ESRF	Experiment title: In-line phase contrast imaging in radiology	Experiment number :MD 94
Beamline:	Date of experiment:	Date of report:
ID17	from: 10.05.2006 to: 13.05.2006	24.07.2006
Shifts: 6	Local contact(s): Dr. A. Bravin	Received at ESRF:
Names and affiliations of applicants (* indicates experimentalists): Francis R. Verdun, Institut universitaire de Radiophysique Appliquée, Lausanne * Pascal Monnin, Institut universitaire de Radiophysique Appliquée, Lausanne * Daniel Gutierrez, Institut universitaire de Radiophysique Appliquée, Lausanne		

Report:

The aim of the experiments was to reveal the potential of the in-line phase contrast imaging of tissue shape samples. Images of metallized and non-metallized casts made of mercox and polyurethane were recorded with a FRELON camera with an effective pixel size of 7.9 μ m. Images were recorded at three defocusing distances of 10 cm (absorption imaging), 80 cm (Fresnel diffraction imaging) and 156 cm (Franunhofer diffraction imaging) with a beam energy of 25 keV.

Samples

The samples imaged were Mercox casts of chick chorioallantoic membrane (CAM) assay of fertilized eggs. The chick CAM were prepared after an incubation of ten days. Chick CAM vessels were prepared as described elsewhere [1]; the systemic vascular architecture was perfused with a freshly prepared solution of Mercox containing 0.1 ml of accelerator per 5 ml of resin. One hour after perfusion, the CAM were transferred to 15% potassium hydroxide solution, for the dissolution of tissue over the course of 3 to 4 weeks. After washing, the casts were dehydrated in ethanol and dried in a vacuum desiccator. One of the samples was sputtered with gold to a thickness of 10 nm and the others were left without a metallic layer.

Results

An example of a chick CAM can be seen in Figure 1. The radiography of a chick CAM kidney made of mercox can is shown in Figure 2. The diameter of the smallest vessels in the radiography is about 20 μ m. Figure 2 was obtained by merging several images to visualize the whole cast. The effect of phase contrast on the fine structure visibility can be seen by comparing the images obtained at the three defocusing distances. The detail of a phase contrast radiography of a chick CAM vessel made of non-metallized polyurethane is shown in Figure 3. The graph next to Figure 3 shows the perpendicular profiles taken across the edge of a vessel. The visibility of the small vessels is dramatically improved by phase contrast.

The images taken at large defocusing distances show the significant edge contrast enhancement due to phase effects. The mercox and polyurethane cams are very weakly absorbing objects, giving a contrast below the limit of 5% that is usually required to detect a low contrast structure in conventional radiology. The images of the Mercox casts show that in-line holography techniques can be used to image very thin structures. Small vessels of 10 to 30 microns are clearly visible. It is to note that without a metallic layer around the sample, nearly no absorption contrast is visible. The phase contrast produces a high resolution two-dimensional image with sufficient contrast of a weakly absorbing material such as resin. The vessels are visible due to the edge enhancement produced by phase modulations at each change of electron density. The particular advantage of this technique is that it provides high quality images without requiring any time consuming (and potentially destructive) sample preparation – such as is required by standard transmission electron microscopy (TEM) or scanning electron microscopy (SEM).

[1] Djonov, V., Schmid, M., Tschanz, S. A., Burri, P. H.: Intussusceptive angiogenesis: its role in embryonic vascular network formation. Circ. Res. **86** (2000) 286-292.

Figures



Figure 1: Example of a chick CAM in a small box



156 cm 80 cm 10 cm Figure 2: Phase contrast radiography of a chick CAM kidney at three defocusing distances



156 cm10 cmIntensity profilesFigure 3: Phase contrast radiography of a chick CAM kidney at three defocusing distances