alterations. Brain sample from transgenic animals and patients with neurodegenerative diseases were imaged with a polychromatic "pink" incident X-ray beam of 26 keV energy. Tomographic reconstructions were performed using the single-distance phase-retrieval approach ("Paganin" method) with PyHST2 software, the δ / β ratio being set to 1000, yielding an isotropic voxel size of 6.5 μm .

So far, the outputs of these experiments are the following:

- whole-brain myelin mapping in white-matter injury models

Preliminary observations were presented at the European Molecular Imaging Meeting [1] and, after validating original tools for visualisation and analysis of white-matter tracts, quantitative results were published in Biomedical Optics Express [2].

- 3D morphologies of amyloid- β plaques in Alzheimer's disease models

Preliminary observations were presented at the European Molecular Imaging Meeting [3] and, after validating original tools for segmentation and analysis of plaque morphologies, quantitative results were published in Biomedical Optics Express [4].

In particular, we demonstrated that the white-matter contrast is related to a mismatch of refractive indices between myelin and surrounding tissue, occurring after the removal of water by ethanol. Indeed, we used brain clearing, a technique that removes lipids (and hence the myelin sheath) to render the brain translucent to light before ethanol dehydration, and observed a loss of white-matter tract contrast after lipid removal in both human (Fig. 3(B)) and mouse brains (Fig. 3(D)), suggesting a role of myelin in contrast generation.

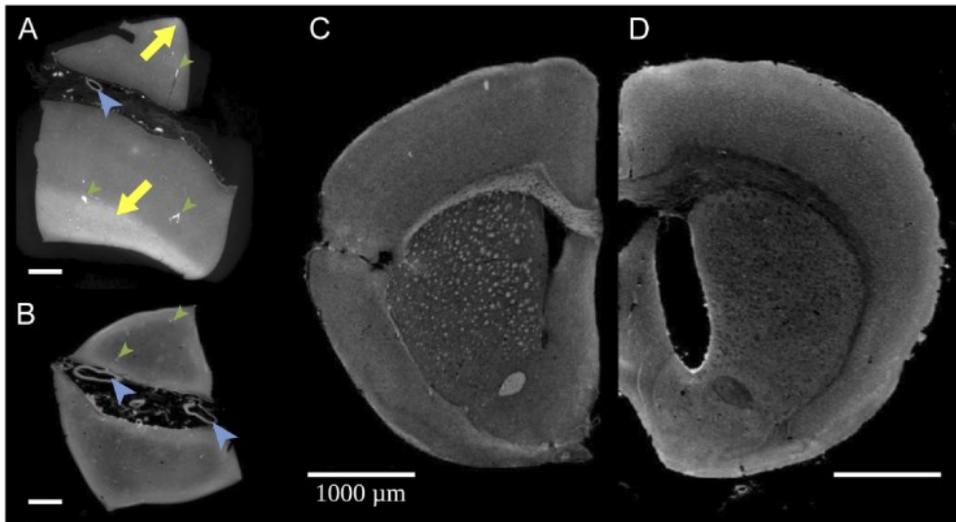


Fig. 3.

Citation
 Matthieu Chourout, Hugo Rositi, Elodie Ong, Violaine Hubert, Alexandre Paccalet, Louis Foucault, Awen Autret, Barbara Fayard, Cécile Olivier, Radu Bolbos, Françoise Peyrin, Claire Crola-da-Silva, David Meyronet, Olivier Raineteau, Hélène Elleaume, Emmanuel Brun, Fabien Chauveau, Marlene Wiart, "Brain virtual histology with X-ray phase-contrast tomography Part I: whole-brain myelin mapping in white-matter injury models," *Biomed. Opt. Express* 13, 1620-1639 (2022); <https://www.osapublishing.org/boe/abstract.cfm?URI=boe-13-3-1620>

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To go further in the analysis of XPCT data, we explored reconstructing the orientation of brain fibers to shed light on brain structural connectivity. A Diffusion Tensor Imaging-like algorithm was used, based on digital gradient analysis to detect fiber orientation, and a custom analysis tool was developed (by NOVITOM). Visual examination of brain-wide connectivity maps showed good agreement between the XPCT-derived maps and MRI-based maps.

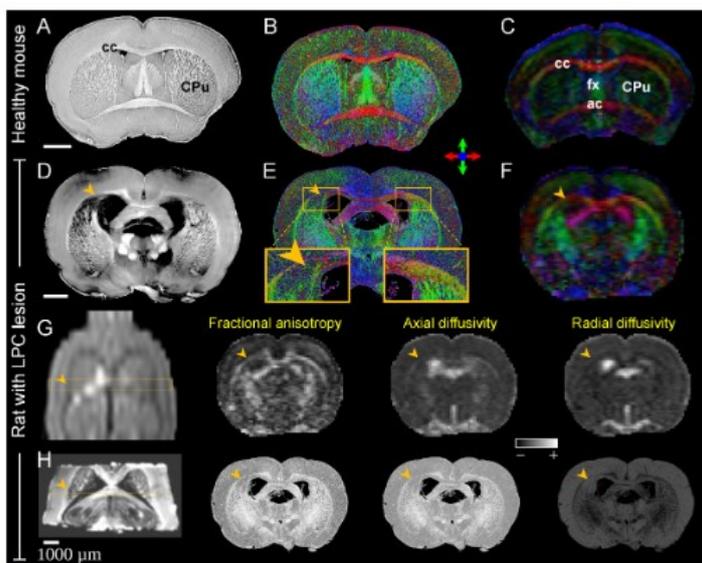


Fig. 6.

Citation
 Matthieu Chourout, Hugo Rositi, Elodie Ong, Violaine Hubert, Alexandre Paccalet, Louis Foucault, Awen Autret, Barbara Fayard, Cécile Olivier, Radu Bolbos, Françoise Peyrin, Claire Crola-da-Silva, David Meyronet, Olivier Raineteau, Hélène Elleaume, Emmanuel Brun, Fabien Chauveau, Marlene Wiart, "Brain virtual histology with X-ray phase-contrast tomography Part I: whole-brain myelin mapping in white-matter injury models," *Biomed. Opt. Express* 13, 1620-1639 (2022); <https://www.osapublishing.org/boe/abstract.cfm?URI=boe-13-3-1620>

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As for the detection of amyloid- β (A β) plaques, we went beyond previous pioneer reports of A β detection with XPCT by :

- i) studying a complete experimental group (10 brains in total vs 1–2 samples);
- ii) comparing 3 different strains (vs. a single one);
- iii) and providing access to multiple morphometric parameters (while previous reports were confined to plaque number, volume and sphericity).

We showed that a new class of 3D parameters, with distribution easily measured on multiple plaques can be obtained, and we provided basic examples of how to handle this set of parameters: e.g., building correlation matrices or searching for spatial patterns.

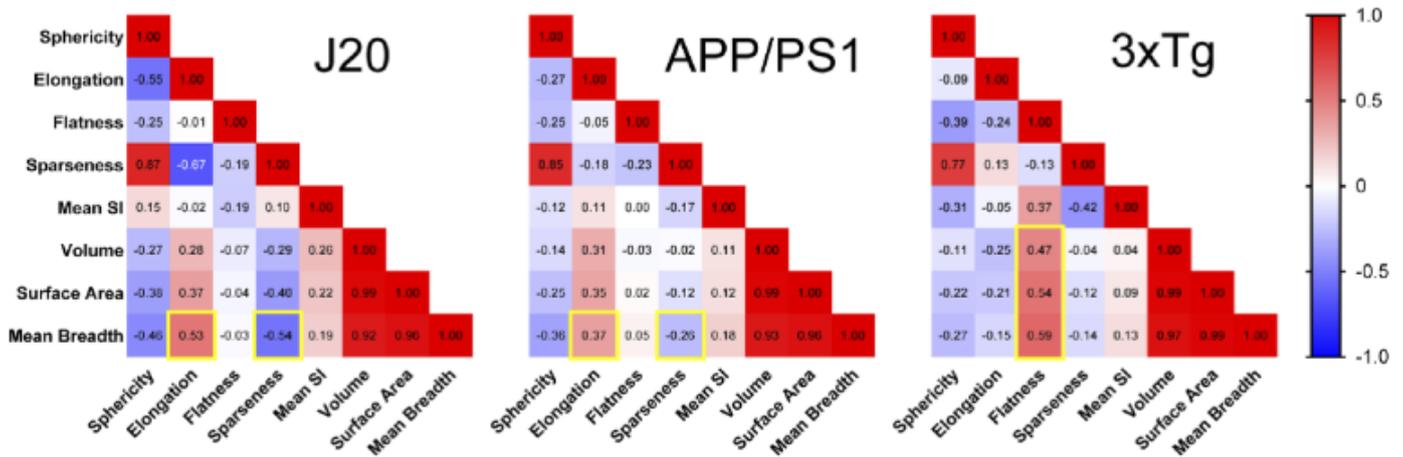


Fig. 6.

Citation

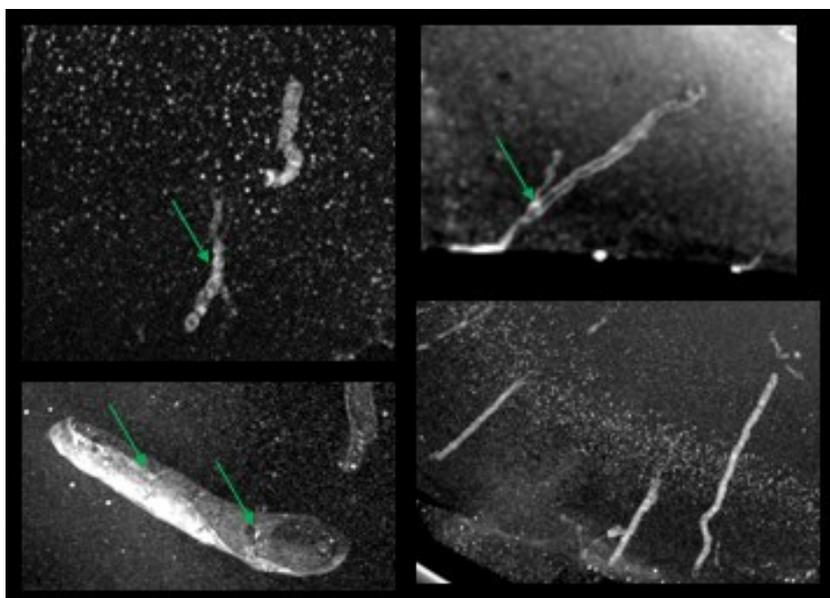
Matthieu Chourout, Margaux Roux, Carlie Boisvert, Coralie Gislard, David Legland, Ignacio Arganda-Carreras, Cécile Olivier, Françoise Peyrin, Hervé Boutin, Nicolas Rama, Thierry Baron, David Meyronet, Emmanuel Brun, Hugo Rositi, Marlène Wiart, Fabien Chauveau, "Brain virtual histology with X-ray phase-contrast tomography Part II: 3D morphologies of amyloid- plaques in Alzheimer's disease models," Biomed. Opt. Express 13, 1640-1653 (2022); <https://www.osapublishing.org/boe/abstract.cfm?URI=boe-13-3-1640>

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Visualization showing the distribution of mean breadth values on APP/PS1 hippocampus <https://opticapublishing.figshare.com/s/b67dc8e839059e885974>

An additional contribution consisted in building and sharing a "biologist-friendly" analysis workflow, by assembling multiple Fiji plugins: segmentation editor, Trainable Weka Segmentation 3D, MorphoLibJ, 3D ImageJ Suite [5]. We were especially interested in the ability of the workflow to correctly detect a significant proportion of A β plaques with very different appearances and morphologies.



We will use these newly developed tools to segment and analyze vascular amyloid deposits which are clearly detected in human brain tissue of familial and sporadic Alzheimer patients (green arrows).

References

- [1] Rositi H, Desestret V, Chauveau F, Cho T-H, Weber L, Ong E, Berner L-P, Olivier C, Langer M, Frindel C, Nighoghossian N, Berthezène Y, Peyrin F, Rousseau D, Wiart M. Fast virtual histology of unstained mouse brains using in-line X-ray phase tomography. In: *10th European Molecular Imaging Meeting (EMIM), Tuebingen, Germany*. **Mar. 2015**. [oral presentation by Marlène Wiart]
- [2] Chourrout M, Rositi H, Ong E, Hubert V, Paccalet A, Foucault L, Autret A, Fayard B, Olivier C, Bolbos R, Peyrin F, Crola-da-Silva C, Meyronet D, Raineteau O, Elleaume H, Brun E, Chauveau F, Wiart M. Brain virtual histology with X-ray phase-contrast tomography. Part I: whole-brain myelin mapping in white-matter injury models, *Biomed. Opt. Express* **2022**:13, 1620-1639. DOI : [10.1364/BOE.438832](https://doi.org/10.1364/BOE.438832)
- [3] Chauveau F, Boutin H, Meyronet D, Rositi H, Olivier C, Elleaume H, Brun E, Peyrin F, Wiart M. Virtual histology of animal and human brains with Alzheimer's disease. In: *13th European Molecular Imaging Meeting (EMIM), San Sebastian, Spain*. **Mar. 2018**. [oral presentation by Fabien Chauveau]
- [4] Chourrout M, Roux M, Boisvert C, Gislard C, Legland D, Arganda-Carreras I, Olivier C, Peyrin F, Boutin H, Rama N, Baron T, Meyronet D, Brun E, Rositi H, Wiart M, Chauveau F. Brain virtual histology with X-ray phase-contrast tomography. Part II: 3D morphologies of amyloid- β plaques in Alzheimer's disease models. *Biomed. Opt. Express* **2022**:13, 1640-1653. DOI : [10.1364/BOE.438890](https://doi.org/10.1364/BOE.438890)
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