Proceedings of the 28th IEEE
EMBS Annual International Conference
New York City, USA, Aug 30-Sept 3, 2006

Noninvasive measurement of total hemoglobin and hemoglobin derivatives using multiwavelength pulse spectrophotometry -In vitro study with a mock circulatory system-

Hironori Suzaki, *Student Member, IEEE*, Naoki Kobayashi, Takashi Nagaoka , *Member, IEEE*, Kiyotaka Iwasaki, Mitsuo Umezu, Sunao Takeda and Tatsuo Togawa, *Senior Member, IEEE*

***Abstract*—Total hemoglobin (tHb), carboxyhemoglobin (COHb), and methemoglobin (MetHb) are usually measured with a CO-oximeter. Noninvasive and continuous measurement of these blood components is expected to decrease the pain of a patient. Therefore, we developed an instrument to measure oxygen saturation (SpO_2), tHb, COHb, and MetHb non-invasively. Multiwavelength LED (600, 625, 660, 760, 800, 940, and 1300 nm) and a combined detector (Si, InGaAs) were built into the instrument (Seven wavelengths transparent pulse spectrophotometer). We used the Waseda mock circulatory system, which can simulate blood circulation in tissues and generate a pulse wave mechanically, to estimate the instrument's performance. Furthermore we proposed new calculation formula including DC components of optical density (this method). Under conditions without any change of other components, the mean error ± standard deviation between this method and the CO-oximetry were SaO_2=0.0±1.4%, tHb=0.0±0.0 g/dl, COHb=0.0±2.0%, and MetHb=0.0±0.3%. When the concentration of other components was changed, this method showed mean errors and standard deviations of SaO_2=0.2±1.6%, tHb=0.0±0.4 g/dl, COHb=0.5±4.1%, and MetHb=0.0±0.3%.**

I. Introduction

Pulse oximetry utilizing a pulsatile optical density ratio (Φ) is widely used for diagnosis and patient monitoring. However, conventional pulse oximeters cannot detect the presence of carboxyhemoglobin (COHb) and overestimate oxygen saturation (SpO_2) in patients.[1), 2)] If COHb or methemoglobin (MetHb) is present, SaO_2 has to be measured by blood sampling using a CO-oximeter invasively and intermittently. Noninvasive instruments for total hemoglobin (tHb), COHb and MetHb have already been developed [3), 4), 5)]. An improved formula described here, which utilizes the DC components (DCi) and DC components ratios ($\psi i/j$) to calculate the concentration of blood components, showed highly accurate results. This method showed great potential as a novel instrument for measuring tHb, SpO_2, COHb and MetHb.

Manuscript received April 3, 2006. This work was supported in part by Nihon Kohden Corporation.

H. Suzaki is with Graduated School of Science and Engineering, Waseda University, 3-4-1 Ohkubo, Shinjuku-ku, Tokyo, Japan (corresponding author to provide phone: +81-42-947-6788; fax: +81-42-947-6788; e-mail: hironori-suzaki@asagi.waseda.jp). He implements an internship with Nihon Kohden Corp., 1-31-4 Nishi-Ochiai, Shinjuku-ku, Tokyo, Japan.

N. Kobayashi and S. Takeda are with Nihon Kohden Corp., 1-31-4 Nishi-Ochiai, Shinjuku-ku, Tokyo, Japan.

T. Nagaoka, was with Faculty of Science and Enginerring, Waseda University, 3-4-1 Ohkubo, Shinjuku-ku, Tokyo, Japan. He is now with the Shizuoka Cancer Center Research Institute, 1007 Shimo-nagakubo, nagaizumi-cho, Sunto-gun, Shizuoka, Japan.

K. Iwasaki, M. Umezu and T. Togawa are with Waseda University, 3-4-1 Ohkubo, Shinjuku-ku, Tokyo, Japan.

II. Materials and Methods

A. Determination on Wavelengths

The optical density of transmitted light through the finger tissue depends on the wavelength and is relatively low within the range from 600 to 1300 nm. The absorption spectra of oxyhemoglobin (O_2Hb), reduced hemoglobin (RHb), COHb, MetHb, and water in this range are shown in Fig.1. Pulse oximetry calculates SpO_2 from the ratio between the pulsatile optical densities (Φ) derived from two wavelengths in which the RHb level is obtained from 660 nm and O_2Hb is obtained from 940nm. The ratio of pulsatile optical densities is shown as follows.

$$\phi_{i/j} = \Delta A_i / \Delta A_j = \log\left(I_i^S / I_i^D\right) / \log\left(I_j^S / I_j^D\right) \qquad (1)$$

where I^S is transmitted light through tissues in systole, and I^D is transmitted light through tissues in diastole. Specific wavelengths, which are sensitive to changes in concentration, were determined using a light source of continuous wavelength.[8)] For COHb, MetHb, and RHb, wavelengths of 600, 625, and 760 nm were selected, respectively, while 1300nm was selected for tHb. 940 nm was selected for RHb and 660 nm was selected for O_2Hb. 800 nm was selected as being near the isosbestic point of O_2Hb and RHb.

B. Measurement System

Wavelengths of 600, 625, 660, 760, 800, and 940 nm can be measured using Si photo diode. Moreover, 1300 nm can be

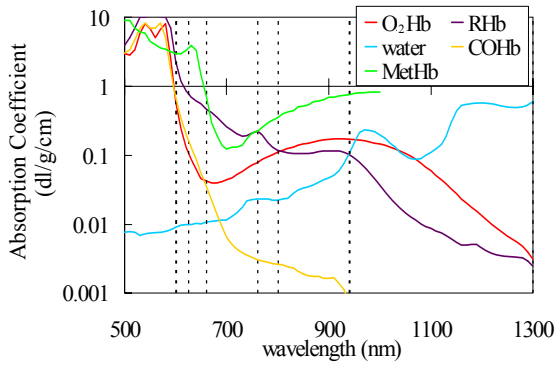


Fig. 1. Absorption spectra of O_2Hb, RHb, COHb, MetHb, and water from 500 to 1300 nm. Dotted lines show selected wavelengths.[6), 7)]

measured using InGaAs. Therefore, a combined detector having Si and InGaAs (K1713-09, Hamamatsu Photonics, Shizuoka, Japan) was used, and measurements of tHb, SO_2, COHb, and MetHb concentrations were examined. The data acquisition system comprised an optical sensor, seven wavelengths LED, low noise amplifier, timing pulse generator, and a 16-bit A/D converter. Signal processing was executed using the LabVIEW software system (National Instruments, Texas, USA) (Fig. 2). The DC and AC components from the optical sensor were displayed and recorded, and the recorded DC and AC pulse components were averaged. The ratios of pulsatile optical densities ($\Phi i/j$), DC components ratios ($\psi i/j$) and DC components (DCi) were then calculated.

A. Experimental System

It is difficult to perform in vivo experiments with COHb and MetHb in physiological condition. Therefore, we conducted an in vitro study using Waseda mock circulatory system developed by M. Umezu and T. Iwasaki (Waseda University) (Fig. 3)[9), 10)]. Concentrations of O_2Hb and COHb in the circulatory system were controlled by the concentration of oxygen and carbon monoxide in the air that flowed through an artificial lung. The concentration of MetHb was controlled by infusing a sodium nitrite solution into the circulatory system, and tHb was controlled by diluting the blood with phosphate buffered saline. The optical sensor (Fig. 2) was attached to the pulsation flow cell, which was inserted in the circuit of the circulatory system (Fig. 3), and the transmitted light of the pulsating blood was measured.

B. Statistical Analysis

The concentration of each blood component was assumed a dependent variable in multiple regression analyses. The ratios of the pulsatile optical densities ($\Phi i/j$), the ratios of the mean photocurrent ($\psi i/j$), and the mean value of the pulse wave (DCi) obtained from the photo detector were assumed to be independent variables. $\Phi i/j$ was calculated as follows.

$$\phi_{i/j} = \Delta A_{\lambda i} / \Delta A_{\lambda j} = \left(AC_{\lambda i} / DC_{\lambda i}\right) / \left(AC_{\lambda j} / DC_{\lambda j}\right) \quad (2)$$

Moreover, $\psi i/j$ was calculated as follows

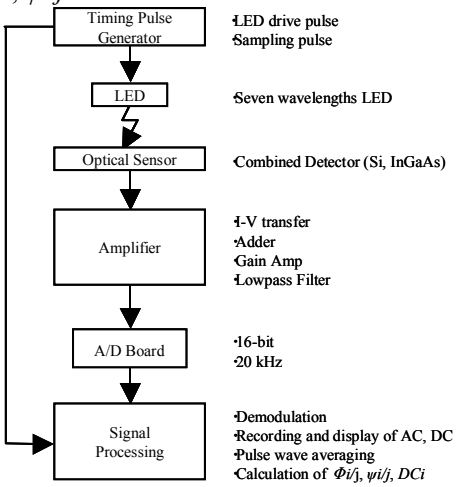


Fig. 2. Schematic diagram of the measurement system.

$$\psi_{i/j} = DC_{\lambda i} / DC_{\lambda j} \quad (3)$$

Wavelength j in formulas (2) and (3) was selected between 800, 940, and 1300 nm because the absorbance of COHb and RHb is small compared with O_2Hb at 800 and 940 nm, and the absorbance of water is large compared with O_2Hb and RHb at 1300 nm. The wavelength i in formulas (2) and (3) was selected between seven wavelengths (600, 625, 660, 760, 800, 940, and 1300nm). Moreover, 600, 625, and 660 nm were used for wavelengths i and j in formulas (2) and (3) because the absorbance of COHb is large compared with O_2Hb at 625 nm, and 600 and 660 nm are almost at the isosbestic point of the absorbance of O_2Hb and COHb. Multiple correlation coefficients adjusted for their degree of freedom were used to confirm the reliability of the formula constructed by multiple regression analysis. Thus, the independent variables were selected, and the following formula was constructed to calculate each concentration of the blood component:

$$\begin{aligned} Cp &= a_0 + a_1 \cdot \phi_1 + a_2 \cdot \phi_2 + \cdots + a_l \cdot \phi_l \\ &+ b_1 \cdot \psi_1 + b_2 \cdot \psi_2 + \cdots b_m \cdot \psi_m \\ &+ c_1 \cdot DC_1 + c_2 \cdot DC_2 + \cdots c_n \cdot DC_{nN} \end{aligned} \quad (4)$$

The concentration calculated by this formula was compared with the concentration measured by a CO-oximeter (OSM3, Radiometer, Copenhagen, Denmark).

C. The Effect of the Changes of Other Components

Interfering substances such as COHb in the blood causes overestimates in pulse oximetry. A similar error was expected using the measurement system described here. Therefore, statistical analyses were performed on the three data sets. The first analysis was for data sets that included only substances to be measured, and represented the training set. The second analysis was performed to estimate the error arising from the change of other components. The third analysis was for data that included the changes of the amount of all components in the training set. As an example, this process was performed for COHb measurements that considered SaO_2, MetHb and tHb as other components. Moreover, the standard deviations of the second analysis were compared with that of the third analysis for tHb, SaO_2, COHb, and MetHb.

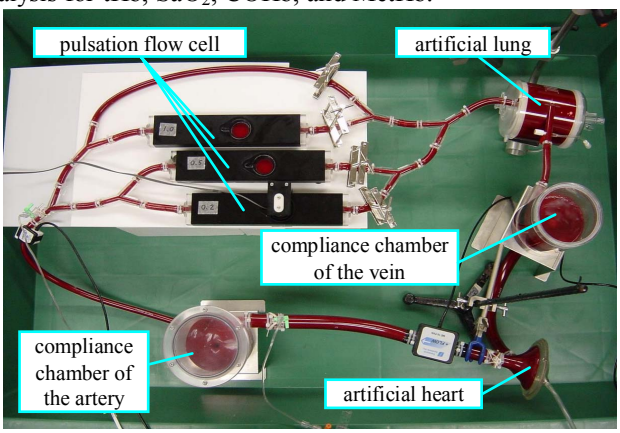


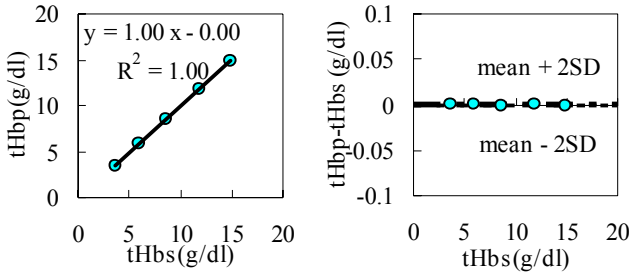
Fig. 3. Waseda mock circulatory system.

III. Results

Independent variables used in the multiple regression analysis are shown in Table 1. The correlation and Bland-Altman plot between concentrations from this method and the concentrations from blood samplings of tHb, SO_2, COHb, and MetHb are shown in Figs. 4-7. Standard deviations of COHb measurements were calculated when each concentration of SaO_2, MetHb, and tHb was changed (Fig.9). It was found that COHb was overestimated by about 40% when SaO_2 varied from 100% to around 80%. When the amount of other components was changed in the blood, the number of independent variables had to be increased. For this reason, the multiple regression analysis was repeated. This included the independent variables for each concentration

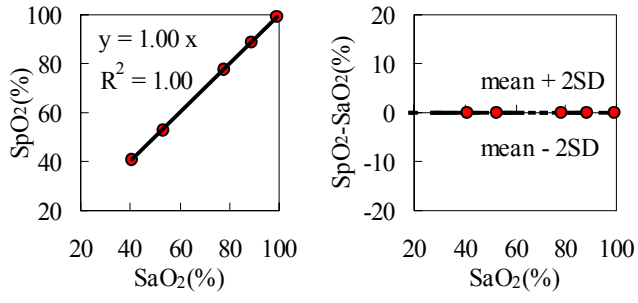
TABLE 1
RATIOS OF PULSATILE OPTICAL DENSITIES (Φi/j), DC COMPONENT RATIOS (Ψi/j) AND DC (DCi) USED TO CALCULATE CONCENTRATIONS OF tHb, SaO2, COHb AND MetHb.

tHb	SaO_2	COHb	MetHb
ψ760/940	Φ660/940	ψ625/800	ψ600/625
ψ760/800	Φ940/1300	Φ760/800	ψ800/940
Φ800/1300	ψ760/940		
ψ625/800	ψ760/1300		



(a) (b)

Fig. 4. (a) Total hemoglobin concentration values from this method (tHbp) versus the arterial total hemoglobin concentration obtained from CO-oximetry (tHbs). (b) The difference between the two sources was plotted. Mean differences and standard deviations were 0.0±0.0 g/dl.



(a) (b)

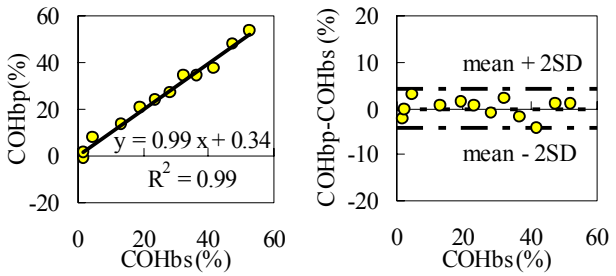
Fig. 5. (a) Oxygen saturation values from this method (SpO_2) versus the arterial oxygen saturation obtained from CO-oximetry (SaO_2). (b) The difference between the two sources was plotted. Mean differences and standard deviations were 0.0±0.0%.

when the concentration of other components was changed. Independent variables, which were redetermined by multiple regression analysis, are shown to Table 2. The correlation and Bland-Altman plot between concentrations obtained from this method and the CO-oximetry are shown in Fig.8. Comparisons of standard deviations in the case that the amount of a specific component without taking into account of the changes in other components was estimated (before) with that in the case that the amount of each component taking into account of all components was estimated (after) are shown in Fig. 9.

IV. Discussion

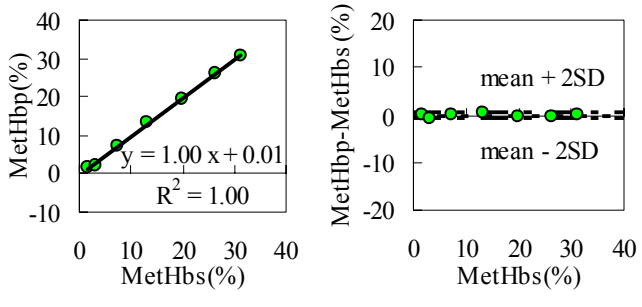
It is clinical important to measure multiple components of blood, simultaneously. For example, there will be cases CO poisoned patient with the change of tHb by bleeding and/or the change of O_2Hb by medical treatment, etc. Thus, measurement for multiple components, as well as their calibration under affection of the change in other components is required. [8)]

Moreover, it is important that taking account of the DC component ratios and the DC values were effective for the improvement in accuracy. Before taking direct-current



(a) (b)

Fig. 6. (a) Carboxyhemoglobin saturation values from this method (COHbp) versus the arterial carboxyhemoglobin saturation obtained from CO-oximetry (COHbs). (b) The difference between the two sources was plotted. Mean differences and standard deviations were 0.0±2.0%.



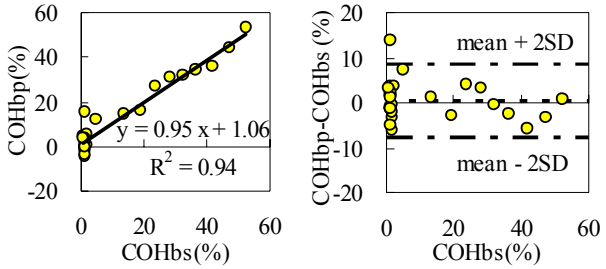
(a) (b)

Fig. 7. (a) Methemoglobin saturation values from this method (MetHbp) versus the arterial methemoglobin saturation obtained from CO-oximetry (MetHbs). (b) The difference between the two sources was plotted. Mean differences and standard deviations were 0.0±0.3%.

components into consideration, standard deviation was around 8%. Then, when multiple linear regression analysis including direct-current components was performed, standard deviation was around 4% (Fig. 8). Carbon monoxide poisoning is diagnosed when COHb>20% in blood: 40% is the fatal dose. If the standard deviation is around 4%, medical treatment can be started when COHb is around 24%. However, if the standard deviation is around 8%, there is a possibility that carbon monoxide poisoning will not be diagnosed, even if COHb=28%. Improvement in accuracy by DC components and the DC components ratios depends on the non-linearity between optical density and concentration of the target substance caused by scattering by blood cells[10].

TABLE 2
RATIOS OF PULSATILE OPTICAL DENSITIES (Φi/j), DC COMPONENT RATIOS (Ψi/j), AND DC (DCi) USED TO CALCULATE THE CONCENTRATION OF tHb, SaO2, COHb, AND MetHb AFTER TAKING INTO ACCOUNT OF THE CHANGES IN OTHER COMPONENTS

tHb	SaO2	COHb	MetHb
DC940	Φ760/800	Φ600/1300	ψ660/625
Φ600/625	Φ600/1300	Φ600/625	Φ600/1300
ψ660/1300	Φ760/940	ψ625/940	ψ800/940
Φ600/1300	Φ800/940	ψ660/625	Φ660/600
Φ660/940	ψ600/625	Φ940/1300	Φ660/940
Φ625/1300	DC660	ψ625/800	Φ600/625
ψ760/940	Φ660/625	ψ625/1300	Φ625/800
		ψ800/1300	DC600
		ψ660/600	



(a) (b)

Fig. 8. (a) COHb values from this method (COHbp) versus the arterial COHb obtained from CO-oximetry (COHbs) after taking into account of the changes in other components. (b) The difference between the two sources was plotted. Mean differences and standard deviations were 0.4±4.1%.

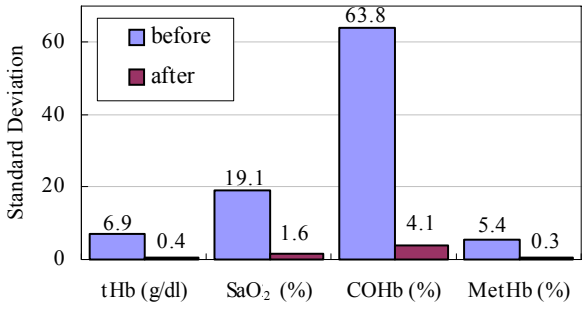


Fig. 9. Comparison between the standard deviations in the case that the amount of a specific component without taking into account of the changes in other components was estimated (before) and in the case that the amount of each component taking into account of all components was estimated (after).

V. CONCLUSION

Pulse spectrophotometry was applied at seven wavelengths and a noninvasive, continuous, and highly accurate instrument for measuring tHb, SaO_2, COHb, and MetHb was developed. Because of multiple regression analyses that considered the change of other components and DC components for optical density, highly accurate measurements of tHb, SaO_2, COHb and MetHb were achieved.

ACKNOWLEDGMENT

This research was performed as one of the research projects (05P29) entitled "Biomedical engineering research on advanced medical treatments" organized by the Advanced Research Institute of Science and Engineering, Waseda University. It was also supported by the Academic Frontier and ASMeW Projects of the Japanese Ministry of Education and Science.

REFERENCES

[1] SJ. Barker, KK. Tremper, "The effect of carbon monoxide inhalation on pulse oximetry and transcutaneous PO_2", Anesthesiology 66, 1987, pp. 677–679

[2] JB. Eisenkraft, "Pulse oximeter desaturation due to methemoglobinemia", Anesthesiology 68, 1988, pp. 279–282

[3] E. Noiri, N. Kobayashi, Y. Takamura, T. Iijima, T. Takagi, K. Doi, A. Nakano, T. Yamamoto, S. Takeda, T. Fujita, "Pulse total-hemoglobinometer provides accurate noninvasive monitoring", Crit. Care. Med. 33, 2005, pp. 2831–2835

[4] T. Usuda, Y. Sato, N. Kobayashi, M. Kanemoto, Y. Takamura, "Monitoring of blood carboxyhemoglobin concentration using pulse spectrophotometry", Ann. Biomed. Eng. 28, 2000, S-50

[5] B. Manzke, J. Schwider, N. Lutter, K. Engelhardt, W. Stork, "Multi wavelength pulse oximetry in the measurement of hemoglobin fractions", Proc. SPIE, 2676, 1996, pp. 332–340

[6] T. Aoyagi, K. Miyasaka, "Pulse oximetry and its simulation", IEEE Tokyo section Denshi 29, 1990, pp. 184–187

[7] N. Kobayashi, Y. Takamura, T. Usuda, S. Takeda, "Technique of non-invasive measurement for total hemoglobin using pulse-spectro-photometry", The Japanese J. Med. Inst. 73, 2003, pp. 587–588

[8] H. Suzaki, N. Kobayashi, H. Kubota, T. Aomi, T. Nagaoka, S. Takeda, A. Uchiyama, "Newly developed equipment for blood ingredients using pulse-spectro-photometry and the calibrator for it", 6th Asian-Pacific Conf. Med. Bio. Eng., April 2005, PA-2-85 [CD-ROM]

[9] K. Iwasaki, Y. Takeuchi, W. Saeki, M. Umezu, K. Ishihara, K. Imachi, "A novel methodology for pre-screening anti-thrombogenicity of artificial organs under physiologically identical pulsatile environments", International J. Artificial Organs, vol. 27, No. 7, 2004, pp. 567

[10] H. Suzaki, N. Kobayash, K. Iwasaki, M. Umezu, S. Takeda, U. Uchiyama, "Relation between hematocrit and optical density in pulse oximetry–in vitro study with Waseda mock circulatory system–", Proc. 27th Ann. Int. Conf. IEEE/EMBC05, 2005, 4.4.2–4, 1247 [CD-ROM]