Rapid multispectral endoscopic imaging system for *in vivo* assessing morphological and physiological characteristics of mouse intestinal

Yunhe Zhang,[a] Yixin Lu,[a] Zhanqin Zhang,[b] Zhuowen Liang,[c] Qianqian Xiao,[b] Kaijian Shao,[a] Yu Wang,[a], Jiawei Zhang,[c] and Shuang Wang [a, *]

[a] Institute of Photonics and Photon-Technology, Northwest University, Xi'an, Shaanxi, 710069, China

[b] Department of Anesthesiology, The First Affiliated Hospital of Xi'an Jiaotong University, Xi'an, Shaanxi, 710061, China

Corresponding author: Shuang Wang, swang@nwu.edu.cn

-- Supplementary Information--

Spectral pre-processing and analysis algorithms

The spectral pre-processing and analysis algorithms could be referenced to the previous publications, including DOI: 10.1117/1.2337529, DOI: 10.1117/1.2337529, and DOI: 10.1364/ol.29.000587. Therefore, we presented a brief introduction on the algorithms. Authors, who are interested on the setup of the spectral pre-processing and analysis algorithms, could refer to the previous three papers for detailed information.

The formula for our method of rapid quantitative analysis, which is based on linear matrix inversion of the ratio between the absorption coefficient $\mu_a(\lambda)$ and reduced scattering coefficient $\mu_s^{'}(\lambda)$ obtained from reflectance spectra after spectral calibration and preprocessing, is expressed as follows:

$$\gamma(\lambda) = \mu_a(\lambda) / \mu_s^{'}(\lambda) \quad (1)$$

Where, $\gamma(\lambda)$ represents the relative absorption spectrum.

Theoretically, the tissue reflectance spectra $R_d(\lambda)$ at each wavelength can be obtained using Fick's law:

$$R_d(\lambda) = \frac{-j(z,\lambda)}{I_0}\bigg|_{z=0} = \gamma^{-1}\nabla\phi(z,\lambda)\big|_{z=0} \tag{2}$$

where, ϕ is the light fluence spatial distribution, j is the difues flux, I_0 is the incidengt power, and γ is the diffusion constant,which depends on the tissue optical properties.According to our system, which uses wide-beam visible light irradiation and point measurements to obtain reflectance spectra, we employed a one-dimensional δ-P1 approximation model. Under this model, the first two terms of the Henyey-Greenstein phase function (g and f respectively) are assumed to be $f = g^2$ and $g^* = g/(g+1)^2$.

By modifying the δ-P1 approximation model, for planar illumination the source term is given by:

$$\nabla^2\phi(\vec{\omega}) - \mu_{eff}^2\phi(\vec{\omega}) = -3\mu_s'\mu_t^* E(\vec{\omega},\hat{z}) + 3g^*\mu_s'\nabla E(\vec{\omega},\hat{z})\hat{z} \tag{3}$$

$$j(\vec{\omega}) = -\frac{1}{3\mu_t^*}[\nabla\phi_d(\vec{\omega}) - 3g^*\mu_s'\nabla E(\vec{\omega},\hat{z})\hat{z}] \tag{4}$$

$$E(z,\vec{\omega}) = E_0(1-R_S)\exp(-\mu_t^* z)\delta(1-\vec{\omega}) \tag{5}$$

where, $\mu_s' \equiv \mu_s(1-f)$ is the isotropic scattering coefficient, $\mu_t^* \equiv \mu_a + \mu_s'$ is the transport coefficient, $\mu_{\text{eff}} = (3\mu_a\mu_a)^{1/2}$ is the effective attenuation coefficient. R_s is the specular reflectance for unpolarized light, E_0 is the irradiance, $\vec{\omega}$ is the unit direction vector, and $\hat{z}$ is the inward pointing unit vector normal to the surface of the medium and is colinear with the z coordinate axis.

We were able to directly calculate the relative absorption spectrum γ(λ) from the tissue reflectance spectra $R_d(\lambda)$:

$$R_d = A[\alpha(\gamma) + \beta(\gamma)] \tag{6}$$

With $\alpha = \dfrac{[3+3(g^*+1)\gamma]}{2+2g_e-\gamma^2}$, $\beta = \dfrac{-\alpha a2[(1+a1)a2+(1+a1)]-a2\gamma}{1+3g_e/\gamma+a1(3+3g_e/\gamma)^{1/2}}$

Where, $A' = \frac{E(1-R_s)}{2+2g_e-\lambda^2}$, $a1 = 2A'/3$, $a2 = 2Ahg^*$, $g_e = (1-g^*)/(1-f)$, $A = \frac{1+R2}{1-R1}$, $h = 2/3(\mu_a + \mu_s')$, and $\nu = \mu g$.

Here R1 and R2 are the first and second moments of the Fresnel reflection coefficient for unpolarized light and are given by:

$$R_1 = \int_0^1 r_F(\nu)\nu d\nu \qquad R_1 = 3\int_0^1 r_F(\nu)\nu^2 d\nu \tag{7}$$

The relative absorption spectra $\gamma(\lambda)$ can be further modeled in in terms of the tissue chromophores and scattering properties as follows:

$$\mu_a(\lambda) = BS\mu_{a,oxy(\lambda)} + B(1-S)\mu_{a,deoxy(\lambda)} + W\mu_{a,water(\lambda)} \tag{8}$$

$$\mu_s'(\lambda) = V_s\lambda^{-\kappa} \tag{9}$$

where, B is the blood volume fraction, S is the blood oxygen saturation index, W the water volume fraction, V_s is the effective scattering volume fraction, and k is a constant related to effective scattering size. Meanwhile, $\mu_{a,oxy(\lambda)}$, $\mu_{a,deoxy(\lambda)}$ and $\mu_{a,water(\lambda)}$ are the excitation coefficients of the oxyhemoglobin, deoxyhemoglobin, and water respectively. Re-arranging up-mentioned two functions, and use matrix formulation we obtained the linear matrix inversion scheme below, which is used to quantify the tissue's blood supply condition:

$$X = A^{-1} * Y \tag{10}$$

Where, $A = \begin{vmatrix} B'S \\ B'(1-S) \\ W \end{vmatrix}$, $X = \begin{vmatrix} \frac{\mu_{a,oxy(\lambda 1)}}{\lambda_1^{-\kappa}} & \frac{\mu_{a,deoxy(\lambda 1)}}{\lambda_1^{-\kappa}} & \frac{\mu_{a,water(\lambda 1)}}{\lambda_1^{-\kappa}} \\ \frac{\mu_{a,oxy(\lambda 2)}}{\lambda_2^{-\kappa}} & \frac{\mu_{a,deoxy(\lambda 2)}}{\lambda_2^{-\kappa}} & \frac{\mu_{a,water(\lambda 2)}}{\lambda_2^{-\kappa}} \\ \cdots\cdots\cdots\cdots & & \\ \frac{\mu_{a,oxy(\lambda n)}}{\lambda_n^{-\kappa}} & \frac{\mu_{a,oxy(\lambda n)}}{\lambda_n^{-\kappa}} & \frac{\mu_{a,water(\lambda n)}}{\lambda_n^{-\kappa}} \end{vmatrix}^{-1}$, $Y = \begin{vmatrix} \gamma_{\lambda 1} \\ \gamma_{\lambda 2} \\ \gamma_{\lambda 3} \\ \cdots \\ \gamma_{\lambda n} \end{vmatrix}$ and $B' = B/V_s$.