Instrumentation and Technique

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Embryo imaging using phase-contrast X-ray technique

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Introduction

Morphogenetic changes during embryonic period occur dynamically and complicatedly. The analysis of such morphogenetic changes require the visualization of embryonic structures in three-dimensions (3D). Conventional micro x-ray computed tomography and magnetic resonance microscopy cannot provide high-resolution images of soft tissue structures such as embryos. The phase-contrast X-ray imaging detects the x-ray phase-shift through an object and produces images with high spatial resolution without any contrast agents. Using the imaging system set up in the BL-14C [1, 2], we analyzed ten mouse embryos at E10.5-17.5, spanning the period of major organogesis.

This study is the first application of the phase-contrast imaging technique for embryo imaging. This novel imaging technique may be useful for observing surface and inner structures of the embryos in detail.

Images of embryos

Mouse embryos were fixed by formalin, and preserved in 70% ethanol. Sequential images of the embryos were obtained by diffraction-enhanced imaging (DEI) system (Figure 1). Detailed observation of the internal organs was made throughout the early to late stages of mouse development, as well as of the external appearance by surface reconstruction. The developing bone structures did not affect the phase-contrast images (see E17.5 in Figure 1).

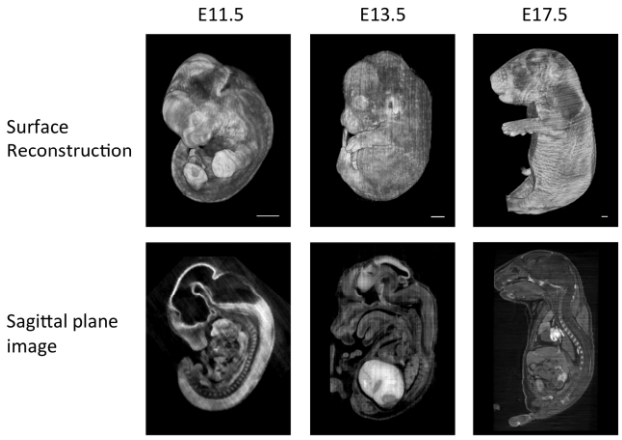

Fig. 1 Sequential images of mouse embryo development. Bars = 1 mm.

The images of embryos were also obtained by X-ray interferometric imaging (XII) system. The comparison of images by XII and DEI systems were shown in Figure 2. Both systems provided fine surface reconstruction and images of the internal structure. Images by the XII system seemed to be better than those of the DEI system for the same embryo, although the scan time of DEI (1 hr) was much shorter than that of XII (4–5 hrs). The image sharpness can be affected by the direction of the rotation of the samples; the axis of rotation was vertical in the DEI system, and horizontal in the XII system. Some samples were not glued directly on the stage but were embedded in agar, which was then fixed on the stage by an adhesive agent. Therefore, small deformation of the agar by gravity may affect the images by the XII system.

These images show that the phase-contrast X-ray CT has a wide enough field and high enough resolution for observation and analyses of morphological changes during embryo development.

References

[1] K. Hyodo, et al., PF Activity Report Part A. 27, 68-69 (2009).

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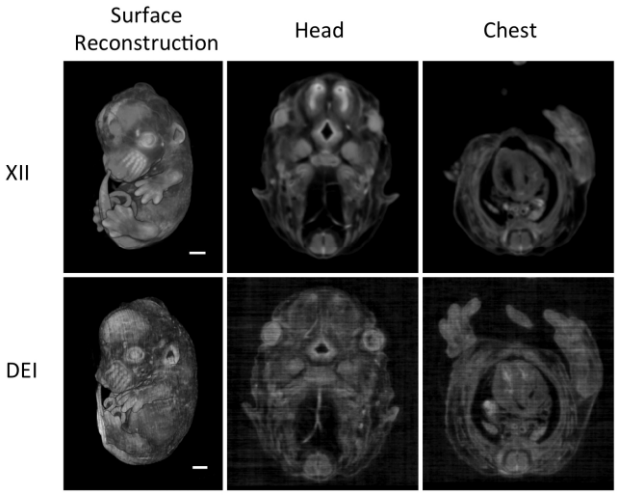

Fig. 2 Images by XII and DEI systems for E13.5 mouse embryo in Fig. 15. Bars = 1mm.

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