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Multispectral Imaging for Hemoglobin Estimation by PCA

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Abstract— **Tissular blood perfusion is helpful to assess the health condition of a subject and even monitor superficial lesions. Current state of the art is focused on developing non-invasive, quantitative and accessible methods for blood flow monitoring in large areas. This paper presents an approach based on multispectral images on the VIS-NIR range to quantify blood perfusion. Our goal is to estimate the changes in deoxygenated hemoglobin. To do so, we employ principal component analysis followed by a linear regression model. The proposal was evaluated using in-vivo data from a vascular occlusion protocol, and the results were validated against photoplethysmography measurements. Although the number of subjects in the protocol was limited, our model made a prediction with an average similarity of 91.53% with a mean R-squared adjusted of 0.8104.**

I. INTRODUCTION

The microcirculatory perfusion considers the supply of oxygen and nutritional exchange, through smaller arteries, arterioles, capillaries, and venules. Adequate tissue perfusion is indispensable to meet the metabolic demands of tissues allowing them to maintain normal body functions [1]. Blood perfusion can be also evaluated by considering non-invasive optical techniques. Furthermore, this methodology can been adopted in a clinical setting because it provides a quantitative and qualitative non-contact assessment [2], [3]. The principal techniques used to measure tissue oxygenation and hemoglobin concentration are near-infrared (NIR) spectroscopy, laser Doppler flowmetry, pulse-oximetry, photoplethysmography (PPG), hyperspectral and multispectral imaging (MSI). These devices are used in different medical applications such as anemia [4], diabetic foot [5] and retinopathy [6], skin ulcer [7], burn and wound analysis [8], diagnosing peripheral arterial disease, monitoring microcirculation to identify shock in critical care [2], monitoring blood flow at surgical sites, and surgical revascularization procedures [9].

There are several tissue perfusion parameters for non-invasive evaluations. Some of the techniques employed are pulse-oximetry, plethysmography, and spectroscopy. One of the most popular parameters is SpO2. Other useful parameters are perfusion indexes, changes of the oxygenated and deoxygenated hemoglobin content, hemoglobin indexes, and water content measurements to name a few [10].

The color of human skin is the result of a complex combination of absorption and scattering events. This property occurs as a consequence of the interaction between skin chromophores and light [3], [11]. The main chromophores in skin and tissue are hemoglobin, melanin, fat, water and other substances, like bilirubin or β-carotene [7]. These molecules are found in various skin layers. For example, melanin is found mostly in the epidermis, while hemoglobin has the most predominance in the dermis layer [3].

MSI is a noninvasive, non-ionizing, and contact-free method that is suitable for clinical diagnostics and studies [1], [12], [13]. Multispectral images can collect data from the visible wavelengths, as well from other portions of the electromagnetic spectrum, such as the NIR region [3], [8], [13]. NIR data is useful to estimate concentrations of chromophores like melanin and hemoglobin [5], [11].

In this work, we propose a procedure to test the variation of blood oxygenation in-vivo throughout an occlusion experiment by using MSI. The protocol consists of interrupting the blood flow in an extremity by applying different pressure levels through a sphygmomanometer. As reported in [14], the SpO2 values are not reliable since they exhibit no considerable changes during low-pressure occlusions. Besides that the measurements are inaccurate when there is a total occlusion state due to the absence of arterial pulsations. The main contribution of this work is to model the changes in Hb without using a reference spectra and validate them against PPG measurements. Section 2 details the methodology for the acquisition protocol of MSI and PPG signals by an occlusion experiment. Also, we describe the linear regression model (LRM) proposed for the estimation process. In Section 3, we show the results obtained after applying the model in multispectral images, and we validate the estimation with values obtained using a commercial system. Finally, the conclusions are shown in Section 4.

II. METHODS

In this work, we explored the feasibility of measuring changes in tissular perfusion by employing a multispectral camera. We recorded multispectral images of an extremity during a vascular occlusion protocol. The multispectral camera employed was a CMS-V1-C-EVR1M-USB3 (Silios Technologies) [15]. The camera records eight different channels centered at 559, 595, 632, 672, 714, 751, 791, and 828 nm. Each channel image has a dimension of 426 x

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339 pixels, and they were recorded by using an exposure time of 16.7 ms and a frame rate of 23 Hz. We used a Fiber-Lite Mi-150 Illuminator Series, equipped with a 150 W Halogen light, as a light source. The PPG data was recorded by using a MAX30102 sensor, and an Arduino microcontroller to operate it. A MightySat Pulseoximeter from Masimo [16] was employed to record reference pulse-oximeter measurements.

A. Experimental protocol

The protocol is based on the one reported in [14], and was approved by the Ethics Committee of the Universidad Autonoma de Aguascalientes. The participants were informed about the protocol, and all gave informed consent. The protocol was applied to six healthy participants (4 females and 2 males), ranging from 25 to 36 years. First, the blood pressure of the participants was measured using an automatic pressure monitor BP7450 (Omron). The resulted diastolic and systolic pressures of each participant at the beginning of the protocol are detailed in Table I.

TABLE I
RECORD OF DIASTOLIC, SYSTOLIC AND TOTAL OCCLUSION (TO) PRESSURES.

Patient ID	Diastolic Pressure	Systolic Pressure	TO Pressure
1	71 mmHg	103 mmHg	120 mmHg
2	75 mmHg	100 mmHg	120 mmHg
3	66 mmHg	86 mmHg	110 mmHg
4	68 mmHg	85 mmHg	110 mmHg
5	76 mmHg	122 mmHg	140 mmHg
6	83 mmHg	124 mmHg	140 mmHg

The subject sits straight with the palm of its dominant hand facing the multispectral camera. The camera is placed 80 cm above the image area, and the light source is placed at 45 degrees of the camera. Cuff pressure was placed on the dominant hand's upper arm. The cuff is connected to a sphygmomanometer to control the pressure during the occlusion protocol. The pulse-oximeter and PPG sensors are connected to the thumb and index fingers, respectively.

The protocol considers five stages of 2 minutes each one: (i) Start, (ii) venous occlusion (VO), (iii) rest, (iv) total occlusion (TO), and (v) reperfusion. During the start stage, no pressure is applied to the handcuff. At the beginning of the VO stage, a pressure of 60 mmHg is applied constantly. The pressure is released at the beginning of the resting stage. Once the resting stage is over, a pressure equal to the systolic pressure of the volunteer plus 20 mmHg is applied to induce a total occlusion. This pressure is maintained during the complete TO stage, and released at the 8:00 minutes mark. The multispectral camera and the pulse-oximeter and PPG sensors keep recording data until the end of the reperfusion stage.

B. PPG processing

The PPG measurements provide us a reference to the state of the tissue perfusion in a finger. Although this parameter is calculated in a single measurement position, it is a non-invasive and well-known reference method [17]. First, we extracted the AC and DC components from the raw PPG signals provided by the sensor MAX30102, using the methodology from [18]. Later, we estimated the changes in oxygenated hemoglobin (HbO_2) and deoxygenated hemoglobin (Hb) by the methodology in [14], which is based on the modified Beer-Lambert Law. This method estimates the attenuation changes from the DC components as

$$\Delta A_{\lambda_{660}} = \ln\left(\frac{\mathrm{DC}(0)_{\lambda_{660}}}{\mathrm{DC}_{\lambda_{660}}}\right), \quad \Delta A_{\lambda_{880}} = \ln\left(\frac{\mathrm{DC}(0)_{\lambda_{880}}}{\mathrm{DC}_{\lambda_{880}}}\right) \quad (1)$$

where $\ln(\cdot)$ stands for the natural logarithm function, and DC represents the average component extracted at each of the wavelengths of the PPG sensor. The values $\mathrm{DC}(0)_{\lambda_{660}}$ and $\mathrm{DC}(0)_{\lambda_{880}}$ are the initial measurements for both wavelengths.

Given both wavelengths, a system of equations is solved to estimate the change in Hb and HbO_2 concentrations as

$$\Delta[\mathrm{Hb}] = \frac{\Delta A_{\lambda_{880}} \varepsilon_{\mathrm{HbO}_{2},\lambda_{660}} - \Delta A_{\lambda_{660}} \varepsilon_{\mathrm{HbO}_{2},\lambda_{880}}}{\left(\varepsilon_{\mathrm{HbO}_{2},\lambda_{660}} \varepsilon_{\mathrm{Hb}_{\lambda_{880}}} - \varepsilon_{\mathrm{HbO}_{2},\lambda_{880}} \varepsilon_{\mathrm{Hb}_{\lambda_{660}}}\right) \cdot d \cdot \mathrm{DPF}} \quad (2)$$

$$\Delta[\mathrm{HbO}_2] = \frac{\Delta A_{\lambda_{660}} \varepsilon_{\mathrm{Hb}_{\lambda_{880}}} - \Delta A_{\lambda_{880}} \varepsilon_{\mathrm{Hb}_{\lambda_{660}}}}{\left(\varepsilon_{\mathrm{HbO}_{2},\lambda_{660}} \varepsilon_{\mathrm{Hb}_{\lambda_{880}}} - \varepsilon_{\mathrm{HbO}_{2},\lambda_{880}} \varepsilon_{\mathrm{Hb}_{\lambda_{660}}}\right) \cdot d \cdot \mathrm{DPF}} \quad (3)$$

where the extinction coefficients $\varepsilon_{\mathrm{HbO}_2}, \varepsilon_{\mathrm{Hb}}$ for each wavelength λ were taken from [19], and they are depicted in Table II. In (2) and (3), *DPF* stands for differential pathlength factor, while d is the distance between the light emitter and the photodetector. In the case of the MAX30102 sensor, we could not find these values reported in the literature. So, in a similar fashion to [14], we proceeded to estimate the relative changes in Hb and HbO_2 concentrations in $mm \cdot cm$([absolute chromophore concentration] $\times$ [optical pathlenght]). These are the tissue perfusion characteristics, which we will employ to validate our methodology.

TABLE II
EXTINCTION COEFFICIENTS FOR THE MAX30102 WAVELENGTHS

λ (nm)	Hb ($Mm^{-1}cm^{-1}$)	HbO_2 ($Mm^{-1}cm^{-1}$)
660	3.4408	0.3346
880	0.8412	1.2846

C. Multispectral Image Preprocessing

The multispectral images were calibrated to extract reflectance images according to the equation:

$$B(x,y,\lambda_i) = \frac{I(x,y,\lambda_i) - I_D(x,y,\lambda_i)}{I_W(x,y,\lambda_i) - I_D(x,y,\lambda_i)} \quad (4)$$

where $I(x,y,\lambda_i)$ stands for reflectance intensities measured during the protocol on (x,y) pixel and λ_i spectral channel. Meanwhile $I_W(x,y,\lambda_i)$ and $I_D(x,y,\lambda_i)$ were the intensities from the white and dark reference multispectral images, respectively. In this work, we employed the normalized

reflectance images, which were calculated for each spectral channel λ_i as:

$$R(x,y,\lambda_i) = \frac{B(x,y,\lambda_i)}{\sum_i B(x,y,\lambda_i)} \quad (5)$$

D. Linear Regression Model using PCA

We applied PCA to five reference multispectral images. Three reference images were selected at the half time of the first three stages (minutes 1:00, 3:00 and 5:00). The last couple of reference images were selected according to the min and max values of the Hb values during TO and Reperfusion stages. Only the first three principal components (PCs) were employed, which accounted for more than 86 % of the total variance. These PCs were extracted and used to project the data to a space of reduced dimension. The extracted PCs were employed to map every multispectral dataset recorded during the protocol.

Five different regions of interest (ROIs) were defined to study perfusion changes during to the occlusion protocol. The ROIs are the distal pulp zone of the middle finger (MF), the annular finger (AF) and of the little finger (LF), distal transverse arch upper zone or palmar region (PR), and the proximal transverse arch zone or central region (CR). The ROIs are depicted in Fig.1 a). The mean value of each ROI was calculated for each PC for every dataset in the experiment as illustrated in Fig.1 c).

According to [20], the middle and thumb fingers from the dominant hand present the most accurate values that reflect the arterial oxygen saturation. So, our hypothesis is that perfusion in the MF could be highly correlated to the measurements from the PPG sensor in the thumb. Next, Pearson correlation was evaluated between the PCs signals versus ΔHb and ΔHbO_2 from equations (2) and (3). Hence, several linear regression models (LRM) were evaluated to predict the changes in Hb and HbO_2.

III. RESULTS

The occlusion protocol was applied to six volunteers without declared diseases. The multispectral images acquired were dimensionally reduced in terms of their first three PCs obtained by PCA. We calculated the mean values for each component in the MF region. The mean Pearson's correlation of these values with Hb and HbO_2 was 0.7476 and 0.3805. Therefore, Hb had the highest linear correlation in all patients and was defined as the dependent variable. The first three PCs were employed as predictor variables in different LRMs. Using the data from only 6 patients, we found a model with a R-squared adjusted coefficient of 0.8104 ± 0.2541. The best result was obtained using the following model

$$\hat{y} = \beta_0 + \beta_1 x_1 + \beta_2 x_3 \quad (6)$$

where $\hat{y}$ is the estimated value for each position, the β coefficients are the model parameters, and the x_i variables correspond to the i-th PC, respectively. This model was applied to all reduced multispectral datasets. The results obtained for patient 2 are shown in Fig. 1).

The prediction obtained from the model (6) for patient 2 is shown in Fig. 1. Depicted in subfigure b), from right to left, images were estimated at the middle of each stage: (i) Start, (ii) VO, (iii) rest, (iv)TO, and (v) reperfusion. In Fig. 1 c), we compared the prediction from the LRM in different ROIs with the values obtained from the PPG measurement (depicted in black), throughout the complete occlusion experiment for patient 2.

We evaluated the cosine similarity (CS) between the PPG measurements $\Delta\vec{H}b$ versus the prediction for other ROIs according to

$$CS(\Delta\vec{H}b, \vec{\hat{y}}) = \frac{\Delta\vec{H}b \cdot \vec{\hat{y}}}{\left\|\Delta\vec{H}b\right\| \left\|\vec{\hat{y}}\right\|} \quad (7)$$

were $\vec{\hat{y}}$ represents the vector containing the predictions for other ROIs throught the experiment and the operator $\|\cdot\|$ represents the Euclidean norm. We found that the major average similarity considering the six patients occurs in the MF region with a value of 0.9153. The results obtained for AF and LF were 0.8178 and 0.6906, respectively. The average similarity value for the PR region was 0.4992, and for the CR was 0.5267.

IV. CONCLUSIONS AND FUTURE WORK

There are several definitions of tissular perfusion that depend mostly on a chromophore of interest. In clinical applications, it is important to measure water content, hemoglobin in different redox states and the amount of melanin to assess damage in a certain lesion. The accurate measurement of perfusion characteristics is challenging due to the complex interaction of light and tissue. Most of the approaches in the state of the art are based on know spectral profiles at given reference wavelengths. The drawback of theses methods is that they assume a similar spectral response for different race groups.

In this work, our goal was to measure induced changes in Hb and HbO^2 concentrations using MSI. We applied multiple linear regression to reduce multispectral images using PCA. A novelty in this work is the validation against clinical in vivo data. We evaluated the correlation between the trained model and the PPG reference data. We found a high correlation with Hb in all patients. In the case of HbO_2, this occurred only in a single patient. The predicted changes in Hb in the MF region had an average similarity of 91.53% with respect to the measurements provided by the PPG sensor in the thumb. These results show the proposal feasibility for the non-invasive estimation of deoxygenated hemoglobin in large areas of tissue. This method relies on the selection of a set of multispectral images for PCA. The proposal is not suitable for a live estimation of perfusion changes and is also dependent on the number of PCs selected. Future work will focused on the spectral decomposition of data and the automatic extraction/selection of the chromophore's spectral response and their contribution.

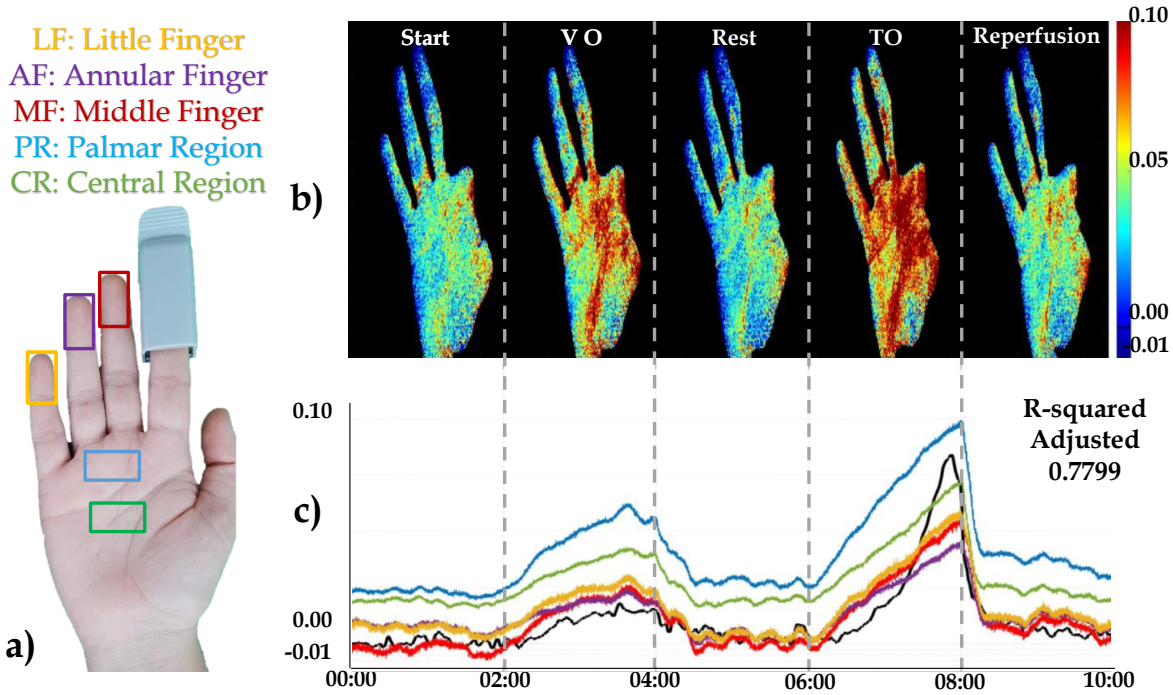

Fig. 1. a) Interest regions (Yellow: LF, Purple: AF, Red: MF, Blue: PR and Green: CR). b) Representative images of Hb predictions in Start, VO, Rest, TO and Reperfusion stages. c) Hb values from PPG signal in black and predictions from interest regions preserve the colormap of a).

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