



Non-invasive hemoglobin measurement using embedded platform

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ABSTRACT

This paper is based on the design and development of an in-house algorithm system to Estimate Total Hemoglobin in human blood using the Arduino based Embedded system. In our present society, monitoring Hemoglobin of subjects is very important in hospitals to keep track of low/high hemoglobin count during operations, delivery, dialysis, blood donations, perform stress test, monitoring patients in intensive care units, etc. is often done using invasive methods. In our approach, we have designed a finger probe using five LEDs on a multi-chip having wavelengths 670 nm/770 nm/810 nm/850 nm and 950 nm, and a single silicon photo detector with an inbuilt trans-impedance amplifier to acquire the signals through the finger using the Photoplethysmography (PPG) principle. The non-invasive method can be replaced in hospitals and homes to monitor the total hemoglobin level which is in real-time, painless, and infection-free from needles. We have tested our system on 8 subjects for three and five wavelengths and the values were compared to the hemoglobin measured using invasive methods in the pathology laboratory. The result demonstrates that the five wavelengths PPG method significantly improves the measurement accuracy of total hemoglobin, reducing the mean absolute error between the reference and the estimated hemoglobin values from 1.3778 g/dL for three wavelengths PPG to 0.3499 g/dL for five wavelengths PPG. The five wavelengths PPG system designed showed good linearity to estimate hemoglobin with the Regression Coefficient ($R^2 = 0.964$) compared to three wavelengths PPG system with the Regression Coefficient ($R^2 = 0.688$).

1. Introduction

In Today's world scenario, Anemia is a major worldwide public health issue. According to the World Health organisation (WHO), around 1.6 billion people which is about 30% of the total population are suffering from anemia. Pregnant women, Preschool children and Teenagers are a part of this vulnerable group of anemia [1,2]. Iron-deficiency anemia is the most common type of malnutrition is ranked as the eighth leading cause of disease in young women in developing countries [3]. Hemoglobin is found in red blood corpuscles (RBC) contains iron and protein, is responsible for carrying oxygen from the lungs to the various parts of the body and carrying carbon dioxide from various parts of the body to the lungs. The normal range of hemoglobin in human blood is given below [4].

- Women 12–16 g/dL
- Men 14–18 g/dL
- Newborn 14–20 g/dL

If the concentration of hemoglobin is less than the normal levels, it is called anemia and this results in disorders such as kidney and liver-

related diseases. If hemoglobin concentration is more than the normal range levels, it is called polycythemia [5]. In the medical field, determination of total hemoglobin before any surgery or blood transfusion is important and therefore needs a quick estimation of hemoglobin with an accurate value for critical conditions. Currently, in hospitals and pathological laboratories, invasive methods are widely used to measure the total hemoglobin concentration. In this setup, a blood sample is taken from a subject and analyzed in a laboratory. The drawback of this invasive system method is painful and the delay between the collection of blood and the determination of the hemoglobin level. Also, it does not permit real-time hemoglobin levels while monitoring subjects in hospitals or at homes. But using the non-invasive method, allows painless and real-time patient monitoring with no risk of infection, as no needles are utilized [6]. The absorption of entire blood in the visible and near-infrared range is ruled by the distinctive hemoglobin derivatives and the blood plasma which consists of water [7]. In a typical PPG measurement unit working in transmission mode, a light-emitting diode (LED) is used to illuminate the skin, and the variation of light absorption by the skin microvascular bed is monitored by a photo detector placed on the opposite side of tissue. The pulsatile change in the volume of blood can be monitored by measuring the amount of transmitted

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light or reflected light. This method of detecting changes in blood volume is known as PPG [8,9].

A lot of research work was carried out in estimating Total Hemoglobin in blood using the PPG principle. Tatiparti Padma et al. [10] estimated non-invasive hemoglobin concentration using PPG where signals were acquired by illuminating the finger with Infrared LED and a Red LED wavelength. The empirical equation for estimating total hemoglobin level in blood was obtained using a light attenuation model for skin-tissue- blood along with extinction coefficients of oxy-hemoglobin and deoxy-hemoglobin. O. Abdallah et al. [11], illuminated light source on the earlobe and a finger as an extremity and detected PPG signals. These signals were then digitalized and processed using LabVIEW Software to calculate hemoglobin in the blood. Kumar Abhishek et al. [12] proposed a device to measure Hemoglobin using a PPG procedure. The LEDs were used as the light source and the photo transistor was used to record the changes in the transmitted light through the finger. These changes in the electrical signal were applied to analog filters to remove noise and the resultant filtered signal was processed in MATLAB to estimate total hemoglobin. A. Mohamed Abbas et al. [13] measured hemoglobin level using two wavelengths and extinction coefficients of oxyhemoglobin and deoxy-hemoglobin derived from the clinical research. Akanksha Deep et al. [14] measured hemoglobin using 810nm and 1300nm LED with a photodiode. The major factor of the photodiode is its responsivity in ampere/watt. The placement of LED and photodiode was very important because the angle of incidence of light from the LED source affects the output at photodiode thus disturbing the output current from the photodiode. Kumar. R et al. [15] calibrated the designed system by measuring Hemoglobin levels via the Cyanmethemoglobin method. We have seen that many researchers have used two or three wavelengths of light to estimate total hemoglobin level in blood using the Photoplethysmo-graphic technique. Most of them have used microcontrollers with inbuilt ADC to detect the spectra and use mathematical equations and statistics to obtain the desired results. Our method of estimation total hemoglobin with 5 LED probe holds the advantage of being non-invasive, real-time, and cost-effective as it involves the use of Arduino Uno as an Embedded Platform, besides providing good accuracy, its mean absolute error is 0.3499 g/dL.

2. Materials and methods

2.1. Designed system for non-invasive hemoglobin meter

The main objectives of this paper are to design a finger probe with multiple LEDs on a single chip with a photo detector, to implement signal conditioning circuit to remove noise from the PPG signal, to develop an in-house algorithm to calculate total hemoglobin with formula based equations with three wavelengths LEDs and five wavelength LEDs. Here the ultimate moto is to measure total hemoglobin count with an error of less than lgm/dL. The complete design for the Non-Invasive Hemoglobin Meter is shown in Fig. 1.

The section consists of the Finger Probe with Multi-Chip LEDs

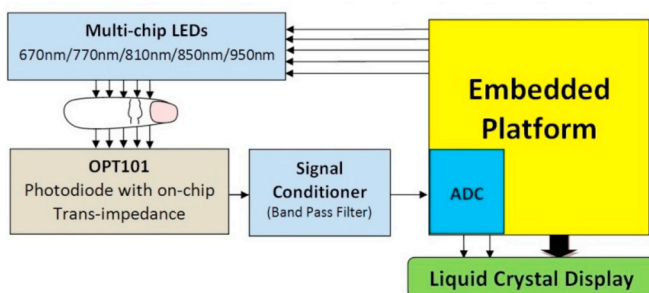


Fig. 1. Block diagram of non-invasive hemoglobin meter.

670 nm, 770 nm, 810 nm, 850 nm & 950 nm), OPT101 with an on-chip photo detector and Trans-impedance amplifier, Signal Conditioning (Band Pass Filter), Liquid Crystal Display (LCD) and the Embedded System. The five LEDs were controlled and turned on one at a time in the order of $\lambda = 670 \text{ nm}, 770 \text{ nm}, 810 \text{ nm}, 850 \text{ nm},$ and 950 nm using the Arduino Uno. The illumination passes through the finger when placed inside the finger probe. Due to changes in the blood volume in the finger due to heartbeat, the absorption of light changes, which leads to variation in the amount of light transmitted out of the finger. The changes in the transmitted signal are detected by the OPT101 which converts the transmitted light into voltage (PPG signal). The PPG signal is of an order of millivolts along with noise. Since the frequency of the human heart is around 1–2 Hz, the PPG signal was separated from noise using an active bandpass filter (0.72 Hz to 2.82 Hz) with an appropriate gain. The amplified PPG signal was then digitized using a 10 bit ADC in the Arduino Uno. The information was processed in the controller and the algorithm was executed to estimate hemoglobin level in human blood using three and five wavelengths.

2.2. Experimental set up

In most of the Pulse Oximeters, only two LEDs (Red/Infrared) are used along with a Silicon Photo detector and a current to voltage converter. In our design, we have used five LEDs on the single Chip and OPT101 (Photo detector with inbuilt trans-impedance amplifier) as shown in Fig. 2. OPT101 operates on a single supply voltage and the quiescent current is only $120\mu\text{A}$. It also has high responsivity from 600nm to 1000nm with its maximum responsivity at 0.45 Amperes/W at 650 nm [16]. The Multi-chip Emitter has all the sources in a small densely packaged area. Also, the power output of all the LEDs is around 5mW when the forward current is 20 mA [17]. The reason behind using the above two was because of its compact size, less space, and less dark current across the OPT101 when the finger is placed inside the finger probe. The Multi-Chip LEDs are mounted on the upper side of the finger probe and OPT101 is mounted on the opposite side of the finger probe.

The finger Probe was interfaced with the signal conditioner along with Arduino Uno and the PPG signal for each wavelength was acquired using a 10-bit analog to digital converter as shown in Fig. 3.

3. Theory/calculations

3.1. Wavelength selection for non-invasive hemoglobin detection

According to previous research studies [18,19], it was found that shorter wavelengths of light like blue and green can only reach up to the capillary bed, the yellow light can penetrate only upto the arterioles in the dermis, while longer wavelengths like red and near infrared can easily penetrate more through the skin and reach the arteries in the tissue. In the research work carried out, it was observed that both the Hemoglobin species have good absorption between 650 to 1000 nm and large Molar Extinction Coefficient difference between the oxy-hemoglobin and deoxy-hemoglobin as shown in Fig. 4. A higher penetration level was observed in the wavelength range from 600 nm to

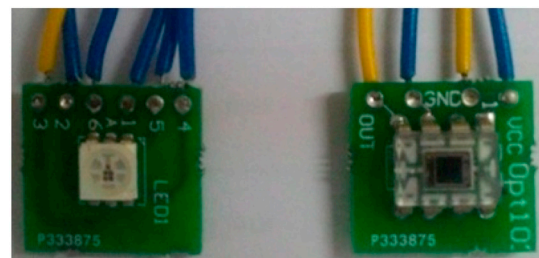


Fig. 2. Multi-chip Emitter MTMD6788594SMT6 and OPT101.

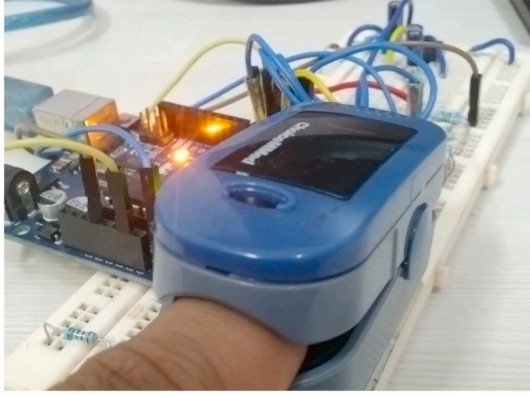


Fig. 3. Finger probe interfaced with signal conditioner and Arduino Uno.

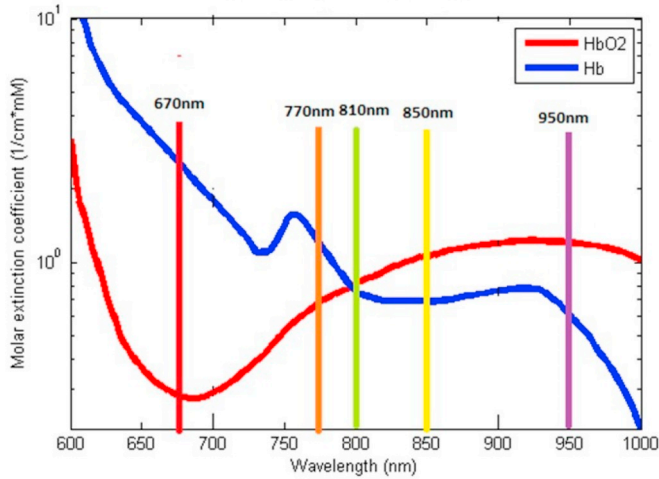


Fig. 4. Light absorbance (extinction coefficient) versus wavelength for two hemoglobin species [22].

950 nm with low Attenuation levels. As per the literature survey [20,21], it was observed that deoxy-hemoglobin blood was absorbed more in the wavelength range from 600 to 800 nm and oxy-hemoglobin blood was absorbed more in wavelengths from 820 to 1000 nm. At 810 nm, the isosbestic point was observed, where the absorption of oxy-hemoglobin and deoxy-hemoglobin were the same. Here, we have selected wavelengths in such a way that one wavelength is in between 900 nm to 1000 nm with significant absorption for both oxy-hemoglobin and deoxy-hemoglobin, in our case it was 950 nm. Second wavelength was chosen at the valley of oxy-hemoglobin i.e. 670 nm. Third wavelength was chosen at the interception (Isosbestic) of oxy-hemoglobin and deoxy-hemoglobin i.e. 810 nm. The other two wavelengths were chosen at either side of the interception with clear distinction of absorption between oxy-hemoglobin and deoxyhemoglobin i.e. 770 nm and 850 nm. However the choice of wavelengths can be slightly varied depending upon the availability of good quality LED sources in this region.

3.2. Detection of blood parameters by photoplethysmography

The principle of detection of blood parameters by photoplethysmography is based on differential absorption of Infrared/Non-Infrared radiations. The absorbance of a particular blood component for a particular wavelength is obtained using Beer-Lambert Law.

Absorbance can be calculated using Eq. (1).

$$A = \epsilon^\lambda * C * L \quad (1)$$

where A is absorbance and λ is the wavelength, C is the concentration

of absorbent (mol), L is optical path length in the cm, ϵ is the extinction coefficient of the substance at a specific wavelength, mol-l cm-l.

Eq. (2) shows that the Absorbance of a mixture is a sum of the absorbances of the components.

$$A = \epsilon^{\lambda 1} * C_1 * L + \epsilon^{\lambda 2} * C_2 * L \quad (2)$$

where $[C_1]$ and $[C_2]$ are unknown concentrations of components 1 and 2 and $\epsilon^{\lambda 1}$ is the extinction coefficient of component 1 and $\epsilon^{\lambda 2}$ is extinction coefficient of component 2 for a given wavelength [23].

3.3. Analysis of two-component mixture (Oxy-hemoglobin and Deoxy-hemoglobin)

To find two unknown components, we require a minimum of two wavelengths. The wave-lengths were chosen to maximize the differential molar extinction coefficient. Initially, we used three wavelengths ($\lambda = 670$ nm, 810 nm, and 950 nm) and later increased to five wave-lengths ($\lambda = 670$ nm, 770 nm, 810 nm, 850 nm, and 950 nm) to improve accuracy in estimating hemoglobin. Of the five wavelengths (two in the red region, two in the infrared and one at the isosbestic region). The five equations listed below were used to estimate oxy-hemoglobin and deoxy-hemoglobin.

$$PPG_Ratio_{670} = \epsilon Hb^{670} * Hb * L + \epsilon HbO_2^{670} * HbO_2 * L \quad (3)$$

$$PPG_Ratio_{770} = \epsilon Hb^{770} * Hb * L + \epsilon HbO_2^{770} * HbO_2 * L \quad (4)$$

$$PPG_Ratio_{810} = \epsilon Hb^{810} * Hb * L + \epsilon HbO_2^{810} * HbO_2 * L \quad (5)$$

$$PPG_Ratio_{850} = \epsilon Hb^{850} * Hb * L + \epsilon HbO_2^{850} * HbO_2 * L \quad (6)$$

$$PPG_Ratio_{950} = \epsilon Hb^{950} * Hb * L + \epsilon HbO_2^{950} * HbO_2 * L \quad (7)$$

where PPG_Ratio is the ratio of AC voltage of the PPG to DC voltage of the PPG for each wavelength when finger placed into the probe. ϵHb^λ is molar extinction coefficient for deoxy-hemoglobin for a particular wavelength, ϵHbO_2^λ is the molar extinction coefficient for oxy-hemoglobin for a particular wavelength. Hb & HbO₂ is the concentration of oxy-hemoglobin and deoxy-hemoglobin in moles per liter which is converted into grams per deciliter.

Formula to Calculate Total Hemoglobin for five wavelengths using Eqs. (3), (4), (5), (6) and (7).

$$\begin{bmatrix} PPG_Ratio_{670} \\ PPG_Ratio_{770} \\ PPG_Ratio_{810} \\ PPG_Ratio_{850} \\ PPG_Ratio_{950} \end{bmatrix} = \begin{bmatrix} \epsilon Hb^{670} & \epsilon HbO_2^{670} \\ \epsilon Hb^{770} & \epsilon HbO_2^{770} \\ \epsilon Hb^{810} & \epsilon HbO_2^{810} \\ \epsilon Hb^{850} & \epsilon HbO_2^{850} \\ \epsilon Hb^{950} & \epsilon HbO_2^{950} \end{bmatrix} * \begin{bmatrix} Hb \\ HbO_2 \end{bmatrix} \quad (8)$$

The unknown oxy-hemoglobin and deoxy-hemoglobin are calculated by rearranging the Eq. (8).

$$\begin{bmatrix} Hb \\ HbO_2 \end{bmatrix} = \begin{bmatrix} PPG_Ratio_{670} \\ PPG_Ratio_{770} \\ PPG_Ratio_{810} \\ PPG_Ratio_{850} \\ PPG_Ratio_{950} \end{bmatrix} * inv \begin{bmatrix} \epsilon Hb^{670} & \epsilon HbO_2^{670} \\ \epsilon Hb^{770} & \epsilon HbO_2^{770} \\ \epsilon Hb^{810} & \epsilon HbO_2^{810} \\ \epsilon Hb^{850} & \epsilon HbO_2^{850} \\ \epsilon Hb^{950} & \epsilon HbO_2^{950} \end{bmatrix} \quad (9)$$

From Eq. (9), total hemoglobin is calculated by adding the concentration of oxyhemoglobin and deoxy-hemoglobin.

$$Thb = Hb + HbO_2 \quad (10)$$

Eq. (10) indicates the concentration of total hemoglobin in Moles per liter which is converted into grams per deciliter.

3.4. Software description for non-invasive hemoglobin meter

Once the hardware setup is ready, the next step is to program the

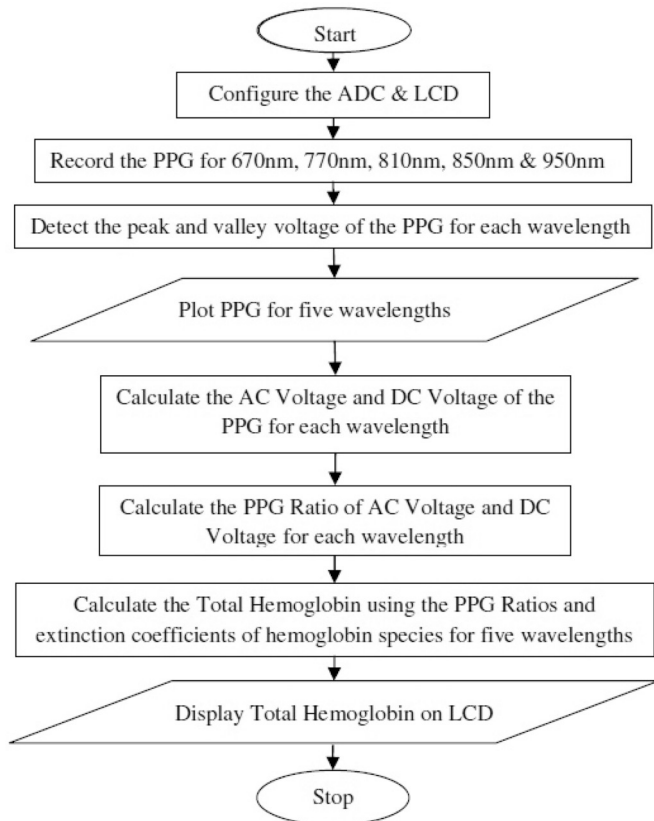


Fig. 5. Flowchart for Hemoglobin estimation using five wavelengths PPG.

device to estimate total hemoglobin. Here the LCD and ADC is configured. The PPG is recorded for five wavelengths of LEDs (670 nm, 770 nm, 810 nm, 850 nm & 950 nm). Next, the peak and valley voltages of the PPG signal are detected for each wavelength. The PPG signal was plotted on the serial Monitor and saved for further offline analysis. The AC voltage (Max Voltage — Min Voltage) and DC voltage (Minimum Voltage) for each wavelength are calculated. Next, the PPG Ratio of AC voltage and DC voltage for each wavelength is calculated. Total hemoglobin of the subject for five wavelengths is calculated using well-known extinction coefficients of oxy-hemoglobin and deoxy-hemoglobin and the PPG ratio for five wavelengths. Finally, the total Hemoglobin level with five wavelengths using non-invasive methods is displayed. The flowchart depicting the details steps is shown in Fig. 5.

4. Result and discussions

The entire experimental setup designed to measure non-invasive hemoglobin levels using five wavelengths of LED was safe because less power was emitted and the fingertip was exposed to less than one minute when placed inside the finger probe. Therefore, no ethical approval was required. Also, the consent of the subject was taken for undergoing analysis during non-invasive hemoglobin measurement. The PPG signals were recorded, while the subjects were in the seated rest position. The invasive measurement was done with same subject in the pathology laboratory to validate with the result.

Each LED was triggered in a sequential manner and kept ON for four seconds for acquiring the PPG signal with one second gap between two PPG signals. It was observed that the amplitude of the PPG signal changes from one wavelength to other wavelength due to the response of hemoglobin as shown in Fig. 6. Also, there could be slight variations with the signal pattern exhibited by the single wavelength itself due to the statistical variation in heart rate based on mental stress of the subject. Also, the PPG can vary based on the skin texture. If the

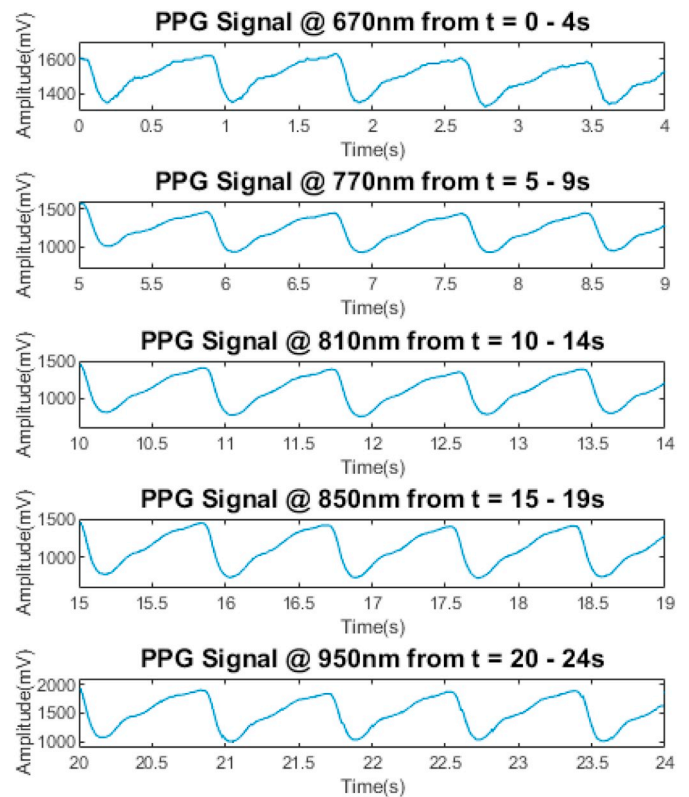


Fig. 6. Signal conditioned PPG signal for different wavelengths.

hemoglobin concentration is high, more incident light is absorbed by it and less light is transmitted producing a small PPG signal and vice versa. Taking into account all of this an algorithm was created to estimate total hemoglobin in the blood.

The result Table 1 of indicates the non-invasive measurement of total hemoglobin using three wavelengths PPG and five wavelengths PPG. Also, a comparative study was made with the hemoglobin measured using the invasive method in the pathology laboratory.

The hemoglobin was estimated with three wavelengths of PPG, $\lambda_1 = 670$ nm, $\lambda_2 = 810$ nm and $\lambda_3 = 950$ nm. The blue rhombus dotted line indicates reference hemoglobin measured in the pathology laboratory and the red squared dotted line indicates the estimated hemoglobin for the above three wavelengths as shown in Fig. 7. The above values for plotting were obtained from Table 1. It was observed that the total hemoglobin values for $\lambda_1 = 670$ nm, $\lambda_2 = 810$ nm and $\lambda_3 = 950$ nm match with these values obtained from the invasive method. The mean absolute error between the reference values and estimated hemoglobin with the above three wavelengths was 1.3778 g/dL. To improve the accuracy and reduce the error, another two different wavelengths were included. The hemoglobin was estimated with five wavelengths of PPG, $\lambda_1 = 670$ nm, $\lambda_2 = 770$ nm, $\lambda_3 = 810$ nm, $\lambda_4 = 850$ nm and $\lambda_5 = 950$ nm. The green triangle dotted lines indicates the estimated hemoglobin for the above five wavelengths as shown in Fig. 7. The above values for plotting were obtained from Table 1. It was observed that the total hemoglobin values for $\lambda_1 = 670$ nm, $\lambda_2 = 770$ nm, $\lambda_3 = 810$ nm, $\lambda_4 = 850$ nm and $\lambda_5 = 950$ nm match with these values obtained from the invasive method. The mean absolute error between the reference values and estimated hemoglobin with the above five wavelengths was 0.3499 g/dL.

The invasive hemoglobin was measured in the pathological laboratory. At the same time, the hemoglobin was estimated with three wavelengths PPG and five wavelengths PPG. Fig. 8 shows the comparison of eight subjects with estimated hemoglobin in Non-Invasive and Invasive hemoglobin.

Table 1
Total hemoglobin estimation for subjects.

Subjects	Gender	Age	Total hemoglobin measured in Pathology Laboratory (g/dL)	Estimated total hemoglobin with designed system (Non-Invasive method)		Error in total hemoglobin measured	
				Three wavelengths PPG (g/dL)	Five wavelengths PPG (g/dL)	Three wavelengths PPG (g/dL)	Five wavelengths PPG (g/dL)
1	M	37	15.7	14.7137	16.3231	0.9863	0.6231
2	F	47	11.7	10.7470	11.6089	0.9530	0.0914
3	M	30	11.0	9.6064	10.5729	1.3936	0.4271
4	M	28	14.6	13.3068	14.6614	1.2932	0.0614
5	M	22	13.5	14.1778	13.9230	0.6778	0.4230
6	F	21	14.0	11.4301	13.4208	2.5699	0.5792
7	M	20	13.0	12.3778	12.7710	0.6222	0.2290
8	F	21	12.8	10.2734	12.4343	2.5266	0.3657

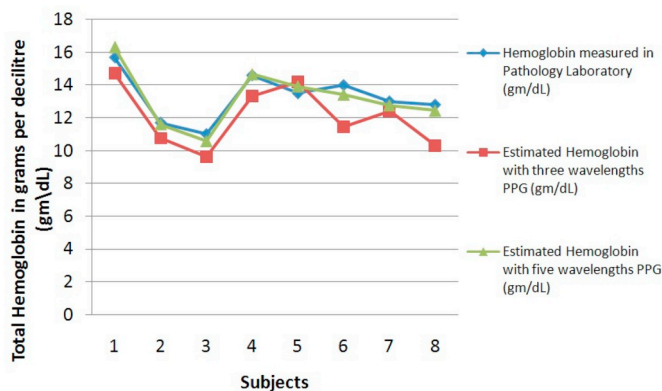


Fig. 7. Total hemoglobin vs subjects.

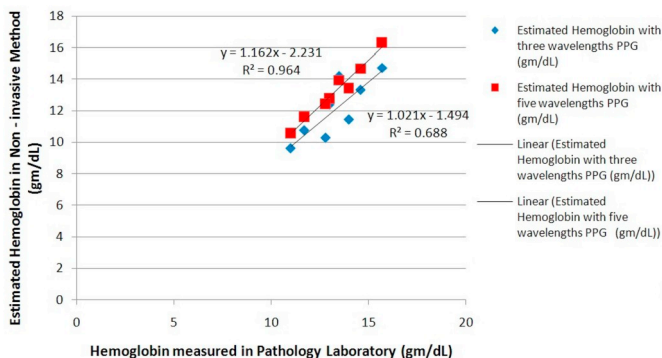


Fig. 8. Estimated hemoglobin with designed system (Non-Invasive) vs Invasive hemoglobin.

Initially, we started with three wavelengths ($\lambda_1 = 670$ nm, $\lambda_2 = 810$ nm and $\lambda_3 = 950$ nm) to estimate hemoglobin and we got the mean absolute error as 1.377 g/dL with the Regression Coefficient of $R^2 = 0.688$, but our main aim was to improve the error below 0.5 g/dL. Hence we selected another two wavelengths, 770 nm and 850 nm to get more information about oxyhemoglobin and deoxy-hemoglobin. By adding these two wavelengths, the mean absolute error significantly improved to 0.3499 g/dL and the Regression Coefficient ($R^2 = 0.964$) which is also quite good. It may be possible to further improve the accuracy by including more number of wavelengths, but the resources required for such design will be large and may not be possible to achieve the protability of the design. Also, the present mean absolute error is 0.3499 g/dL is more than sufficient for the diagnosis. For this reason the five wavelengths PPG is an excellent choice for the measurement of non-invasive blood hemoglobin.

5. Conclusions

This system was developed to estimate total Hemoglobin with a finger probe using five LEDs on the Multi-chip Emitter LED (670/770/810/850/950 nm) and OPT101 a photo detector with Trans-impedance amplifier. It was observed that hemoglobin estimation with three wavelengths (670 nm/810 nm/950 nm) was good with the mean absolute error between reference values and the estimated hemoglobin to 1.3778 g/dL. To improve our measurement accuracy and error, another two different LED wavelengths (770 nm/850 nm) were also used. In this approach, our measurement system produced results with good accuracy, reducing the mean absolute error to 0.3499 g/dL. The five wavelength PPG system designed showed good linearity to estimate hemoglobin with the Regression Coefficient ($R^2 = 0.964$) compared to three wavelengths PPG system with the Regression Coefficient ($R^2 = 0.688$). The PPG signal for the five LEDs was obtained for different subjects. It was concluded that the amount of the PPG signal depends upon the concentration of hemoglobin and the subject's skin color.

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Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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