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Towards the reconstruction of the mouse brain vascular networks with high- resolution synchrotron radiation X-ray tomographic microscopy

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The formation and progression of several vascular diseases in the brain is accompanied by changes in the vessel micro-structure and morphology. A clear visualisation and an in-depth knowledge of the vascular system is essential for better understanding the pathophysiological mechanisms of neurovascular disorders. Micro-Optical Sectioning Tomography has shown potentials in imaging the vessel network of an entire mouse brain with a voxel resolution of 0.35×0.4×2.0 µm3 [1]. However, available imaging tools are unsuited for non-destructive cerebral mapping of the three-dimensional vascular microstructures. To overcome these difficulties, the brain vasculature architecture is currently documented at 16 µm resolution in micro-Computed Tomography (CT) [2] and about 5.9 µm pixel size with synchrotron-radiation based micro-CT [3]. Within the context of the Human Brain Project (HBP), we aim at using synchrotron radiation X-ray tomographic microscopy at the Swiss Light Source of the Paul Scherrer Institute (Switzerland) as a key technology for reconstructing, in a non-descructive way, the entire vascular system of the mouse brain at 1 µm resolution. During the experimental work, PCO.Edge camera with high efficiency (QE>70%) coupled with 10× objective and filtered white-beam radiation are used to further decrease exposure times. This configuration yields a pixel size of 0.65 µm and an effective resolution of about one micron. Filtered white-beam refers to the polychromatic configuration of the beamline where 95% of the total beam power is filtered out of the beam incident on the sample. The bandwidth of the X-ray beam is narrowed down around a mean energy of 25–30 keV. The exposure time in such conditions is set to 30 ms. The sample is prepared by intravascular filling with consecutive embedding of the tissue, adopting a protocol suggested by [1]. Local CTs are performed for a total of 792 scans in 30 hours scanning time to cover the whole brain volume. In total, 7 TB of datasets are acquired and need to be processed. To address this challenge, we extend the method in order to work on several scans by enabling the use of many machines in parallel, thus allowing the stitching and analysis of such large datasets. At this point, these pioneering efforts are pointing towards new horizons in the investigation of large biological samples with 3D high spatial resolution.

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