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Estimation Method of Fluorescent Donaldson Matrices Based on Multispectral Imaging Data

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Abstract

This paper proposes a compact and reliable method to estimate the bispectral Donaldson matrices of fluorescent objects by using multispectral imaging data. We suppose that an image acquisition system allows multiple illuminant projections to the object surface and multiple response channels in the visible range. The Donaldson matrix is modeled as a two-dimensional array with the excitation range (350, 700 nm) and the reflection and emission ranges (400, 700 nm). The observation model is described using the spectral sensitivities of a camera and the spectral functions of reflectance, emission, and excitation. The problem of estimating the spectral functions is formulated as a least squares problem to minimize the residual error of the observations and the roughness of the spectral functions. An iterative algorithm is developed to obtain the optimal estimates of the whole spectral functions. The performance of the proposed method is examined in simulation experiments using multispectral imaging data in detail.

Introduction

The fluorescent characteristics are well described in terms of the bispectral radiance factor, which is a function of two wavelength variables: the excitation wavelength of incident light and the emission and reflection wavelength [1]. A discrete representation of the bispectral radiance factor is called a Donaldson matrix [2]. This matrix can be measured using two monochromators or one monochromater (spectral imasing device) with many short-wavelength cutout filters [3]. However, these measurement method were inconvenient and impractical.

Several approaches were proposed for estimating the Donaldson matrices using an imaging system without using filters. Fu et al. [4] tried to estimate the reflectance spectrum and the relative spectrum of emission and absorption in the visible range (400, 700 nm) using an RGB color camera data. Blasinski et al. [5] used the Alternating Direction Method of Multipliers (ADMM) approach to estimate the reflectance spectrum and the absolute spectra of emission and absorption in two fluorophores based on an imaging system consisting of many narrowband light sources and transmission filters. Suo et al. [6] described an imaging system that used programmable spectral filters placed on both sides of light source and camera. In the above approaches, the Donaldson matrices were constructed in a visible wavelength of 400 nm and more.

We note that the fluorescent excitation occurs in ultraviolet wavelengths, and natural light includes illuminant spectral power distribution in this range. Tominaga et al. [7-8] presented approaches using a spectral imaging system where the Donaldson matrices were constructed on the broader wavelength range over ultraviolet and visible wavelengths. One was a graphical approach, where two illuminants were projected onto an object surface, and the Donaldson matrix estimation was based on a spectral difference between two radiance factors. Another was a numerical approach, where multiple illuminants were projected sequentially onto the object, and the spectral functions were estimated by iterative calculation at each wavelength. However, as well known, spectral imaging has an essential disadvantage that it requires expensive equipment and time-consuming.

The present paper proposes a compact and reliable method to estimate the bispectral Donaldson matrices using multispectral imaging data. We suppose a general image acquisition system that allows multiple illuminant projections to the object surface and multiple response channels in the visible range. The Donaldson matrices are modeled as a two-dimensional array where the excitation range is defined in a 350 to 700 nm wavelength range, and the reflection/emission range is defined in a 400 to 700 nm wavelength range. The observation model is described using the spectral sensitivities of a camera and the spectral functions of reflectance, emission, and excitation constructing the Donaldson matrices. The estimation problem is solved as a least squared problem to minimize the residual error of the observations acquired by a multispectral imaging system and the roughness of the spectral functions.

Bispectral Fluorescence Modeling

A Donaldson matrix $D(\lambda_{em},\lambda_{ex})$ is represented as a two-variable function of the excitation wavelength λ_{em} and the emission/reflection wavelength λ_{ex}. The excitation wavelength for all fluorescent materials starts from about 330 to 350 nm. In this paper, the excitation range is set at $350 \le \lambda_{ex} \le 700$. On the other hand, since we suppose an imaging system operating in a visible wavelength range, the emission/reflection range is set at $400 \le \lambda_{em} \le 700$. The Donaldson matrix is decomposed into two components of the reflected radiance factor $D_R(\lambda_{em},\lambda_{ex})$ by light reflection and the luminescent radiance factor $D_L(\lambda_{em},\lambda_{ex})$ by fluorescent emission. The matrix $D_R(\lambda_{em},\lambda_{ex})$ is diagonal and has values at $\lambda_{em} = \lambda_{ex}$ which corresponds to surface-spectral reflectance $S(\lambda)$. A fluorescent object usually contains a single fluorescent chemical compound. The luminescent radiance factor is separated into a multiplication of the excitation spectrum and the excitation spectrum $D_L(\lambda_{em},\lambda_{ex}) = \alpha(\lambda_{em})\beta(\lambda_{ex})$. In the multiplication of $\alpha(\lambda_{em})$ and $\beta(\lambda_{ex})$, one of two spectra can be arbitrarily rescaled. Therefore, we assume that the excitation spectrum is normalized $\int_{350}^{700} \beta(\lambda_{ex}) d\lambda_{ex} = 1$.

A discrete form of the Donaldson matrix with the above properties is represented in an (n-by-m) matrix as

$$\mathbf{D} = \mathbf{D}_{\mathrm{R}} + \mathbf{D}_{\mathrm{L}}$$

$$= \begin{bmatrix} \alpha_1\beta_1 & \cdots & \alpha_1\beta_{m-n} & s_1 & 0 & \cdots & 0 \\ \alpha_2\beta_1 & & \alpha_2\beta_{m-n} & \alpha_2\beta_{m-n+1} & s_2 & \ddots & \vdots \\ \vdots & & \vdots & \vdots & \ddots & \ddots & 0 \\ \alpha_n\beta_1 & \cdots & \alpha_n\beta_{m-n} & \alpha_N\beta_{m-n+1} & \cdots & \alpha_n\beta_{m-1} & s_n \end{bmatrix}, \quad (1)$$

where s_i (i=1, 2, .., n), α_i (i=1, 2, .., n), and β_i (i=1, 2, .., m-1) express, respectively, the discrete spectral representations of reflectance $s(\lambda_{em})$, emission $\alpha(\lambda_{em})$, and excitation $\beta(\lambda_{ex})$. When the spectral functions are sampled in equal wavelength intervals of 5 nm, the Donaldson matrix is rewritten with m=71 and n=61.

Observation Model

Let us suppose a multispectral camera with M channels in the visible wavelength range. The camera outputs z_k (k=1,2,.., M) are described as

$$z_k = \int_{400}^{700} y(\lambda_{em}) R_k(\lambda_{em}) d\lambda_{em}, \qquad (k = 1,2,..,M), \tag{2}$$

where $y(\lambda_{em})$ is the spectrum observed from a target object and $R_k(\lambda_{em})$ (k=1,2,…,M) are the spectral sensitivity functions of the camera. The target object is a fluorescent object with a matt surface without specularity. Let $E(\lambda)$ be the illuminant spectrum of a light source in the wavelength range (350, 700 nm). The spectral radiances observed from the surface under this illuminant are described as a sum of the diffuse reflection component and the luminescent component as follows:

$$\begin{aligned} y(\lambda_{em}) &= S(\lambda_{em})E(\lambda_{em}) + \alpha(\lambda_{em}) \int_{350}^{\lambda_{em}} \beta(\lambda_{ex})E(\lambda_{ex})d\lambda_{ex} \\ &= S(\lambda_{em})E(\lambda_{em}) + \alpha(\lambda_{em})C(\lambda_{em}), \end{aligned} \tag{3}$$

where

$$C(\lambda_{em}) = \int_{350}^{\lambda_{em}} \beta(\lambda_{ex})E(\lambda_{ex})d\lambda_{ex}. \tag{4}$$

Suppose that the same object is observed under N different light sources. We define several matrices for the purpose of discrete representation of the model as follows:

$$\mathbf{z}_i = \begin{bmatrix} z_{i1} \\ z_{i2} \\ \vdots \\ z_{iM} \end{bmatrix}, \mathbf{Z} = \begin{bmatrix} \mathbf{z}_1 \\ \mathbf{z}_2 \\ \vdots \\ \mathbf{z}_N \end{bmatrix}, \mathbf{y}_i = \begin{bmatrix} y_i(\lambda_1) \\ y_i(\lambda_2) \\ \vdots \\ y_i(\lambda_n) \end{bmatrix}, \mathbf{Y} = \begin{bmatrix} \mathbf{y}_1 \\ \mathbf{y}_2 \\ \vdots \\ \mathbf{y}_N \end{bmatrix},$$

$$\mathbf{s} = \begin{bmatrix} S(\lambda_1) \\ S(\lambda_2) \\ \vdots \\ S(\lambda_n) \end{bmatrix}, \boldsymbol{\alpha} = \begin{bmatrix} \alpha(\lambda_1) \\ \alpha(\lambda_2) \\ \vdots \\ \alpha(\lambda_n) \end{bmatrix}, \boldsymbol{\beta} = \begin{bmatrix} \beta(\lambda_1) \\ \beta(\lambda_2) \\ \vdots \\ \beta(\lambda_m) \end{bmatrix}, \tag{5}$$

$$\mathbf{e} = \begin{bmatrix} E(\lambda_1) \\ E(\lambda_2) \\ \vdots \\ E(\lambda_m) \end{bmatrix}, \mathbf{C} = \begin{bmatrix} C(\lambda_1) \\ C(\lambda_2) \\ \vdots \\ C(\lambda_n) \end{bmatrix},$$

where $\mathbf{z}_i$ is an M-dimennsional column vector of the camera outpus under i-th illuminant, $\mathbf{Z}$ is an MN-dimensional column vector for all N light sources, $\mathbf{y}_i$ is an n-dimensional radiance vector under i-th illuminant, and $\mathbf{Y}$ is an nN-dimensional column vector for all N light sources. We should note the wavelength ranges in Eq.(5). The vectors $\mathbf{y}_i$, $\mathbf{s}$, $\boldsymbol{\alpha}$, and $\mathbf{C}$ are defined at wavelengths of 400 to 700 nm such as (400, 405, 410, ⋯, 700), and the vectors $\boldsymbol{\beta}$ and $\mathbf{e}$ are sampled in the same intervals as the above but in the wider range of 350 to 700 nm such as (350, 355, ⋯ 395, 400, 405, ⋯, 700). The model equation (3) is then described in matrix form as follows:

$$\mathbf{Y} = \begin{bmatrix} \mathrm{diag}(\mathbf{e}_1) \\ \mathrm{diag}(\mathbf{e}_2) \\ \vdots \\ \mathrm{diag}(\mathbf{e}_N) \end{bmatrix} \mathbf{s} + \begin{bmatrix} \mathrm{diag}(\mathbf{C}_1) \\ \mathrm{diag}(\mathbf{C}_2) \\ \vdots \\ \mathrm{diag}(\mathbf{C}_N) \end{bmatrix} \boldsymbol{\alpha}, \tag{6}$$

where the symbol diag(**a**) denotes an (n-by-n) diagonal matrix constructed with the elements of **a**.

When we apply the finite dimensional linear model to the spectral functions $S(\lambda_{em})$ and $\alpha(\lambda_{em})$, we have

$$S(\lambda) = \sum_{i=1}^{K} w_{Si}B_{Si}(\lambda), \quad \alpha(\lambda) = \sum_{i=1}^{L} w_{\alpha i}B_{\alpha i}(\lambda), \tag{7}$$

where $B_{si}(\lambda)$ and $B_{\alpha i}(\lambda)$ are the basis functions for the spectral reflectance and the emission spectrum, and w_{si} and $w_{\alpha i}$ are the weighting coefficients. Equivalently (7) is represented in matrix form using column vectors as

$$\begin{aligned} \mathbf{s} &= [\mathbf{b}_{s1}, \mathbf{b}_{s2}, \cdots, \mathbf{b}_{sK}]\mathbf{w}_s \equiv \mathbf{B}_s\mathbf{w}_s, \\ \boldsymbol{\alpha} &= [\mathbf{b}_{\alpha 1}, \mathbf{b}_{\alpha 2}, \cdots, \mathbf{b}_{\alpha L}]\mathbf{w}_\alpha \equiv \mathbf{B}_\alpha\mathbf{w}_\alpha \end{aligned} \tag{8}$$

With this linear model expression, the model equation is written as

$$\mathbf{Y} = \begin{bmatrix} \mathbf{G}_1 & \mathbf{F}_1 \\ \mathbf{G}_2 & \mathbf{F}_2 \\ \vdots & \vdots \\ \mathbf{G}_N & \mathbf{F}_N \end{bmatrix} \begin{bmatrix} \mathbf{w}_s \\ \mathbf{w}_\alpha \end{bmatrix}, \tag{9}$$

where

$$\mathbf{G}_i = \mathrm{diag}(\mathbf{e}_i)\mathbf{B}_s, \quad \mathbf{F}_i = \mathrm{diag}(\mathbf{C}_i)\mathbf{B}_\alpha, \quad (i = 1,2,...,N) \tag{10}$$

The whole observations including the camera outputs are represented as

$$\mathbf{Z} = \mathbf{R}'\mathbf{Y}, \tag{11}$$

where $\mathbf{R}'$ is an (NM-by-Nn) matrix defined using the (M-by-n) sensitivity matrix $\mathbf{R}$ for M camera channels as

$$\mathbf{R}' = \begin{bmatrix} \mathbf{R} & \mathbf{0} & \cdots & \mathbf{0} \\ \mathbf{0} & \mathbf{R} & \ddots & \mathbf{0} \\ \vdots & \ddots & \ddots & \mathbf{0} \\ \mathbf{0} & \cdots & \mathbf{0} & \mathbf{R} \end{bmatrix}. \tag{12}$$

Estimation Algorithm

Our purpose in the estimation problem is to estimate $\mathbf{w}_s$ and $\mathbf{w}_\alpha$ from a set of the camera outputs $\{\mathbf{z}_i\}$ observed under M light sources. To solve this problem, we consider minimizing the residual error by using the least squared method, and also the smoothness of the spectral reflectance and the emission spectrum. Then performance index (a cost function) is described as

$$\begin{aligned} L &= \left\| \mathbf{R}'\mathbf{Y} - \mathbf{R}' \begin{bmatrix} \mathbf{G}_1 & \mathbf{F}_1 \\ \mathbf{G}_2 & \mathbf{F}_2 \\ \vdots & \vdots \\ \mathbf{G}_N & \mathbf{F}_N \end{bmatrix} \begin{bmatrix} \mathbf{w}_s \\ \mathbf{w}_\alpha \end{bmatrix} \right\|^2 + \mu_s \|\nabla\mathbf{s}\|^2 + \mu_\alpha \|\nabla\boldsymbol{\alpha}\|^2 \\ &= \left\| \mathbf{Z} - \mathbf{R}'[\mathbf{G} \quad \mathbf{F}] \begin{bmatrix} \mathbf{w}_s \\ \mathbf{w}_\alpha \end{bmatrix} \right\|^2 + \mu_s \|(\mathbf{I} - \mathbf{J})\mathbf{B}_s\mathbf{w}_s\|^2 + \mu_\alpha \|(\mathbf{I} - \mathbf{J})\mathbf{B}_\alpha\mathbf{w}_\alpha\|^2 \\ &\equiv \left\| \mathbf{Z} - \mathbf{R}'[\mathbf{G} \quad \mathbf{F}] \begin{bmatrix} \mathbf{w}_s \\ \mathbf{w}_\alpha \end{bmatrix} \right\|^2 + \mu_s \|\mathbf{B}'_s\mathbf{w}_s\|^2 + \mu_\alpha \|\mathbf{B}'_\alpha\mathbf{w}_\alpha\|^2, \end{aligned} \tag{13}$$

where μ_s and μ_α are weighting parameters for the smoothness, and the symbol ∇ is an operator making differences between adjacent elements in a vector. Let us define (n-by-n) matrices **I** and **J** for the differentiation operator as

$$\mathbf{I}=\begin{bmatrix}0 & 0 & \cdots & 0\\ 0 & 1 & \ddots & 0\\ \vdots & \ddots & \ddots & 0\\ 0 & \cdots & 0 & 1\end{bmatrix},\qquad \mathbf{J}=\begin{bmatrix}0 & 0 & \cdots & 0\\ 1 & 0 & \ddots & 0\\ \vdots & \ddots & \ddots & 0\\ 0 & \cdots & 1 & 0\end{bmatrix}. \qquad (14)$$

The estimates $\mathbf{w}_s$ and $\mathbf{w}_\alpha$ to minimize the cost function L are given as a solution for the following set of linear equations

$$\begin{bmatrix}(\mathbf{R'G})^T(\mathbf{R'G})+\mu_s\mathbf{B'}_s{}^T\mathbf{B'}_s & (\mathbf{R'G})^T(\mathbf{R'F})\\ (\mathbf{R'F})^T(\mathbf{R'G}) & (\mathbf{R'F})^T(\mathbf{R'F})+\mu_\alpha\mathbf{B'}_\alpha{}^T\mathbf{B'}_\partial\end{bmatrix}\begin{bmatrix}\mathbf{w}_s\\ \mathbf{w}_\alpha\end{bmatrix}.$$

$$=\begin{bmatrix}(\mathbf{RG})^T\\ (\mathbf{RF})^T\end{bmatrix}\mathbf{Z} \qquad (15)$$

The estimates of $S(\lambda)$ and $\alpha(\lambda)$ are obtained with the solutions $\hat{\mathbf{w}}_s$ and $\hat{\mathbf{w}}_\alpha$ in the form

$$\hat{\mathbf{s}}=\mathbf{B}_s\hat{\mathbf{w}}_s,\quad \hat{\boldsymbol{\alpha}}=\mathbf{B}_\alpha\hat{\mathbf{w}}_\alpha \qquad (16)$$

On the other hand, the excitation spectrum $\beta(\lambda)$ can be estimated based on a physical model describing a relationship between the excitation spectrum and the reflection spectrum

$$\beta(\lambda)=Q(\lambda)(1-S(\lambda)), \qquad (17)$$

where $Q(\lambda)(\lambda\geq 350)$ is the luminescence efficiency (see [6]). For the limited wavelength range $400\leq\lambda\leq 700$, Eq.(17) is represented in a matrix form as

$$\boldsymbol{\beta}=\mathbf{Q}(1-\mathbf{s}). \qquad (18)$$

where **Q** is a diagonal matrix with elements $Q(\lambda_1), Q(\lambda_2)$, ..., $Q(\lambda_n)$, and **1** denotes an n-dimensional column vector with elements 1. Since the wavelength ranges of excitation and reflection are $350\leq\lambda\leq 700$ and $400\leq\lambda\leq 700$, respectively, the relationship in Eq.(17) is available only for estimation of $\beta(\lambda)$ in $400\leq\lambda$ due to the observation limitation. The spectral curve of $\beta(\lambda)$ in $350\leq\lambda<400$ is estimated by interpolation based on the estimates $S(\lambda)$ in $400\leq\lambda$ and the terminal condition of $\beta(350)=0$.

It should be noted that although the estimates of spectral reflectance $S(\lambda)$ and emission spectrum $\alpha(\lambda)$ are obtained by solving Eq.(15), we need the excitation spectrum $\beta(\lambda)$, which is nested in (4) and (17). In this paper, we adopt an iterative procedure. The initial estimate of $\beta(\lambda)$ is set based on (17) where $S(\lambda)$ is constant in $400\leq\lambda$. The estimates $\hat{\mathbf{s}}$ and $\hat{\boldsymbol{\alpha}}$ are updated by solving Eqs. (15)-(16), and the estimate $\hat{\boldsymbol{\beta}}$ is updated in Eq.(17) at each iteration step. This iterative process is so effective that the performance becomes sufficiently small in about two steps.

Simulation Experiments

We execute simulation experiments to examine the performance of the proposed estimation method from various points of view in detail. We used a red sample containing an orange fluorescent color as a target object. The spectral functions of reflectance, excitation, and emission are shown in [9]. Three light sources with continuous spectra were used, so that N=3. Figure 1 shows the spectral-power distributions for the light sources of (1) an incandescent lamp, (2) an artificial sunlight lamp, and (3) a flood daylight lamp for the photograph used in this experiment. All spectral functions were sampled in 5 nm intervals, and the Donaldson matrix **D** has the size of m=71 and n=61. The spectral radiances were calculated with $\mathbf{y}=\mathbf{De}$ for each illuminant. Furthermore, assuming observation noises of 1% for the camera outputs, normal random numbers were generated with a standard deviation of $\sigma=0.01$ and added to the calculated observations.

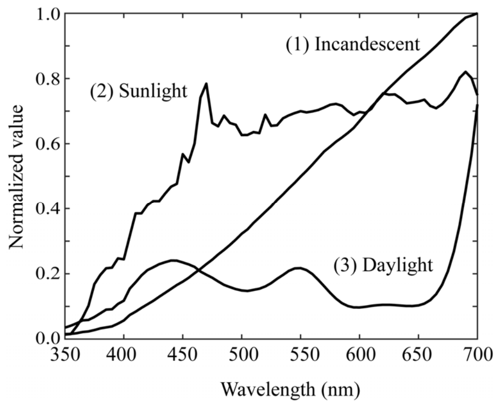

***Figure 1** Illuminant spectral-power distributions of three light sources..*

A. Estimation results using a multispectral camera data

We suppose the use of a multispectral imaging system with eight band-pass channels in the visible wavelength range. Figure 2 shows the total spectral sensitivity functions of the imaging system, which was constructed with a monochrome CCD camera and eight interference filters for practical use in digital archiving (see [10]). We collected Donaldson matrices for different fluorescent materials to create a database of spectral reflectances and emission spectra of real fluorescent objects. These include our fluorescence database consisting of the measurements of fluorescent paints, cloths, papers, boards [7-8], and also MCSL fluorescence database of different printed materials [11].

The basis functions to approximate the spectral reflectance and the emission spectrum were derived from singular value decomposition (SVD) analysis of the respective spectral datasets. Figure 3 shows five basis functions (K=5) for the spectral reflectance, and Figure 4 shows three basis functions (L=3) for the emission spectrum. The weighting parameter are set to $\mu_s=\mu_\alpha=0.5$

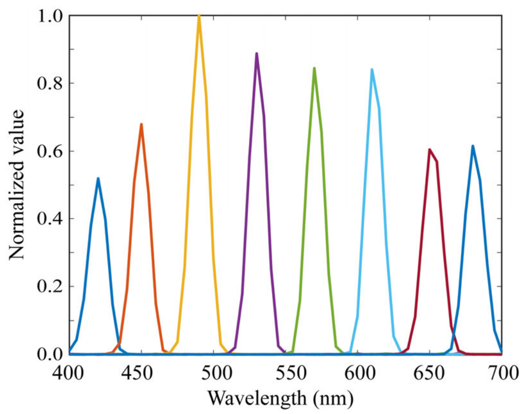

***Figure 2** Total spectral sensitivity functions of the imaging system used.*

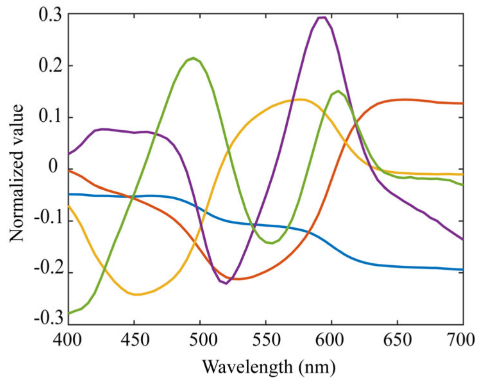

Figure 3 *Basis functions for the spectral reflectance estimation.*

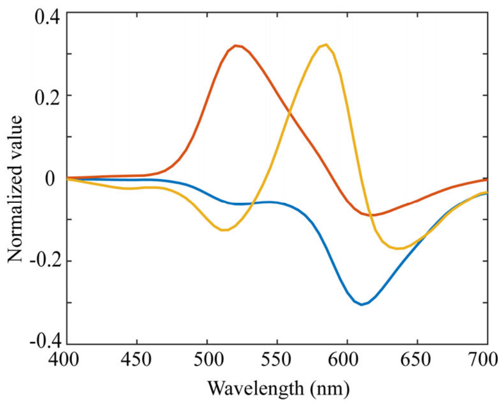

Figure 4 *Basis functions for the emission spectrum estimation.*

The estimation results for reflection and emission spectra of the target fluorescent object are depicted in Figure 5 (a), where the bold curves represent the estimates and the broken curves represent the original spectra. Figure 5 (b) shows the estimation results for the excitation spectrum. The Donaldson matrix reconstructed with the estimated spectral curves. The root mean squared error between the original and estimated Donaldson matrices with the size 71-by-61 was 6.96e-05. The effective processing time was 0.00874 sec.

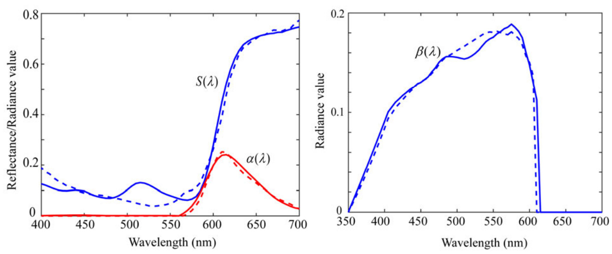

Figure 5 *Estimation results for the red sample, where bold and broken curves represent the estimate and original spectra, respectively. (a); Spectral reflectance and emission spectrum, (b); Excitation spectrum.*

B. Performance comparison with the other methods

We compare the above performance with the other methods presented for estimating the Donaldson matrix. The Donaldson matrix **D** is a (71-by-61) rectangular matrix with a wavelength sampling of 5 nm. If the number of channels becomes *M*=61 and the sensitivity matrix **R** is set to a (61-by-61) identity matrix, the proposed algorithm executes the estimation automatically for a spectral imaging system. We performed the estimation under the above situation for the same object. The root mean squared error between the original and estimated Donaldson matrices became 7.01e-05 with $\mu_s = \mu_\alpha = 0.5$. The processing time was a little slower, but the estimation accuracy did not improve much.

Finally, we compared our performance with the method presented by Blasinki et al. [5]. The method was to solve the estimation problem of three spectral functions with basis functional approximation by using ADMM approach. We repeated the experiment using Blasinki's algorithm in this study. The same basis functional representation to reflectance and emission are shown in Figures 3-4 were used. Additionally, we determined three basis functions to excitation in the same way as ours. The estimation error for the Donaldson matrix was calculated as a (61-by-61) array. Then it was 1.55e-4 after five iterations. As a result, the processing time is increased, but the estimation accuracy did not increase.

Conclusions

In this paper, we have proposed a compact and reliable method to estimate the bispectral Donaldson matrices of fluorescent objects by using a multispectral imaging system. We supposed a general image acquisition system that allowed multiple illuminant projections to the object surface and multiple response channels in the visible range. The Donaldson matrix was modeled as a two-dimensional rectangular array where the excitation range was defined in a 350 to 700 nm wavelength range, and the reflection/emission range was defined in a 400 to 700 nm wavelength range.

The observation model was described using the spectral sensitivities of a camera and the spectral functions of reflectance, emission, and excitation constructing the Donaldson matrix. The problem of estimating the spectral functions is formulated as a least squares problem to minimize the residual error of the observations acquired by the multispectral imaging system and the roughness of the spectral functions.

We utilized the fact that a fluorescent object usually contains a single fluorescent material (fluorophore). The excitation spectrum was then estimated using a physical model between excitation and absorption. An iterative algorithm was developed to obtain the optimal estimates of the whole spectral functions. The performance of the proposed method was examined in simulation experiments using a multispectral imaging system. We showed the estimation accuracy in comparison with the other methods, including the spectral imaging method.

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Author's Biography

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