Transillumination Imaging of Physiological Functions by NIR Light

Yoshinori Taka[1], Yuji Kato[1] and Koichi Shimizu[1]

***Abstract* - With a near-infrared (NIR) light, we can get transillumination image of a living body. Further, we can quantify the physiological change in the body as the change in light attenuation. A fundamental study has been conducted to visualize the functional change inside a living biological body using the NIR light. A technique was developed to visualize the attenuation change occurred in a diffuse scattering medium. Transillumination images are obtained before and after the physiological change. By taking the ratio of transmitted intensity of these two images, we can obtain the spatial distribution of attenuation change while suppressing the effect of scattering. This principle was derived in theoretical analysis and its effectiveness was verified in experiments. To examine the applicability of this principle to a biological body, localized physiological changes were made in the mouse abdomen and the rat brain. The hypoxia in one of the mouse kidneys was visualized selectively from another normal kidney. The local increase in the blood volume was detected in the somatosensory area of a rat brain when its forelimb was electrically stimulated. The blood increase occurred in a symmetrical position with respect to the sagittal plane, when the forelimb of the opposite side was stimulated. Through these experiments, it was found that the changes in the tissue oxygenation and the blood volume could be detected noninvasively and that they are visualized in the transillumination images using the NIR light.**

***Key words* – imaging, transillumination, light, functional imaging, near-infrared, scattering**

I. Introduction

Among the wide spectrum of visible and invisible light, near-infrared (NIR) light is known to have the high transmittance through a biological body [1]. In addition to the high transmission, the important chromophores such as hemoglobin show characteristic absorption spectra in this NIR range [2]. Since the spectrum is sensitive to the oxygenated state of the chromophores, there is the possibility to obtain the functional change inside a living body in a transillumination image.

There have been experimental studies to apply the transillumination method to animal bodies [3-7], but they were not widely used in practice. One of the reasons was the problem of strong scattering of mammalian tissues, which degraded the quality of images considerably [8]. The lack of an appropriate light source and a sensitive detector was another reason. With the recent progress of an optical technology, the feasibility of an optical CT has been pointed [9,10]. However, the problem of the diffuse scattering has remained.

We have conducted the fundamental study on the scattering in dense random media, and verified the effectiveness of the scattering suppression techniques [11-15]. This paper presents the experimental study to verify the feasibility of imaging of the physiological functions using the NIR light.

II. Principle

The absorbance of hemoglobin (Hb) is generally dependent on its redox state except at the isosbestic wavelength around 800 nm. Thus, we can evaluate the change in the tissue oxygenation by measuring the absorption of Hb in the tissue. Further, we can measure the change in the amount of Hb, or the blood volume in tissue by measuring the absorption at this isosbestic wavelength.

This technique is based on the measurement of the transmitted light through a biological body. The light intensity transmitted through a non-scattering medium is given by,

$$I / I_0 = \exp(-\varepsilon CD) \qquad (1),$$

where I_0, I, ε, C and D are the intensities of incident and transmitted light, the absorption coefficient, the concentration and the geometrical thickness of the scattering sample, respectively. Here we define the attenuation of light as the optical density OD, or

$$OD = \log(I_0 / I) \qquad (2).$$

Then it becomes,

$$OD = \varepsilon CD \qquad (3).$$

This is the case when there is no scattering in the medium. If the medium has strong scattering property such as in the case of mammalian tissues, there is no analytical solution to obtain the transmitted light intensity. However, if the scattering characteristics of the medium do not change, it has been shown that the change of the transmitted intensity is proportional to the change in the attenuation parameters in the medium [16], i.e.,

$$\Delta OD = B\Delta(\varepsilon CD) \qquad (4),$$

where B is the differential pathlength factor [16] which represents the increase of the optical pathlength due to the scattering process.

In the proposed technique, we measure the change in the transmitted intensity due to the changes in the tissue oxygenation and in the blood volume. When the oxygenation of the tissue (oxygenation of blood in the tissue, strictly

[1] Department of Bioengineering,
Graduate School of Engineering,
Hokkaido University, Sapporo, 060, Japan
shimizu@bme.eng.hokudai.ac.jp

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speaking) changes without the change in blood volume (C), the difference between before and after the change is given by,

$$OD_2 - OD_1 = \log(I_1 / I_2) = (\varepsilon_2 - \varepsilon_1)BCD \quad (5),$$

where the subscripts 1 and 2 indicate before and after the change, respectively. This means that the difference of the optical density is linearly proportional to the change in the absorption coefficient in the scattering medium. Therefore, we can visualize the spatial distribution of this absorption change by calculating the ΔOD at each pixel of the transmitted image.

When the degree of Hb oxygenation does not change, or if we use the light of the isosbestic wavelength, the change in the optical distance due to the blood volume is given by,

$$OD_2 - OD_1 = \log(I_1 / I_2) = (C_2 - C_1)B\varepsilon D \quad (6).$$

This means that we can visualize the distribution of blood volume change in the transillumination image.

III. Method

Fig.1 shows the outline of an experimental setup schematically. A light source of infrared light was placed below a rat (Wistar, 100-150 g). The transmitted light across the rat was focused on a CCD camera placed above the rat. The image signal from the CCD camera was recorded in a video recorder, and processed by an image processor controlled by a microcomputer. As the light source, laser diodes and a Ti:Sapphire laser were used. The optical power at the incident point of the rat surface was about 100 mW. The hair of the rat surface was shaved off to eliminate unnecessary scattering and to simulate the condition of the human skin. To check the vital sign of a rat, an ECG was kept monitored throughout the experiment.

IV. Imaging of Internal Structure

Fig.2 shows the transillumination image of a mouse abdomen. The light source was a laser diode (801 nm wavelength). Without any contrast media, the major structure could be observed such as the intestines, the spleen and the bladder. In the moving image, the peristaltic movement of intestines was clearly observed in a real time.

Fig.3 shows the transillumination image of a rat head and the position of the image. A main structure of the brain could be seen through the skin and the skull. We could see the sagittal vein running between the left and the right cerebral hemispheres and the transverse vein running between the cerebrum and the cerebellum. Further, the large olfactory bulb which is characteristic of a rat could be observed as well.

These results show that the imaging of internal structure is possible to some extent, and that the image provides the information different from that of X-rays.

V. Functional Imaging

The significance of the optical trans-body imaging is the capability of imaging the internal physiological functions. First, the feasibility to detect the internal functional change was examined. As the functional change, one of the mouse kidneys was made hypoxic. The blood circulation of the right kidney was blocked by tying the renal arteries and veins to make the kidney hypoxic. As a light source, Ti:Sapphire laser (775 nm wavelength) was used.

In the transillumination image itself, it was difficult to

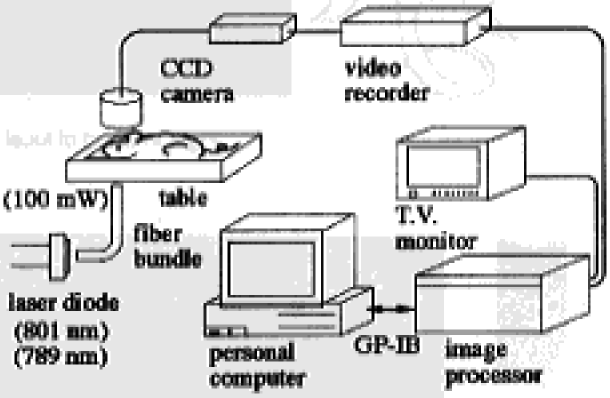

Fig.1 Experimental system for transillumination imaging.

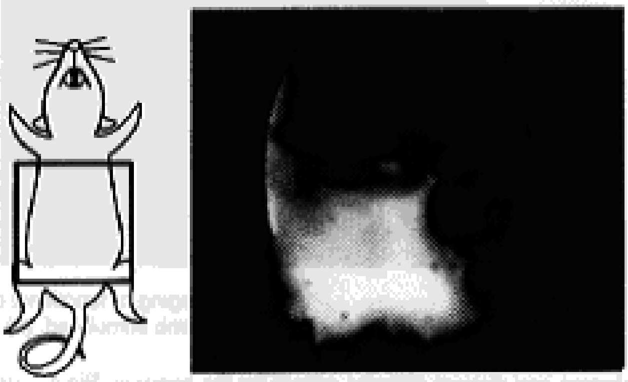
Fig.2 Transillumination image of mouse abdomen.

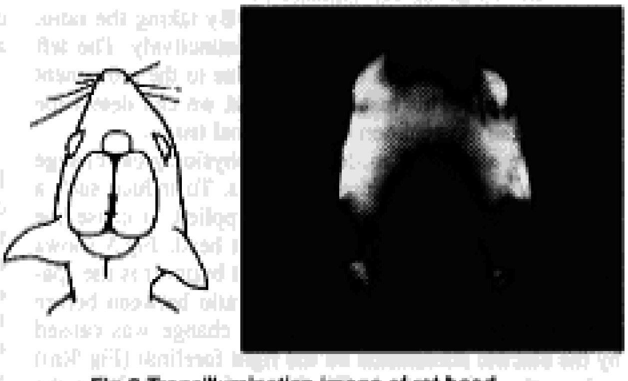
Fig.3 Transillumination image of rat head.

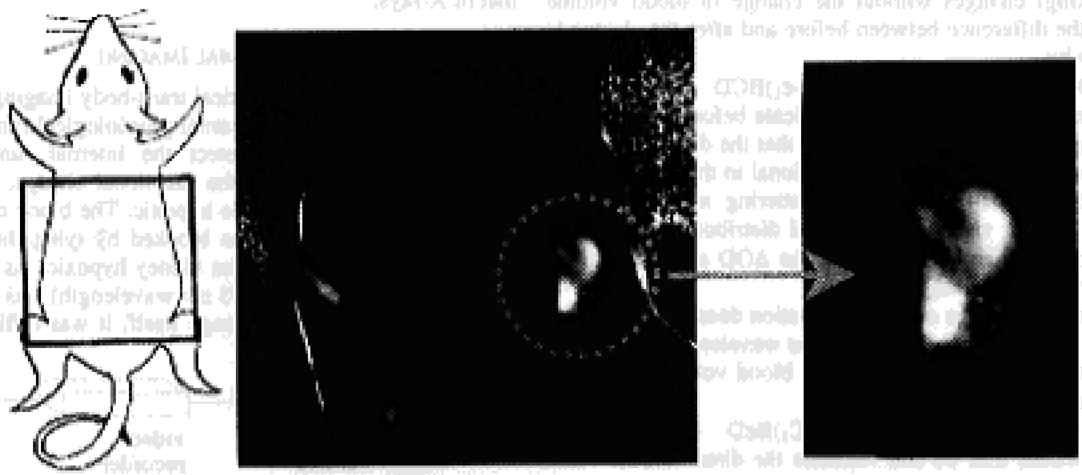

Fig.4 Imaging of local hypoxic part in the body

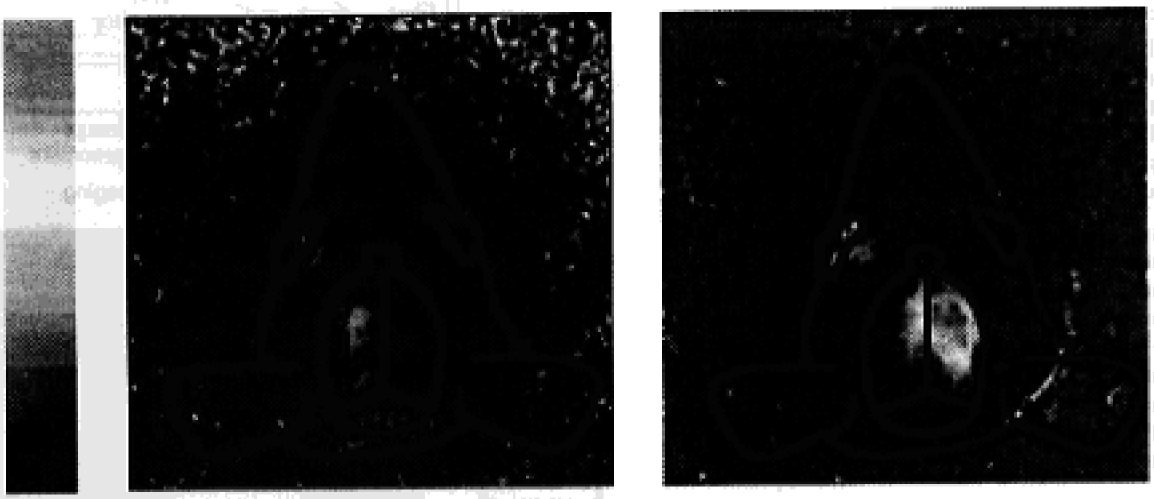

Fig.5 Imaging of functional change in rat brain:
(a) right forelimb stimulated, (b) left forelimb stimulated.

observe any distinct changes caused by the hypoxia. Fig.4 shows the image of the transmittance ratio between the images before and after the hypoxia. By taking the ratio, the part of local hypoxia appeared distinctively. The left bright area seemed to be the artifact due to the movement of the intestine. This result shows that we can detect the change in the oxygenation of the internal tissue.

Next, we attempted to detect the physiological change caused by natural biological functions. To induce such a change, a sensory stimulation was applied to cause the change in blood circulation in the rat head. Fig.5 shows the results of functional imaging of rat brain. It is the spatial distribution of the transmittance ratio between before and after the functional change. The change was caused by the electric stimulation on the right forelimb (Fig.5(a)) and on the left forelimb (Fig.5(b)) of the rat. Since the laser diode of the isosbestic wavelength (801 nm) was used as a light source, the image corresponds to the distribution of the blood volume change. The localized increase of blood volume was well detected in the somato-sensory area of the rat brain.

VI. Conclusions

With the view toward the noninvasive imaging of biological functions with light, a fundamental study has been conducted. Using a near-infrared light, the transillumination images of a mouse abdomen and a rat brain were obtained. The physiological changes such as local hypoxia could be visualized in the transillumination images. It was found that the changes in the blood volume and in the tissue oxygenation could be detected using multiple wavelengths of light. In this way, the spatial distribution of these changes could be visualized in the transillumination images noninvasively. Through this study, the feasibility of optical trans-body imaging of physiological functions was verified.

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