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Change of Image Quality by Fixation Technique in Phase-contrast X-ray Image

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1 Introduction

Phase contrast X-ray imaging technique for detecting contrast from differences in refractive index, was developed. This technique has approximately 1000 times greater sensitivity than that of the conventional absorption technique [1]. Various biological specimens such as brain, heart, cancer, et.al., could be imaged clearly without contrast agents, however image contrast decreases in high spatial resolution as 0.005mm. To enhance the image contrast, we changed the fixation technique of the biological sample from commonly used formalin to ethanol because the physical property increases density differences against back ground and the effect of dehydration by ethanol increases the soft tissue density.

Here, the quality and degree of contrast enhancement of ethanol-fixed biological specimens such as brain, heart and kidney was compared to that of the formalin-fixed specimens in phase-contrast X-ray images.

2 Experiment

The phase-contrast X-ray computed tomography (CT) system consisted of an asymmetric silicon crystal (220), a two-crystal X-ray interferometer, a phase shifter, and a lens coupling X-ray CCD camera. The X-ray energy was set to 35 keV by the monochromator.

Rats were anesthetized, and cannulation to the apex of left ventricle was carried out surgically for perfusion. First, physiological saline solution was injected from the apex, and the whole blood in the vessel was replaced. Then, 10% formalin or 100% ethanol was perfused for fixation, and the samples were steeped in each fixing solution. This experiment was approved by the Medical Committee for the Use of Animals in Research of the Kitasato University, and it conformed to the guidelines of the American Physiological Society.

3 Results and Discussion

Phase-contrast X-ray CT images of biological specimens fixed by ethanol, showed much better image contrast than that by formalin fixation.

In rat's brain, inner structures such as cerebral cortex, hippocampus, basal ganglion as thalamus, corpus callosum, and mammililiothalamic tract, were depicted clearly by ethanol fixation (Fig.1). The image contrast by ethanol-fixed brain had approximately two-times higher than that by formalin-fixed brain between cortex and white matter [2]. The nerve plexus composed of the white matter in corpus callosum can be extracted well, and 3D structure of white matter could also be reconstructed by image processing software (Realintage, Cybernet) [3].

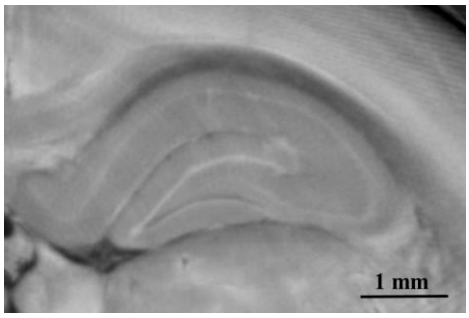

Fig.1. Hippocampus of rat's brain fixed by ethanol

In rat's kidney fixed by ethanol, cortex, tubules in the medulla, and the vessels could be visualized more clearly than that of formalin-fixed tissues [4]. The ethanol fixation technique increased approximately 2.7-3.2 times higher image contrast than that by formalin fixed technique in cortex and outer stripe of the outer medulla, and these density changes could be explained by histological analysis [5].

In rat's heart fixed by ethanol, image quality of myocardium was better than that by formalin fixation. Especially, myocardial three muscle layers such as endocardium, mid-myocardium and epicardium were clearly distinguished in ethanol fixed probably due to separation of each muscle layers. However, the muscle layers cannot be visualized in formalin fixation [6].

Image contrast of various organs of rat in phase-contrast X-ray CT image, has improved significantly by ethanol fixation technique.

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References

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Research Achievements

1. Best Presentation Award in Master Science of Kitasato University (2013)

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