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Three-dimensional Photoacoustic Imaging of Chicken Embryo Vasculature

光音響イメージングを用いたニワトリ胚の心血管系の可視化

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1. Introduction

Today, various imaging modaities are available for diagnosis of cancer or cardiovascular disease. Especially, computed tomography (CT) may have become the first-line diagnostic method in mordern medical practice in Japan. However, excessive use of CT raised a new problem; side effect of the radiation exposure. Thus, non-invasive and repeatable diagnostic method has been always desired.

Besides conventional ultrasound (US) imaging, photoacoustic (PA) imaging is a non-invasive diagnostic approach using PA effect in which ultrasound is generated by thermal expansion of the material induced by the exposure of nano-second pulsed laser light [1]. Intensity of the PA signal mainly depends on the absorption coefficient of the object. Because the absorption of the laser light with the wavelength around 530 nm is large in red colored object, microvasculature or neovascularization is a good target of PA imaging. Therefore, PA imaging is expected to help diagnosis of cancer with neovascularization.

The purpose of this study is to visualize chicken embryo vasculature with a three-dimensional (3D) PA imaging system.

2. Method

2-1. Photoacoustic imaging

PA imaging is based on photoacoustic effect. When an objective is illuminated to a short pulse laser light, light energy is absorbed by the object and it expands thermally because of local temperature rise. Then, ultrasonic is generated by the thermal expansion and detected by a ultrasonic transducer.

PA signals are dependent on various parameters. They can be described as

$$P_0 = (\beta c^2 / C_p)\mu_a F \quad (1)$$

where P_0 is pressure of thermal expansion, β $[K^{-1}]$ is volumetric thermal expansion coefficient, c is velocity of light, C_p $[J/(Kg.K)]$ is specific heat, μ_a is absorption coefficient of object, F $[J/cm^2]$ is light flux [2].

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2-2. Experimental system

Figure 1 shows the block diagram of the experimental system. This system was controlled by LabVIEW program installed in the PC. The stage driver (Mark-202, Sigma-koki Co Ltd) sent a trigger to a function generator (WF1944B, NF, Inc.) when the x-axis stage (SGSP20-20, Sigma-koki Co Ltd) moved. Then, after the function generator sent a trigger to the semiconductor laser (L11038-02Y, Hamamatsu Photonics K.K.), the laser oscillated with a trigger to the PC. Ultrasonic transducer received PA signals after a laser oscillation, then PA signals were acquired by a digitizer card (DP1400, Acqiris Inc) in the PC with a sampling rate of 1Gsamples/sec.

The semiconductor laser had a wavelength of 532nm with a pulse width of 3 ns. The ultrasonic transducer had a center frequency of 50 MHz and a focal distance of 15mm. The transducer had a 1-mm hole in the center to get through an optical fiber. Therefore, the irradiation of the laser light and reception of the ultrasound were co-axially aligned.

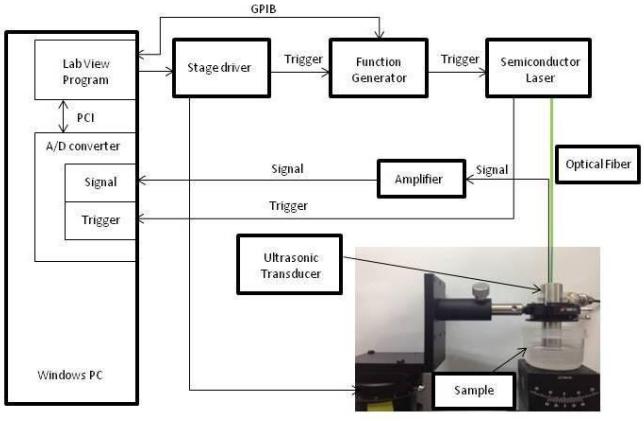

Fig. 1 Block diagram of the experimental system

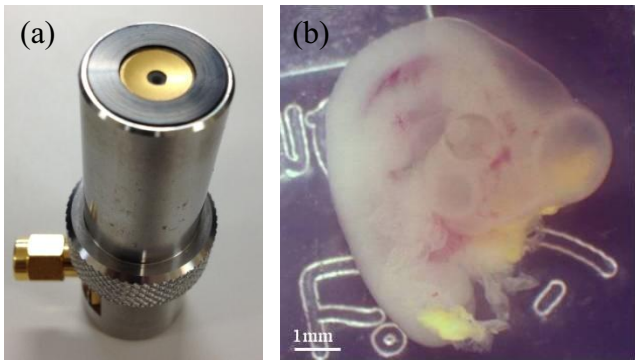

Fig. 2 (a) ultrasonic transducer and (b) optical image of a chicken embryo

Data processing, analysis, and image processing of B-mode images were conducted by MATLAB (The Mathworks Inc). 3D images were reconstructed by Image J (National Institute of Health).

2-3. Chicken embryo

Chicken embryos were chosen as imaging objectives because they were inexpensive, easily obtainable and easy for visualization. Ultrasound and PA imaging were conducted for the identical target after 3 days of incubation when the vasculature was developed. Fig. 2 shows an optical image of the chicken embryo used in the experiment.

3. Results and Discussion

Fig. 3 (a) shows the US image and (b) shows the PA and US merged image. Fig. 3 (c) shows ultrasonic image turned 180 degrees and (d) shows PA and US merged image turned 180 degrees. The B-mode image was 7 mm in width and 3 mm in height. One hundred consecutive B-mode images were acquired at intervals of 70 μm in lateral direction. In these images, US signal is expressed in gray scale and PA signal is expressed in hot scale. Thus, orange parts in Fig. 3 are considered as the vasculature where PA signal was generated.

In Fig. 3, US images show the main body structures including back bones of the chicken embryo and PA images show the great vessels. Moreover, Fig. 3 (c) shows the heart of the embryo in central part of its whole body which cannot be recognized in the optical image of Fig. 2 or ultrasonic images of Fig. 3 (a) or (d). The experiment time was approximately 30 minutes and the red blood cells which were the main source of the PA signal, were circulating in the vasculature during that period. PA intensity would be proportional to the spatial and temporal average distribution of red blood cells.

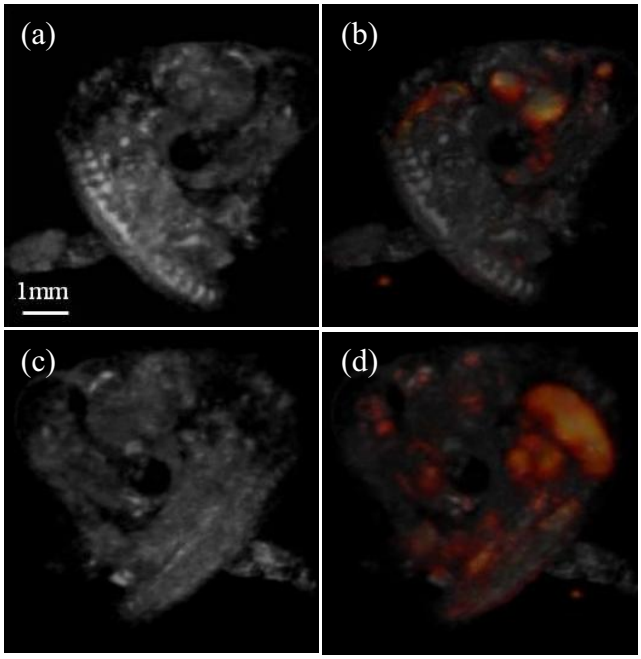

Fig. 3 (a) ultrasonic image, (b) PA image, (c) 180-degree rotated ultrasonic image, and (d) 180-degree rotated PA image of a chicken embryo

4. Conclusion

In the present study, 3D PA imaging successfully visualized the heart and great vessels of the chicken embryo which cannot be visualized by sole US imaging. In other words, combination of US and PA imaging could visualize the distribution of red blood cell, which should be critical information for the diagnoses of various diseases. Moreover, PA imaging is useful for investigation of chicken embryo after it was covered by opaque membrane.

References

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