# Image Restoration of Fluorescent Source at Unknown Depth in Diffuse Medium

Koichi Shimizu, Ruriko Koizumi, Hidenobu Itagaki and Yuji Kato

Graduate School of Information Science and Technology, Hokkaido University, Sapporo, Japan.

## **Abstract**

The transcutaneous image of fluorescent source in an animal body is blurred due to the strong scattering of intervening tissue. To restore the clear image of a fluorescent source, the deconvolution with a point spread function (PSF) is useful. We derived the PSF theoretically from the diffusion equation. To use the PSF, the depth of the fluorescent source is essential. A technique was devised to estimate the depth of the fluorescent source in the deconvolution process. The effectiveness of the proposed technique was verified in the experiment with a relatively thick fluorescent source.

## 1. Introduction

The importance of fluorescent imaging in research and clinical practice has been widely recognized. When we excite the fluorescent source inside an animal body, we can see the spatial distribution of the source through the intervening tissue. In many cases, the image is strongly blurred due to the diffuse scattering of animal tissue. If we can characterize the blurring in terms of the point spread function (PSF), we can obtain a clear image of the fluorescent source by the deconvolution with the PSF. We have derived the PSF theoretically by applying the diffuse approximation to the equation of transfer [1]. The effectiveness of this technique was verified in animal experiments.

However, the PSF was given as a function of the source depth. Thus, the applicability of this technique has been largely restricted, if we do not know the source depth. To solve this problem, we have developed a new technique to obtain a clear image of fluorescent source without knowing the depth of the source in the body. The effectiveness of the proposed technique was tested with a relatively thick fluorescent source.

# 2. Estimation of source depth

Assume that there is a fluorescent source at the depths of  $d_f$  and  $d_b$  from the front and back surfaces, respectively. The outer thickness of the scattering medium  $d_t = d_f + d_b$  can be measured easily. Figure 1 shows their relation. The images of the fluorescent

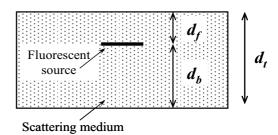


Fig.1 Depths of fluorescent source from front and back sides.

source from the front side  $f_f(x,y)$  and from the backside  $f_b(x,y)$  were obtained with a cooled CCD camera. These images were blurred due to the diffuse scattering of the medium around the fluorescent source.

The image processing based on the deconvolution with the PSF was applied to the blurred images. This deconvolution process is repeated using the PSF's with different depths. We obtained a collection of improved images  $\{g_f(x,y)\}$  and  $\{g_b(x,y)\}$  for the original images  $f_f(x,y)$  and  $f_b(x,y)$ , respectively. Among the combinations of  $g_f(x,y)$  and  $g_b(x,y)$ , the pair that satisfies the condition of  $d_f + d_b = d_t$  were compared. Among many possible combinations, the case in which the similarity of  $g_f(x,y)$  and  $g_b(x,y)$  is the highest should provide us the true depths  $d_f$  and  $d_b$ .

# 3. Experimental test

The effectiveness of the above principle was tested in an experiment. Figure 2 shows the experimental setup. As a fluorescent source, ICG was mixed in the epoxy resin in the shape of rectangular parallelepiped. The size of the fluorescent source was  $10x10x2.5 \text{ mm}^3$ . It was placed in a scattering medium made of Intralipid suspended in agar ( $\mu_s$  '=1.0/mm,  $\mu_a$ =0.1/mm) of 15 mm thickness. The depth of the top and the bottom faces of the fluorescent source were 7.5 mm and 5 mm from the top and the bottom of the scattering agar, respectively.

The light for excitation (765 nm) was illuminated on the front surface of the scattering medium, and the fluorescent image was taken with the cooled CCD

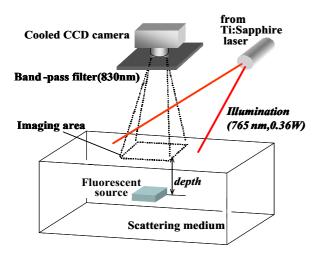


Fig.2 Experimental setup.

camera through a band-pass filter (830 nm). Another image was taken in the same manner with the scattering medium upside down.

Figure 3 shows the images  $f_f(x,y)$  and  $f_b(x,y)$ . Since the fluorescent source is deeper in the case of  $f_f(x,y)$ , the image was blurred more than the case of  $f_b(x,y)$ . Figure 4 shows the examples of the result of deconvolution with the PSF's of different depths. Each pair of the depths satisfies the condition of  $d_f+d_b=15$  mm. Figure 5 shows the correlation between the two images which satisfy this condition. From this curve, we can estimate the depth of the fluorescent source as 9 mm. This is close to the center depth of the fluorescent source.

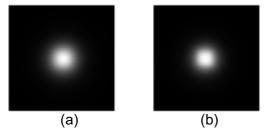


Fig.3 Fluorescent images in scattering medium: (a) front side, (b) back side.

# 4. Conclusions

The transcutaneous image of fluorescent source in an animal body is blurred due to the strong scattering of intervening tissue. A clear image of the fluorescent source can be restored by the deconvolution with a PSF. However, this technique was applicable only when the depth of the fluorescent source was known, a priori. We have devised a technique to estimate the depth of the fluorescent source in the same process of the deconvolution. In an experiment, the effectiveness of the proposed technique was verified.

Further investigation is required to examine the practical usefulness of this technique for the transcutaneous fluorescent image in the animal body.

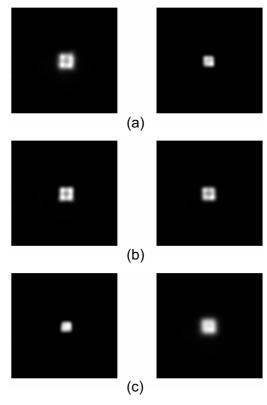


Fig.4 Results of deconvolution with PSF's of;

- (a) 7mm from front and 8mm from back,
- (b) 9mm from front and 6mm from back,
- (c) 11mm from front and 4mm from back.

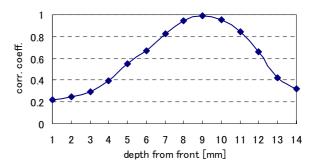


Fig.5 Correlation of two images from front and back sides.

#### Reference

[1] K. Shimizu *et. al.*, Appl. Opt. **44**, 2154-2161 (2005).