

Anatomical and Pathological Analyses of human embryos using phase-contrast X-ray imaging

Shigehito Yamada^{1,2}

¹Congenital Anomaly Research Center, Kyoto University Graduate School of Medicine,
Yoshida-Konoe-cho, Sakyo-ku, Kyoto, 606-8501, Japan

²Human Health Sciences, Kyoto University Graduate School of Medicine,
53 Shogoin-Kawahara-cho, Sakyo-ku, Kyoto, 606-8507, Japan

1 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 human embryos at Carnegie stage (CS) 15-23, spanning the period of major organogenesis.

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

2 Materials

Since 1961, a large number of human conceptuses have been collected in Kyoto University with the collaboration of several hundred obstetricians. In a great majority of the cases, pregnancy was terminated for social reasons during the first trimester of pregnancy (Maternity Protection Law of Japan). The pregnancies were interrupted mainly by dilatation and curettage. Some cases were derived from spontaneous or threatened abortions. Since the attending obstetricians did not examine the aborted materials, the collection of embryos was not biased by their outcome (normal or abnormal, live or dead, etc.), the embryo collection can be considered to be representative of the total intrauterine population in Japan. The embryo collection now comprises over 44,000 specimens, and part of them (~20%) are undamaged well-preserved embryos. When the aborted materials were brought to our laboratory, the embryos were measured, staged, and examined for gross external abnormalities and signs of intrauterine death under a dissecting microscope. The developmental stage of the embryos (Carnegie stage; CS) was determined as described in the previous paper [3].

3 Results and Discussion

Human embryos without major damages were selected suitable for detailed morphological examination. They

were soaked in the formalin solution after fixation by 10% formalin or Bouin's solution. They were transported carefully to the KEK.

From May 2012 to February 2013, 34 scans have been performed and 27 human embryos were imaged (Table 1). Sequential images of the embryos were obtained by X-ray interferometric imaging (XII) system. Fine surface reconstruction and images of the internal structure were provided from the image set. The image sharpness can be affected by the direction of the rotation of the samples; the axis of rotation was horizontal in the XII system. 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.

Table 1: Number of each Carnegie stages

Carnegie stage	Number
15	4
16	5
17	8
18	1
19	1
20	1
21	3
22	2
23	2

The 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, and now the pathological analyses of the images are ongoing.

References

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* shyamada@cac.med.kyoto-u.ac.jp