Diffuse Reflectance Spectral Imaging for the quantification of absorption and diffusion in homogeneous turbid media

Veronica SORGATO¹, Michel BERGER¹, Charlotte EMAIN¹, Anne KOENIG¹, Blandine ROIG¹, Christine VEVER-BIZET^{2,3}, Jean-Marc DINTEN¹, Geneviève BOURG-HECKLY^{2,3}, Anne PLANAT-CHRÉTIEN¹

¹Laboratoire Images et Systèmes d'Acquisition CEA-LETI, Minatec Campus, 17 Rue des Martyrs, F38054 Grenoble, Cedex 9, France

> ²Laboratoire Jean Perrin UPMC Univ. Paris 06, UMR8237, F-75005, Paris, France

> > ³Laboratoire Jean Perrin CNRS, UMR8237, F-75005, Paris, France

veronica.sorgato@cea.fr, anne.planat-chretien@cea.fr, genevieve.bourg-heckly@upmc.fr

Résumé – Dans ce travail, nous utilisons le principe de la Spectroscopie en Réflectance Diffuse (DRS) pour se rapprocher d'une technique en Imagerie multi-spectrale capable de quantifier les propriétés optiques d'un milieu diffusant homogène. Dans une première étape de cette démarche, nous avons validé un système de DRS sans-contact avec des trajectoires séparées pour l'illumination et la détection. Ensuite, la sonde de détection a été remplacé par un capteur CCD offrant une meilleure résolution spatiale de la réflectance diffuse qui résulte d'une source ponctuelle. Actuellement, la méthode de calculs des propriétés optiques permet une bonne détermination de l'absorption au détriment de la diffusion. Nous avons donc développé un algorithme robuste qui permet une quantification optimale des deux paramètres et qui s'adapte à toutes les modalités de mesure.

Abstract – In this work, we make use of Diffuse Reflectance Spectroscopy (DRS) to approach a multi-spectral imaging technique able to quantify optical properties of a homogeneous medium. As a first stage in this approach, we have validated a non-contact DRS system with separated illumination and detection paths. The detection system was subsequently replaced by a CCD detector offering greater spatial resolution of the diffused reflectance originating from a punctual source. While quantification of the absorption coefficient is well achieved with the existing method, the discrimination of the scattering coefficient is deteriorated. Therefore, we have developed a robust algorithm that optimizes the quantification of both parameters and that can be adapted for any measurement modality.

1 Introduction

Quantitative measurements of absorption and diffusion coefficients can be obtained by Diffuse Reflectance Spectroscopy (DRS) rendering it a valuable technique for tissue diagnosis of various pathologies that present optical properties variation. Nevertheless, it is limited by the small area of inspection, which can be overcome by multi-spectral imaging.

Many groups have developed various multi-spectral imaging techniques for clinical applications ([1], [2]). Particularly, notable work has been done in the field of modulated illumination by ([3], [4]), achieving spatially resolved quantification of optical properties for a large field of view under clinical conditions.

Our approach to multi-spectral imaging considers a point spread function from which the optical properties of the medium can be estimated. Major work has already been done in this field by [5] showing different strategies to overcome instrumental limitations. In this paper we describe the evolution of a contact DRS system towards quantitative multi-spectral imaging of spatially resolved reflectance (SRR) resulting from punctual illumination. This punctual technique poses the basis for an extended technique in which a larger field of view will be inspected.

Both non-contact DRS and spectral imaging systems are validated by measuring SRR in Intralipid 1%, 1.5%, 2%, and 3% liquid phantoms with different concentrations of ink resulting in an absorption coefficient $\mu_a = [0.2, 2] \ cm^{-1}$.

A Monte Carlo simulation is employed as a solution of the radiative transfer theory under contact conditions. Therefore, various signal corrections based on the non-contact system's architecture are considered before optical properties are calculated. The principle of optical properties calculation of an unknown phantom relies on the reflectance comparison with a reference phantom for which the optical properties are known. Deterioration of optical properties estimation increases for noncontact systems when using phantoms with different diffusion coefficients. We therefore propose a robust correction algorithm that improves estimation for all techniques and most particularly for non-contact systems.

2 Experimental set-up

A probe featuring a central illumination fibre and concentric detection fibres at 6 different distances is used to measure SRR, as described in the contact DRS system reported in [6] and shown in Fig. 1. The spectrum of the Tungsten Halogen (T - H) Lamp used as illumination source is between 400-900 nm.



FIGURE 1 – Contact DRS with detailed distal end of probe : central illumination fiber with 6 detection fiber rings.



FIGURE 2 – Final Non-contact DRS with separated illumination and detection paths.

After validation of a preliminary simple non-contact DRS set-up (with parallel illumination and detection signals), the illumination and detection paths were separated as shown in Fig. 2. Folded punctual illumination at the sample's surface and vertical detection of the resulting SRR is achieved through a



FIGURE 3 – Quantitative multispectral imaging of SRR originating from punctual illumination.

beam-splitter (BS). In the illumination path, achromatic lenses are used to focus and control the size of the illumination point at the surface set to 500 μm . In the detection path, an achromatic doublet pair with a focal length of 100 mm for both doublets is used to drive the resulting diffused reflectance to the detection fibres of the probe with a magnification of 1.

Considering the non-contact DRS system of Fig. 2, instrumental transfer to spectral imaging of diffused reflectance is easily obtained. The spectral probe is replaced with a CCD detector and a filtering disposition is added between the doublets of the illumination path, where rays are collimated. The resulting Diffuse Reflectance Spectral Imaging system is shown in Fig. 3. The limited dynamic range of the CCD poses a major instrumental constraint of this set-up for the imaging of the point spread function. We have therefore explored various possible solutions that will be shown at the conference.

3 Method and Signal treatment

A pre-computed Monte Carlo simulation is used as a solution of the radiative transfer equation. The simulation describes photons travelling from a determined point at the surface, through a semi-infinite medium, and back to the surface where they are measured.

The essential principle for the calculation of the optical properties through the DRS system described in [6] lies in the reflectance correlation of an unknown sample with respect to that of a reference phantom for which the optical properties are known. These reference optical properties, reduced scattering coefficient μ'_s and absorption coefficient μ_a , determine a theoretical diffusion reflectance for each detection fibre derived from the Monte Carlo simulation. The measured and theoretical reflectance of the reference phantom are compared, resulting in a correction factor that adjusts the measured reflectance of the unknown phantom. This corrected signal is then fitted to a simulated reflectance, by minimizing the difference between both, and from which the unknown phantom's optical properties are determined.

This calculation method is valid only if the reflectance measurements are corrected from additive noise and taken under the same conditions to ensure a stable correction factor for the measured reflectance of both phantoms. It is therefore indispensable to previously correct raw measurements from the additive system's aberrations such as ambient light and instrumental offset. The correction factor is then able to compensate for multiplicative effects, as long as they are kept constant (See 3.2). However, in a non-contact set-up, the calculation method is incapable to make up for the effect of the distorted illumination beam profile which depends on the diffusion of the phantom (See 3.3).

3.1 Additive effect : Offset

The offset and ambient light are additive signals that contain no relevant information about the sample's optical properties. Thus, they are subtracted from the measured signal.

3.2 Constant Multiplicative effect : Optical Transfer Function

The constant multiplicative modification of the signal by the various components is described by their optical transfer function (OTF). The OTF is measured directly by placing the illumination signal at the image plane and comparing it to the signal reaching the object plane or vice versa. The correction of the measured signal is then applied by a simple deconvolution with the overall OTF. We verified that OTF-corrected and uncorrected signals estimate the same optical properties, proving the method's robustness regarding constant multiplicative modification. Thus, the measurement of the OTF is not required.

3.3 Effect of the inaccurate Illumination beam profile

When using a non-contact set-up, the projection of the illumination beam is distorted by the optical aberrations of the components conforming the illumination path, ie. the beamsplitter and achromatic doublets. The blurred border of the point consequently pollutes the closest detection fibre ring. Namely, the diffused reflectance of interest is immersed in the intense combination of specular reflection and aberrant diffusion reflectance originating at the inaccurate border. To overcome this constraint, it was verified that even when the signal measured by the closest fibre is not used, appropriate optical properties estimation is still achieved with the contact DRS set-up.

The rest of the detection fibres rings are polluted only by the less intense aberrant diffusion reflectance which decreases with distance and can be considered negligible for the furthest ring. The most polluted closest fibres and the least polluted furthest fibres are the most sensitive to μ'_s and μ_a , respectively. Therefore, if μ'_s is the same for the reference and unknown phantom, the correction factor will stay constant and optical properties estimation will be relevant. However, if μ'_s differs between the phantoms, correction of the illumination profile is crucial to enable estimation. A possible correction strategy consists in considering the polluting illumination profile in the Monte Carlo simulations as has been done by ([5], [7]). An alternative solution considers a calibration approach. We opted for an adaptive correction algorithm that adjusts the reflectance of an unknown phantom according to the closest μ'_s of a range of reference phantoms.

4 Results

Appropriate quantification of absorption and scattering coefficients is achieved by implementing the developed robust adaptive correction algorithm. Figures 4 and 5 shows the capacity of the non-contact DRS system of Fig. 2 in the estimation of different optical properties of Intralipid (IL) 1% - 3% phantoms with $0.2 - 2 \ cm^{-1}$ in μ_a at 600 nm. Non-contact DRS (NC) measurements are shown in color whilst contact and theoretical optical properties are represented in black.

Similar estimation is obtained by the multi-spectral imaging technique of Fig. 3 after overcoming the instrumental constraints. Evaluation of the calculation errors of both non-contact systems with respect to the reference contact DRS system will be presented at the conference.

The low signal transmission of the lens and the limited quantum response of the detector beyond 750 nm are responsible for the decrease of signal-to-noise ratio. This explains the degradation in μ_a quantification at this spectral range as shown in Fig. 5. These type of instrumental limitations will determine the useful spectral range to be used in the final system.

Phantoms with different %IL and same estimated µ =0.4 ± 0.04 cm⁻¹at 600 nm



FIGURE 4 – Reduced Scattering Estimation.

Phantoms with different μ_a and same estimated $\mu'_s = 20 \pm 0.2 \text{ cm}^{-1}$ at 600 nm



FIGURE 5 – Absorption Estimation.

5 Conclusion

We have validated a non-contact DRS system and its corresponding quantitative multi-spectral imaging set-up, with respect to a well-founded contact DRS system. The systems are able to quantify absorption and reduced scattering coefficients by overcoming instrumental limitations that affect the measurement of spatially resolved reflectance. This work establishes the basis for an extended quantitative imaging technique that will cover a larger field of view to speed up and facilitate tissue diagnosis.

Références

- [1] N. Bedard, R. A. Schwarz, A. Hu, V. Bhattar, J. Howe, M. D. Williams, A. M. Gillenwater, R. Richards-Kortum, and T. S. Tkaczyk, "Multimodal snapshot spectral imaging for oral cancer diagnostics : a pilot study," *Biomedical optics express* 4(6), pp. 938–949, 2013.
- [2] C.-C. Yu, C. Lau, G. O'Donoghue, J. Mirkovic, S. McGee, L. Galindo, A. Elackattu, E. Stier, G. Grillone, K. Badizadegan, *et al.*, "Quantitative spectroscopic imaging for noninvasive early cancer detection," *Optics express* 16(20), pp. 16227–16239, 2008.
- [3] S. Gioux, A. Mazhar, B. T. Lee, S. J. Lin, A. M. Tobias, D. J. Cuccia, A. Stockdale, R. Oketokoun, Y. Ashitate, E. Kelly, *et al.*, "First-in-human pilot study of a spatial frequency domain oxygenation imaging system," *Journal of biomedical optics* 16(8), pp. 086015–086015, 2011.
- [4] D. J. Cuccia, F. Bevilacqua, A. J. Durkin, F. R. Ayers, and B. J. Tromberg, "Quantitation and mapping of tissue optical properties using modulated imaging," *Journal of biomedical optics* 14(2), pp. 024012–024012, 2009.
- [5] F. Foschum, M. Jäger, and A. Kienle, "Fully automated spatially resolved reflectance spectrometer for the determination of the absorption and scattering in turbid media," *Review of Scientific Instruments* 82(10), p. 103104, 2011.

- [6] A. Koenig, S. Grande, K. Dahel, A. Planat-Chrétien, V. Poher, C. Goujon, and J.-M. Dinten, "Diffuse reflectance spectroscopy : A clinical study of tuberculin skin tests reading," *Proc. SPIE 8592, Biomedical Applications of Light Scattering VII* 85920S, 2013.
- [7] F. Foschum and A. Kienle, "Broadband absorption spectroscopy of turbid media using a dual step steady-state method," *Journal of biomedical optics* 17(3), pp. 0370091– 0370097, 2012.