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Research paper

An Alternative Wavelength Range for Noninvasive Assessment of Wound Tissue Oxygenation Status

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Abstract

This study aims to propose an alternative wavelength range for noninvasive prediction of wound transcutaneous blood oxygen saturation, S_tO_2. A pilot study was undertaken on an injured subject with superficial wound on the palm of hand for consecutively seven days. The offline processing of the measured light signals revealed a considerable consistency in the mean and standard deviation of S_tO_2 values calculated as 56.34 ± 4.42 % and 62.05 ± 3.53 % for the considered wavelength range of $520-600$ nm and $520-570$ nm, respectively. The average absolute mean difference was calculated as 5.72 %. This research concluded that $530-570$ nm range is suitably used in the prediction of wound S_tO_2. The use of this range would significantly speed up data acquisition and collection process for the realization of real time imaging of skin oxygen level during different phases of wound healing.

Keywords: *Blood oxygen saturation; Extended Modified Lambert Beer; Multispectral imaging; Wavelength range; Wound healing.*

1. Introduction

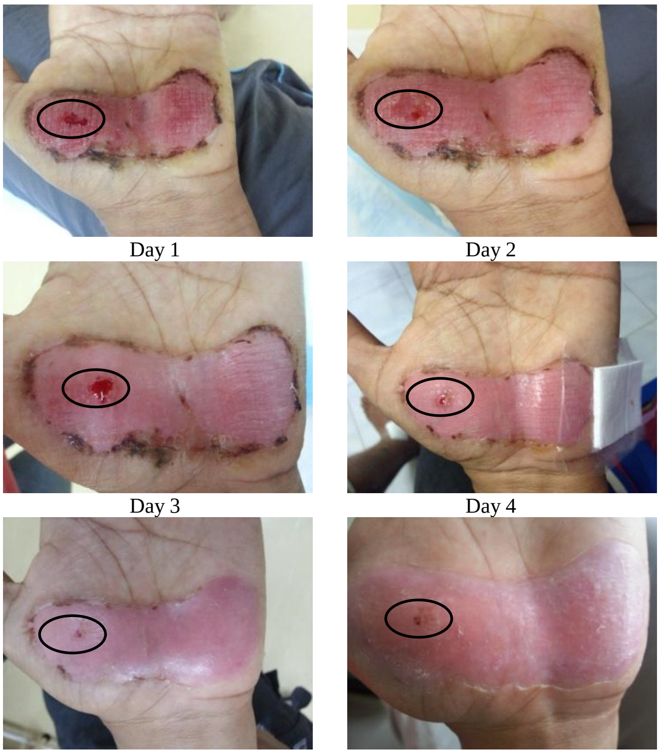
Over the years, researchers worldwide attempted various methodologies to identify a superlative way of measuring blood oxygenation level. The proposed methods vary from the modification of the commonly used pulse oximeter to the use of highly advanced noninvasive spectroscopy imaging system for noninvasive measurement of oxygen saturation level [1, 2]. The basic fundamental of these techniques is almost similar, in which it is based on the changes of the detected light intensity with variation in the absorptivity of hemoglobin components. While the operation of pulse oximeter is based on the ratio of ratios equation involving light signals of two wavelengths [3], i.e. red ($\lambda = 600$ nm) and infrared ($\lambda = 880$ nm), where absorptivity of deoxyhemoglobin (Hb) and oxyhemoglobin (HbO_2) is dominant, optical reflectance spectroscopy technique considers a wider selection of light in the visible to near infrared wavelength range (approximately 400 nm to 750 nm) [4]. Unlike the use of pulse oximeter which is limited to single point measurement, spectroscopy technique provides comprehensive information of tissue oxygen level of the imaged skin region. The transmitted light in the visible range such as blue ($\lambda = 450$ nm) and red ($\lambda = 600$ nm) is able to penetrate the superficial skin layer up to a depth of 100 µm and 550 µm, respectively [5]. This allows light to be reflected, transmitted and absorbed across the multilayer human skin which primarily comprises of pigment-rich epidermal and dermal layer (where HbO_2 and Hb mainly exist). Although the presence of other absorbers such as water, bilirubin and other hemoglobin signatures in the blood medium is widely acknowledged, light absorption by these absorbers is trivial as compared to the former.
Spectroscopy imaging technique has found its application in various fields including clinical diagnosis [6], agricultural and food industry [7], poultry farming [8] and in the studies of wound healing [9]. This is owing to its non-contact and non-destructive monitoring attributes. Previous study [10] demonstrated the feasibility of using 520 – 600 nm range in the prediction of wound S_tO_2. It was concluded that the distinctive S_tO_2 dependent changes in the absorptivity of hemoglobin makes it an excellent choice of wavelength range for wound oximetry. Other work [11] reported the use of mathematical approach to predict tissues oxygen saturation using information of absorber's absorption and scattering coefficients.
In general, wound healing process can be divided into three main stages; starting with tissue inflammation followed by proliferative stage and ends with tissue remodeling stage [12]. The outcome of wound healing depends largely on the supply of oxygen and nutrient in its microcirculatory system. Sufficient supply of both these elements would ensure proper and timely wound healing. Studies [13, 14] have shown that poor oxygen delivery in wound and the surrounding tissue would result in a hypoxic environment. The latter would subsequently interrupt the angiogenesis process that is vital for physiological healing [15]. Bedside clinical evaluation of wound physical state is not reliable to assess wound condition; this is considering the major underlying factor such as wound oxygen status is not taken into consideration. Hence, a proper diagnosis tool to noninvasively monitor tissue oxygen saturation level during the various phases of wound healing progress is highly sought after.
This paper aims to investigate the feasibility of using visible light spectrum in the wavelength range of $530-570$ nm as an alternative range for the prediction of wound tissue oxygenation status. This work compared the S_tO_2 values predicted using this proposed range and the $520-600$ nm range used in [14]. This is to experimentally investigate if the use of a shorter range comprising of five wavelengths for skin oximetry would yield an equivalent

outcome. Furthermore, the presence of absorbing species underneath the human skin in the visible wavelength range are widely documented.

2. Method

2.1. Experimental Subject and Method

In the effort to investigate the consistency in the performance of the considered wavelength ranges, a pilot experiment was performed on an outpatient volunteer who sought treatment at the University Health Centre, University Tun Hussein Onn Malaysia (UTHM) under the observation of professional medical officers and nurses. This research has been reviewed and approved by the clinical committee of the University Health Centre, UTHM. The participation is voluntary whereby corresponding volunteers are not subject to coercion and are well informed that they are free to withdraw from this research at any time without giving a reason. The subject recruited in this study is a 22-year-old male with a wound on the palm of the left hand who sought treatment from the medical centre for daily dressing following a recent motorcycle accident. Prior to the experiment, the subject was briefed on the experimental procedure and required to provide a written informed consent of which personal information of the individual involved is confidentially disclosed. The subject self declared to be a non-smoker with no underlying medical illnesses such as pulmonary disease, severe anemia (hemoglobin <7 g/dL) or diabetes mellitus. The work carried out in this study is noninvasive and merely an observational trial so no treatment or drug were administered to the patient.
Multispectral data collection and wound healing assessment were performed for seven consecutive days to observe changes in wound S_tO_2 during the different phases of healing. A series of wound images is shown in Figure 1, where positive wound healing progress can be visually observed based on the decrease in wound perimeter and gradual restoration of the epidermal integrity. The condition of the wound was physically assessed by medical professionals during each follow-up visit until it was declared healed and dressing was no longer required.



Day 1 Day 2 Day 3 Day 4 Day 5 Day 6

Day 7

Fig. 1: Daily physical state of the wound

2.2. Spectroscopic Measurement of Light Reflectance

The prediction of S_tO_2 is based on the *priori* knowledge of the extinction coefficients of hemoglobin components in the visible wavelength range of $530-570$ nm. Only the absorption of hemoglobin was considered here while light absorption by other absorbers in the blood medium such as melanin and water were disregarded. Optical approach was used to document changes in the wound oxygen level using multispectral imaging system previously introduced in [14] to detect light reflected from the wounded skin site. This employed system is briefly explained here again. A high intensity white light emitting diode (LED) was shone on a diffraction grating into a monochromator; the former was tuned to generate a spectrum of the desired wavelength range. Light of wavelengths in the considered range of $520-600$ nm and $530-570$ nm with sampling size of 10 nm was chosen to illuminate the wounded skin site (indicated by circled region shown in Figure 1). A charge-coupled device (CCD) camera (BUC4-500C from BestScope) was employed to capture image from the centre of the wound at each respective wavelength. In this experiment, the wounded skin site was at a distance of 17 cm from the CCD camera and subject was required to maintain still while reflectance data was captured in a dark room.
Even though there is, thus far, no optimum wavelength range in the measurement of this required health parameter, this work considered values given from wavelength range of $520-600$ nm as the gold standard. This is following a previous simulation study [16] which concluded that the use of this range is viable to predict one's oxygen saturation value. The predicted S_tO_2 is taken here as the mean of that measured across arteries, veins and capillaries.

2.3. Extended Modified Lambert Beer Model

The S_tO_2 value was determined in this work using a nonlinear model extended from the Modified Lambert Beer (MLB) law discussed in [17]. The S_tO_2 was determined by means of iterative fitting of the measured attenuation data using this mathematical model. The fundamental of Extended Modified Lambert Beer (EMLB) model shown in (1) is based on the existing knowledge on tissues' absorbing and scattering of lights underneath skin:

$$A(\lambda)=G_0+\mu_a d_0+G_1\lambda+\lambda\exp(-\mu_a d_1) \tag{1}$$

where light attenuation is defined as a complex function of medium's light scattering and absorption. The hemoglobin absorptivity, μ_a, used in EMLB is given by the sum of product of absorbers' concentration, C, and wavelength dependent extinction coefficient, ε, as followed:

$$\mu_a(\lambda)=\varepsilon_{HbO_2}(\lambda)C_{HbO_2}+\varepsilon_{Hb}(\lambda)C_{Hb} \tag{2}$$

where each absorbing component is represented by its subscript. While the total hemoglobin concentration is expressed as $T=C_{HbO_2}+C_{Hb}$, the S_tO_2 is given by:

$$S_tO_2 = \frac{C_{HbO_2}}{T} \quad (3)$$

3. Results

A comparison was done in the S_tO_2 predicted from the processing of multispectral raw data captured in the wavelength range of $520-600$ nm and $530-570$ nm. These reflectance data were fitted using the EMLB to give the percent S_tO_2 value using the extinction coefficient of oxy- and deoxyhemoglobin published in [18] as its *priori* knowledge.
Shown in Figure 2 and Figure 3 are the processed S_tO_2 maps for the considered wavelength range of $520-600$ nm and $530-570$ nm, respectively. A color bar is included on the right of each diagram to represent the blood oxygen saturation value.

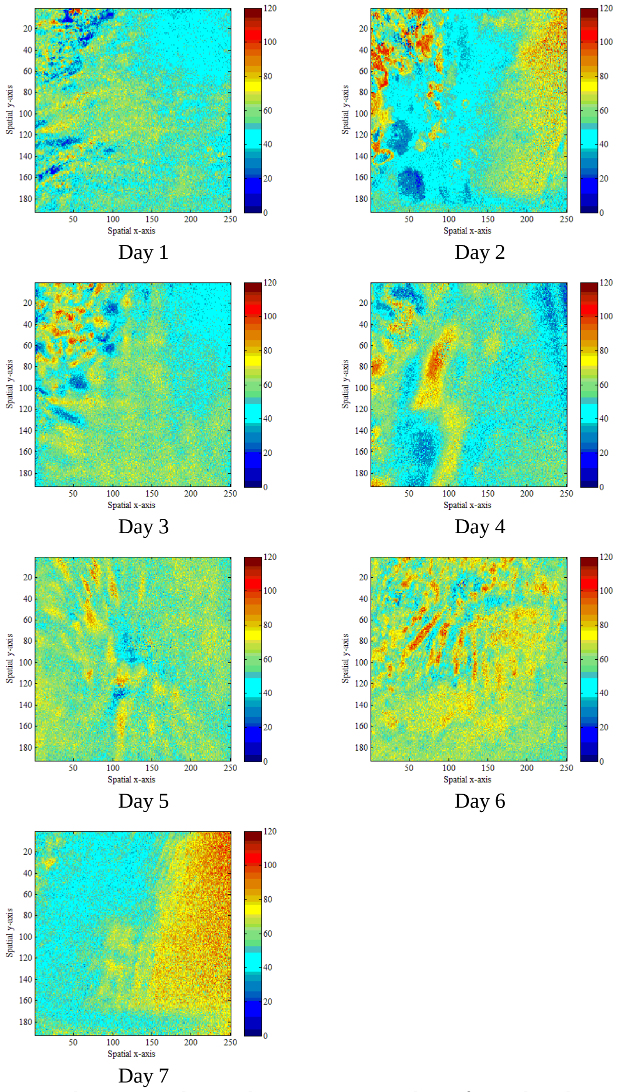
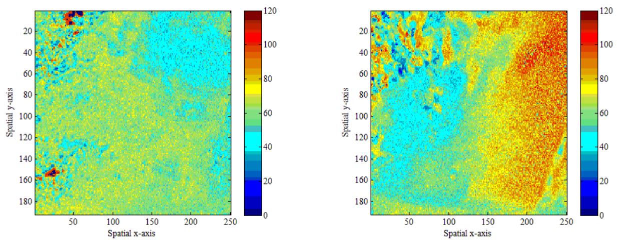


Fig. 2: The processed wound S_tO_2 map using data of wavelength range $520-600$ nm



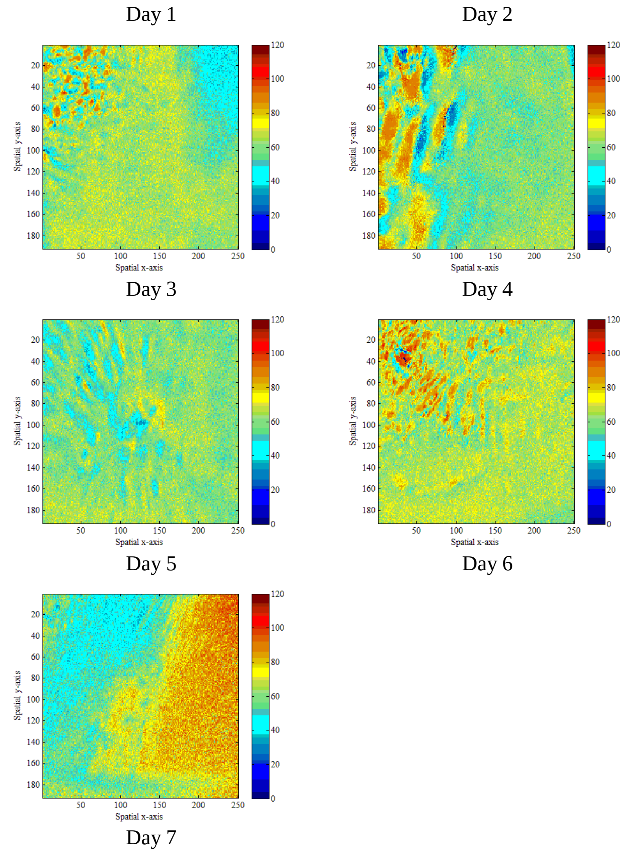


Fig. 3: The processed wound S_tO_2 map using data of wavelength range $530-570$ nm

The mean and standard deviation of S_tO_2 values for both wavelength ranges shown in Figure 2 and 3 are calculated and listed in Table 1.

Table 1: A comparison of wound S_tO_2 predicted using data of wavelength ranges $520-600$ nm and $530-570$ nm

Day	Predicted percent S_tO_2 (%)	
	$520-600$ nm	$530-570$ nm
1	51.74 ± 3.63	57.35 ± 1.12
2	53.47 ± 4.99	64.79 ± 2.25
3	54.88 ± 3.15	61.31 ± 0.89
4	52.01 ± 3.44	59.68 ± 1.42
5	58.73 ± 1.54	59.27 ± 0.77
6	62.88 ± 1.20	66.30 ± 1.03
7	60.65 ± 2.95	65.67 ± 2.39

The average absolute difference in the mean S_tO_2 for both wavelength ranges was calculated as 5.72 %. Next, the mean S_tO_2 values in Table 1 were analyzed and compared using a two-tailed independent samples *t*-test in MaxStat statistical software (MaxStat Lite, Germany) with confidence level of 95 %. The statistical test result revealed statistical significance, $\rho = 0.02$.
Meanwhile, the mean S_tO_2 readings from this work are compared with Huong *et al.* [14] who also studied multispectral wound images collected at wavelength range of $520-600$ nm with a sampling size of 10 nm. In this previous work, noninvasive multispectral measurements were conducted on different parts of the body namely the central forehead, posterior forearm and proximal ankle. In addition, the results were also compared with a similar work conducted on the unwounded skin region of the thenar of palm [19]. The mean and standard deviation of wound S_tO_2 are calculated from the spatial *y*-axis of the S_tO_2 maps in Figure 2 and Figure 3 and compared with the reports of previous related work [14, 19] as presented in Table 2.

Table 2: A comparison of wound S_tO_2 from this work with the reports of Huong *et al*. [14] and Philimon *et al*. [19]

Skin location	Mean ± standard deviation of percent S_tO_2 (%)
Thenar of palm (520 – 600 nm)	56.34 ± 4.42
Thenar of palm (530 – 570 nm)	62.05 ± 3.53
Posterior forearm [14]	60.55 ± 12.84
Central abdomen [14]	62.1 ± 8.23
Frontal foot [14]	60.04 ± 9.8
Unwounded thenar of palm [19]	54 ± 1.9

4. Discussion

Based on the results shown in Figure 2 and Figure 3, this work observed a consistently higher S_tO_2 value predicted using data of $530-570$ nm as compared to $520-600$ nm range. It is interesting to note that the changes in mean S_tO_2 throughout the wound healing process are considerably similar for the two considered ranges. The wound S_tO_2 averaged from values collected for seven consecutive days shown in Table 1 is given by 62.05 ± 3.53 % for the proposed wavelength range. Meanwhile an average S_tO_2 of 56.34 ± 4.42 % is given by its long range counterpart. The relatively low absolute difference in the mean values shown in Table 1 given by 5.72 % is largely owing to the overlapping in the choice of the wavelength range. This study considered both of these wavelength ranges as there is a large variability in the hemoglobin absorptivity. The *t*-test analysis showed that weak statistical significance exists between the values given by the considered ranges. This is likely to be contributed by the limited sample size used in this study. The accuracy of *t*-test analysis and mean difference in the values shown in Table 1 may be improved by either increasing sample size or considering more data points.

This work observed the highest and lowest differences in the S_tO_2 predicted using reflectance data collected on the second and fifth visit, respectively. This is possibly due to the differences in the uniformity of wound bed shown in Figure 1, wherein an irregular wound surface can visibly be seen for Day 2 as compared to that of Day 5. This shows that wound appearance complemented with the above mentioned factors may affect the S_tO_2 reading. Meanwhile an investigation into the trend of S_tO_2 values shown in Table 1 revealed an overall higher S_tO_2 during the sixth day of follow-up. Based on the wound image shown in Figure 1 for the corresponding day, it is reasonable to suspect that the wound has entered proliferative stage. These results are in accordance with the hypothesis that a high S_tO_2, a result of an increase in blood perfusion to the wound bed, is essential to facilitate wound healing.

Table 2 shows a comparison of mean and standard deviation of S_tO_2 value from this study with previous related work on tissue oxygenation status using a longer wavelength range of $520-600$ nm. The S_tO_2 value observed in this work is of close range to that reported in previous works that employed spectroscopy imaging system of similar arrangement [14, 19]. A higher mean S_tO_2 in wounded skin region compared to the normal skin region is, presumably, a result of functioning arterial vasomotion to facilitate healing in wound bed as previously explained. This value would eventually drop to normal as healing progressed and skin has keratinized. Differences in skin color and thickness as well as individual health status among recruited volunteers could account for the variability in reported mean S_tO_2 and standard deviation of the wound, which subsequently influenced the healing progress and time taken for wound recovery. Interestingly, S_tO_2 readings are observed to be higher in regions with less hair follicle, i.e. fingertips [20]. This is explained by the profusion of arteriovenous anastomoses (AVAs) which provides direct shunting of arterial blood into the venous compartment. Nonetheless, it is important to note here that the wound S_tO_2 range presented in this study can only be used as a guide. This is because the predicted S_tO_2 range is largely influenced by the experimental arrangement. This was previously demonstrated in a related work [1], which showed a decrease in S_tO_2 as the distance between detector and skin sample increased.

This is, to the authors' knowledge, the shortest range of wavelength reported for multispectral imaging of skin oximetry. The choice of this range would minimize data acquisition time; this considering only light of five wavelengths is used. This would be beneficial especially during experimental data collection when volunteers need to remain still for a long period. A slight movement during data collection would result in motion artefact, hence, rendering data useless for the study. This study implied that the difference in wound S_tO_2 given by the considered wavelength ranges may be reduced by increasing data size and using spectral data of higher resolution.

5. Conclusion

This study showed the use of an alternative wavelength range of 530 – 570 nm to predict wound S_tO_2 using multispectral imaging system. The performance of the proposed wavelength range was experimentally evaluated by comparing it with the previous proposed longer range. It is concluded that both of these ranges revealed similar trend in changes of wound S_tO_2 throughout the healing process of wound, hence, confirming the feasibility of the proposed shorter range for surface oximetry. Further work would suggest more research be carried out on more wound samples to investigate the effects of spectral resolution on the predicted S_tO_2 range.

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