DEVELOPMENT OF A NON-INVASIVE BLOOD GLUCOSE SENSOR

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ABSTRACT

The objective of this project was to develop a non-invasive blood glucose sensor based on the Beer - Lambert principle of light reflection. This paper describes three main stages of this project. 1) Characterizing the wavelength absorption properties of glucose. 2) Design and construction of a prototype non-invasive blood glucose sensor. The electronic subsystems of this device are separately defined sensor, signal conditioning, and display circuits. The sensor circuit consists of a TCRT1000 reflective optical sensor with transistor output, operating with an emitter/receiver wavelength of 950 nm. The signal conditioning part consists of high-pass filter, non-inverting amplifier, and low-pass filter circuits. From the output signal, we can derive a relationship between blood glucose levels (in micrograms per milliliter) and voltage, which is displayed using a digital oscilloscope. 3) Testing the non-invasive blood glucose sensor. The instrument's calibration curve equation relating blood glucose level with voltage is determined using multiple regression analysis. The device test results demonstrate that blood glucose level is inversely proportional to output voltage, with the relationship described by the equation $y = 252.95x^{-0.779}$, and a coefficient of determination of 0.96.

Keywords: Blood Glucose Meter, Blood Glucose Sensor, Non-invasive Blood Glucose

1. INTRODUCTION

Diabetes is an extremely prevalent disease across the world, with continuously increasing numbers of diagnosed patients [1]. Under healthy conditions, blood sugar levels are measured between 60 and 140 milligrams per deciliter (mg/dL). In diabetic patients this system is dysregulated. Blood sugar levels exceeding the normal range, e.g. 140 to 500 mg/dL, are known as hyperglycemia, whereas levels below 60 mg/dL are known as hypoglycemia [2]. Diabetes is a very dangerous disease if not treated and managed correctly. Vulnerable patients are exposed to increased risk of complications from other diseases, including cardiovascular diseases, stroke, blindness, kidney failure, nervous system diseases and disability [3,4]. Patients must regularly monitor their blood sugar levels, normally 5 - 7 times per day. Conventionally, blood glucose monitoring requires invasive blood collection methods. This is problematic for patients with needle phobia, besides causing a certain degree of pain and discomfort, with increased risk of infection which can induce potentially greater levels of pain and distress [5]. Diabetes also impairs natural wound healing, further cascading these concerns. Reluctance to use invasive blood sampling devices, for the reasons mentioned, prevent patients from knowing their blood sugar level. This form of non-compliance significantly disrupts proper management of diabetes, placing patients at increased risk of serious complications.

At present, there are several non-invasive blood glucose tests. Urine tests provide a rough examination, although their accuracy is below that of measurements taken directly from blood samples; furthermore, urine tests are typically only reliable where the blood sugar level is higher than 180 mg/dL [6]. Breath analysis provides an alternative indirect measure of blood glucose. Devices utilize chemosensors to detect sweet scents from the patient's breath caused by chemical ketones or acetone. Breath analyzers are severely limited in relation to patients who smoke, besides demonstrating inaccuracies and false findings [7].

Currently, near-infrared (NIR) light technology, employing wavelengths of 700 - 1200 nm, is applied in several areas of medical analysis. These applications tend to rely on the Beer-Lambert principle, which states that light absorption of a material is determined by its properties. The most familiar example of this technology is blood oxygen saturation measurement. However, research suggests that other constituents may be detected by analyzing NIR light absorption from vibrations of organic molecules present in the blood. Specifically, the spectral absorption of glucose molecules across the NIR range has been examined [8-9]. By emitting NIR light and detecting its reflected energy, absorption of the reflective medium is calculated, therefore determining the presence of the target molecule. The composition of glucose should be proportional to the amount of light absorbed. When concerning blood glucose analysis, NIR light sufficiently penetrates tissues without inducing any effects of toxicity, thus is suitable for non-invasive investigations [10-12].

A previous study has investigated non-invasive measurement of blood glucose concentration levels using NIR photodiodes centered on wavelengths of 940 and 950 nm [13]. The preferred light wavelength from this study was reported as 950 nm, which more efficiently passed from a light emitting diode through blood glucose concentrations of 0 - 200 mg/dL better than the 940 nm

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source. Importantly, the output voltage from each photodiode was found to be proportional to blood glucose level of the samples under test.

The present study builds upon this research, and aims to incorporate it into the development of a non-invasive blood glucose sensor. Based on the Beer-Lambert principal, utilizing absorption of reflected light, we aim to characterize the relationship between blood glucose level and voltage output from a novel non-invasive glucose meter device.

2. MATERIALS AND METHODS

The methods applied in this project may be separated into three distinct stages. 1) Characterization of glucose light absorption properties. 2) Design and construction of a non-invasive blood glucose sensor. 3) Testing and calibration of the novel blood glucose-sensing device.

2.1 Characterization of glucose light absorption properties

2.1.1 Preparing stock and working solutions of D-glucose

The stock solution of D-glucose (LOBA Chemie) was prepared by dissolving in distilled water until reaching a concentration of 1000 mg/dL. Diluted 25 mL portions of the stock solution with concentrations of 40, 250, and 500 mg/dL were made. To make the appropriate concentration of each working solution, dilutions from the stock solution followed equation (1), assuming the units are in mg/dL.

$$C_1V_1 = C_2V_2 \quad (1)$$

where C_1 is the pre-dilution solution concentration in mg/mL, V_1 is the pre-dilution solution volume in mL, C_2 is the post-dilution solution concentration in mg/dL, and V_2 is the post-dilution solution volume in mL.

If the concentration of stock solution before dilution, C_1 = 1000 mg/dL, the concentrations of working solutions after dilution C_2 = 40, 250, and 500 mg/dL, and the volumes of working solutions after dilution, V_2 = 25 mL, then the volumes of stock solution before dilutions, V_1 = 1, 6.25 and 12.5 mL, respectively.

2.1.2 Testing the wavelength absorption of D-glucose using spectrophotometer

To find the wavelength of maximum absorbance for D-glucose, distilled water was used as a background, and different sample concentrations (40, 250, and 500 mg/dL) of D-glucose solution were examined in turn. The absorption of light caused by each specimen across wavelengths ranging from 400 to 1100 nm was recorded using the spectrophotometer. Each concentration was run in triplicate samples. The wavelength of maximum absorption (λ_{max}), which could readily be absorbed by all three concentrations of the D-glucose solution, was found to be 950 nm, as illustrated in Figure 1. Furthermore, the observed light absorption was proportional to the concentration of D-glucose, as shown in Figure 2.

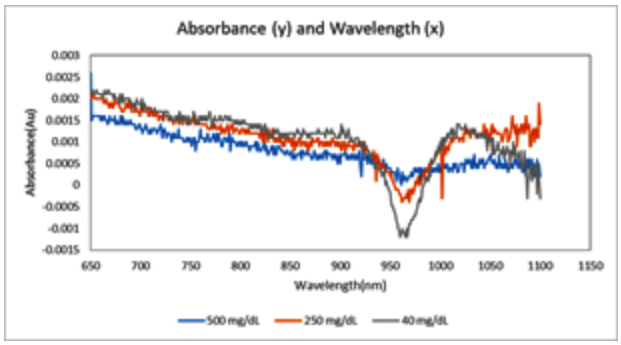

Figure 1. The wavelength absorption of glucose

Concentration of D-glucose (mg/dL)	Absorption of light 1	Absorption of light 2	Absorption of light 3
40	$1.0204x10^{-4}$	$6.1989x10^{-5}$	$2.1219x10^{-4}$
250	$4.0245x10^{-4}$	$3.5429x10^{-4}$	$3.9053x10^{-4}$
500	$5.3692x10^{-4}$	$6.9618x10^{-4}$	$5.0879x10^{-4}$

Figure 2. The absorption at 950 nm of D-glucose

2.2 Design and construction of the non-invasive blood glucose sensor

Design and construction of the non-invasive blood glucose sensor consisted of three electronic subsystems; 1) the sensor circuit; 2) the signal conditioning circuit, and; 3) the display. This general schema for the development of this novel instrument is shown in Figure 3.

Figure 3. The illustrated diagram of development of non-invasive blood glucose sensor

2.2.1 Sensor circuit

For this research, a TCRT1000 reflective optical sensor with transistor output (Vishay Semiconductor GmbH, Heilbronn, Germany), operating at a wavelength of 950 nm, was used in the sensor circuit for blood glucose monitoring. This sensor can be applied to measure blood constituents by placing a finger pad in contact with it; the effective detection distance ranges from 0.2 - 4 mm. The TCRT1000 sensor consists of a short-range infrared LED with a wavelength of 950 nm, functioning as a light transmitter, and a phototransistor which functions as a receiver. This reflective operation combined with

transistor output is well-suited to our application. When the 950 nm light from the LED interacts with blood, the absorption of energy caused by glucose may be detected from the reflected light. The phototransistor receives this reflected light and converts it into an electrical signal; this principle of operation is shown in Figure 4.

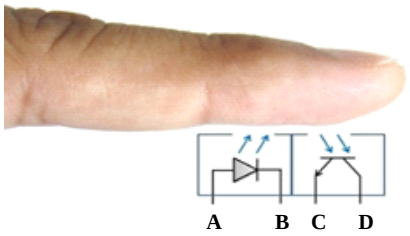

Figure 4. Reflective optical sensor principle

An LED in the TCRT1000 sensor was used as a light transmitter, thus it is necessary to include a current-limiting resistor (R_1) in series, to prevent damaging the LED in the circuit. This resistance can be calculated from equation (2) as follows:

$$R_1 = \frac{V_{CC} - V_F}{I_F} \quad (2)$$

where R_1 is the value of resistance in ohms, V_{CC} is the DC power supply voltage in volts, V_F is the forward voltage drop across the LED in volts, and I_F is the forward current in amperes.

From the TCRT1000 datasheet, V_{CC} = 9 V, V_F = 1.25 V, and I_F = 50 mA, therefore the resistance was calculated according to equation (2):

$$R_1 = \frac{9V - 1.25V}{50mA} = 155\ \Omega$$

The phototransistor in the TCRT1000 sensor was used as a light receiver, thus a resistor (R_2) connected in series with the collector, should be included in the circuit design to limit current, preventing damage to the phototransistor. This resistance can be calculated from equation (3) as follows:

$$R_2 = \frac{V_{CC} - V_{CE(SAT)}}{I_C} \quad (3)$$

where R_2 is the series collector resistance in ohms, $V_{CE(SAT)}$ is the voltage dropped across the collector and emitter of the phototransistor in volts, and I_C is the collector current in amperes.

From the TCRT1000 sensor datasheet, V_{CC} = 9 V, $V_{CE(SAT)}$ = 0.3 V and I_C = 1 mA; hence the resistance (R_2) was determined using equation (3) as follows:

$$R_2 = \frac{9V - 0.3V}{1mA} = 8.7\ \text{k}\Omega$$

R_1 = 155 Ω and R_2 = 8. 7 k Ω were connected in series with the LED and phototransistor, respectively, to complete the sensor circuit, as shown in Figure 5.

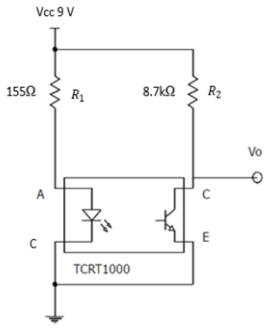

Figure 5. Sensor circuit

2.2.2 Signal Conditioning

The raw signal output obtained from the sensor circuit consists of a small amplitude pulse (max. 200 mV), unsuitable for making accurate measurements of light absorption by glucose. This periodic low-amplitude signal therefore requires modification with a signal conditioning circuit. This consisted of high-pass filter, non-inverting amplifier, and low-pass filter circuits, after which the output signal was displayed with an oscilloscope.

2.2.2.1 High-pass filter circuit

The high-pass filter circuit was used to remove any low-frequency noise, such as baseline wander, to ensure stability of light absorption measurements. This circuit only allows signals with a frequency exceeding the cutoff point to flow, and directs frequencies below the cutoff point to ground; thus diminishing low-frequency signal amplitudes. The desired cutoff frequency in this design was selected to accommodate the sensor pulse rate frequency of 0 .5 - 4 Hz. Implementation of a first-order high-pass filter was achieved by connecting a series capacitor with a resistor tied to ground. To determine the cutoff frequency value of 0.5 Hz, using a capacitance (C_1) of 0.1 µF, the resistance (R_3) of the high-pass filter circuit was calculated from equation (4), as shown:

$$R = \frac{1}{2\pi f_C C_1} \quad (4)$$

$$R_3 = \frac{1}{2\pi f (0.5Hz)(0.1\mu F)} = 3\ \text{M}\Omega$$

Hence the high-pass filter circuit used capacitance C_1= 0.1 µF and resistance R_3 = 3 MΩ.

2.2.2.2 Non-inverting amplifier circuit

In this device, a non-inverting amplifier circuit was used to amplify the output signal from the sensor circuit before reading measurements of blood glucose concentration. This maintained input and output signals in-phase, while amplifying to enhance the resolution of sensor output changes; enhancing the detection of small signal changes from the sensor circuit output, therefore making the device more sensitive to changes in light absorption. A low-power operational amplifier integrated circuit (LM358; Motorola Semiconductors, Hong Kong) was used in the non-inverting amplifier circuit. In order to increase the peak pulse voltage from the sensor (V_{in}) from 200 mV to a maximum output voltage (V_{out}) of 4V, an amplificatory gain (A_V) of 20 for the non-inverting amplifier circuit was selected, as follows:

$$A_V = \frac{V_{out}}{V_{in}} = \frac{4V}{200mV} = 20$$

To achieve this level of amplificatory gain, a potential divider network is used to supply feedback to the inverting input of the operational amplifier. To determine the correct resistance values for this circuit, the first value was allocated an arbitrary value; i.e. $R_i = R_4$ = 10 kΩ. The feedback resistance ($R_f = R_5$) in the non-inverting circuit was then calculated from equation (5), as follows:

$$A_V = \frac{V_{out}}{V_{in}} = 1 + \frac{R_f}{R_i} \quad (5)$$

$$R_f = R_i(A_V - 1)$$

$$R_5 = R_4(A_V - 1)$$

$$= 10k\Omega(20 - 1) \ = 190k\Omega$$

Therefore, the non-inverting amplifier circuit used an LM358 op-amp IC, and resistors R_4 and R_5 were 10 kΩ and 190 kΩ, respectively.

2.2.2.3 Low-pass filter circuit

The low-pass filter circuit allows signal frequencies below its cut-off frequency point (f_c) to pass through freely, while attenuating those of higher frequencies. In this device, a low-pass cutoff frequency of 5 Hz was selected, according to the output characteristics of the sensor circuit. A first-order low-pass filter design can be achieved by connecting a series resistor with a capacitor tied to ground. With a capacitance (C_2) of 0. 1 µF arbitrarily chosen, the resistance (R_6) in the low-pass filter circuit could be calculated from equation (6), as follows:

$$R = \frac{1}{2\pi f_C C_2} \quad (6)$$

$$R_6 = \frac{1}{2\pi f(5Hz)(0.1\mu F)} = 318k\Omega$$

Thus, the low-pass filter circuit consisted of a capacitance C_2 = 0.1 µF and resistance R_6 = 318 kΩ.

To summarize, the non-invasive blood glucose sensor circuitry was composed of a sensor circuit, incorporating series current-limiting resistors, R_1=155 Ω and R_2 = 8.7 kΩ, with a TCRT1000 reflective 950 nm optical sensor; the signal conditioning stage consisted of a high-pass filter circuit, with C_1 = 0. 1 µF and R_3 = 3 MΩ, a non-inverting amplifier circuit using the LM358 op-amp and resistances R_4 =10 kΩ and and R_5= 190kΩ, followed by a low-pass filter circuit with C_2 = 0.1 µF and R_6 = 318 kΩ. The circuit diagram for this design is provided in Figure 6, and the fully-constructed non-invasive blood glucose sensor device within an electronic casing is shown in Figure 7.

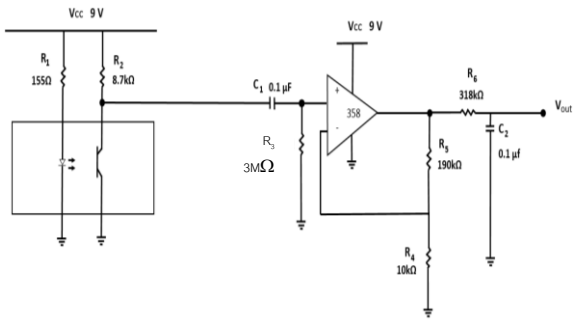

Figure 6. Non-invasive blood glucose sensor circuit

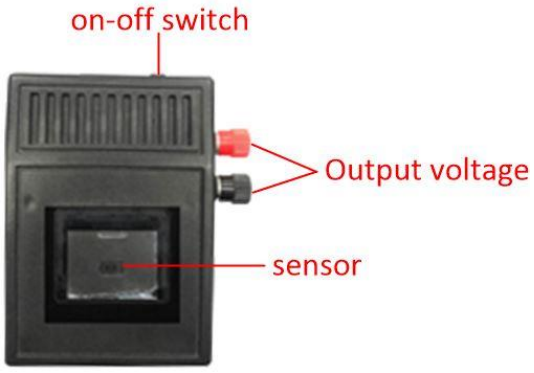

Figure 7. Prototype device with enclosure

2.3.3 Display

A digital oscilloscope (TDS 1001B; Tektronix, USA) was used to display the output voltage signal from

the non-invasive blood glucose sensor. The output generated by the instrument in response to differing concentrations of blood glucose was shown on the oscilloscope screen. Henceforth, the signal read by the oscilloscope was used to evaluate the relationship between blood glucose levels (mg/dL) and output voltage (V) of the device.

2.3 Non-invasive blood glucose sensor test

To develop the non-invasive blood glucose sensor device for practical use, it was an essential first-step to perform testing to establish the calibration curve of the device. By determining an equation to describe the relationship between blood glucose level and the output voltage, we can assess the suitability of this device for real-world applications. The multiple regression analysis method was used. Non-invasive blood glucose sensor voltage output recordings were compared with conventional blood sample measurements taken from a sample of 107 subjects; 83 from Luk-hok Health Promoting Hospital, Pathumthani, Thailand, and 24 from Chaloem Phra Kiat Health Promotion Hospital, Ban Klang, Pathumthani, Thailand. Four stages of this process are described as follows:

1. Record the blood glucose level in mg/dL from each subject by conventional blood sampling and analysis.

2. Record the voltage output of the non-invasive blood glucose sensor from each subject. To obtain this measurement, the subject places their finger in contact with the sensor, and the output voltage signal was displayed on the oscilloscope screen, as shown in Figure 7.

Figure 7. Non-invasive blood glucose sensor test

3. Plot a graph of blood glucose level (mg/dL) versus device output voltage (V).

4. Find the equation of this calibration curve relating blood glucose level with device output voltage, and quantify the precision of this relationship.

3. RESULTS

The frequency of light absorbed by glucose was determined using a spectrophotometer. Light wavelengths over the range of 400 - 1000 nm were transmitted through 40, 250, and 500 mg/dL samples of D-glucose, which correspond to concentration levels found in the blood. The results indicate that light absorbed by D-glucose in the range of 40-500 mg/dL occurs optimally at a wavelength of 950 nm.

The effectiveness of the non-invasive blood glucose sensor design, as described above, was evaluated by comparing the voltage output with actual blood samples, taken and analyzed by conventional methods. This enabled us to calculate the calibration equation, relating output voltage with blood glucose level. Samples from each subject were analyzed with a conventional glucose meter (Model; Manufacturer, Location), with blood glucose level measured in mg/dL. Immediately afterwards, the subject placed a figure upon the non-invasive blood glucose sensor, and the stable peak output voltage of the device was recorded. Example output viewed on the oscilloscope display in response to subjects with blood glucose levels of 99, 127, and 332 mg/dL are shown in Figure 9, 10, and 11 respectively.

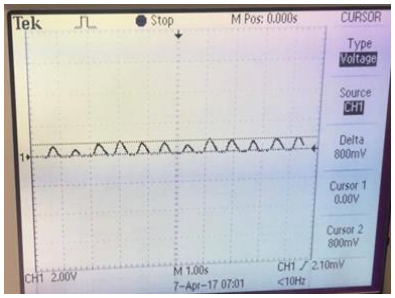

Figure 9. The output voltage signal of glucose level at 332 mg/dL

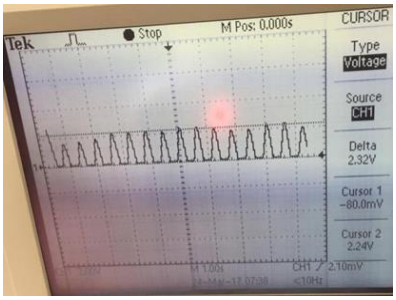

Figure 10. The output voltage signal of glucose level at 127 mg/dL

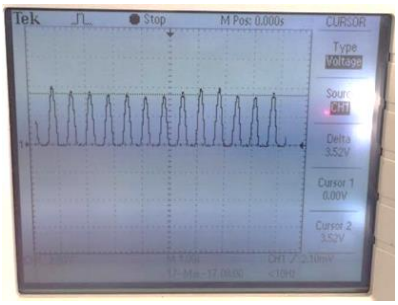

Figure 11. The output voltage signal of glucose level at 99 mg/dL

From the non-invasive blood glucose sensor test, data from 107 subjects were recorded. Blood glucose levels versus device output voltages are plotted in Figure 12. This graph illustrates the relationship between the blood glucose concentration on the y-axis and output voltage on the x-axis.

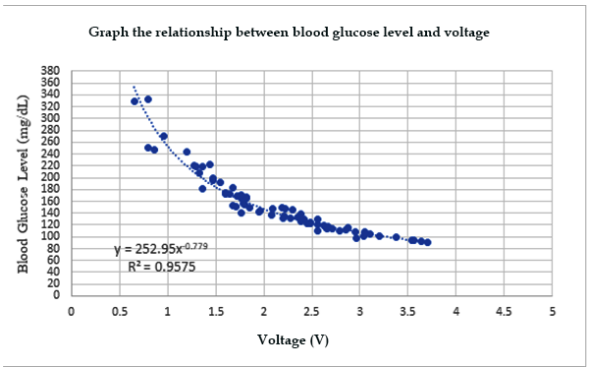

Figure 12. Graph of the relationship between the blood glucose concentration in unit mg/dL and the voltage in unit Volt

4. DISCUSSION

Analysis of D-glucose solution NIR light absorption (Figure 1) indicates that 950 nm wavelength light is maximally absorbed. By examining the spectral absorption properties of D-glucose concentrations ranging from 40 - 500 mg/dL, which cover levels found in the blood, we established that light of 950 nm wavelength was suitable for use in non-invasive blood glucose measurement. This is in agreement with a previous study comparing 940 and 950 nm wavelength light sources [13].

Therefore, in the development a non-invasive blood glucose sensor, it was appropriate to incorporate the reflective optical sensor (TCRT1000) functioning with a 950 nm wavelength. Using this component, blood glucose level may be detected by placing a finger on the sensor. The sensor detects glucose by transmitting light from an LED through the tissue and measuring the degree of absorption reflected back to a phototransistor, which converts the reflected light into a pulsed electrical voltage output. When 950 nm light penetrates a substance containing glucose (at least with concentrations ranging from 40-500 mg/dL), energy is absorbed, and the reflected light intensity will be proportionally decreased, following the Beer-Lambert principle, to represent the level of glucose contained within the sample.

When the graph of blood glucose concentration versus device output voltage is plotted, there is evidently a close relationship the two. It may be noted that a low potential difference from the device indicates that the concentration of blood glucose being measured is high, whereas if the potential difference is high, the concentration of blood glucose being measured is low, therefore the concentration of blood glucose level can be said to demonstrate an inversely proportional relationship to the voltage output from the non-invasive blood glucose sensor.

When using the multiple regression analysis method analyze the recorded data to obtain the equation of relationship between the concentration of blood glucose level and the voltage. The equation was $y = 252.95x^{-0.779}$ where y was the concentration of blood sugar level in mg/dL and x was the voltage in volt and the coefficient of determination was 0.96.

In the testing phase of the project, we struggled to obtain blood glucose samples in the hypoglycemic (70 - 90 mg/dL) and hyperglycemic (250 - 350 mg/dL) range. This led to a data imbalance, with most of the subjects falling within the healthy range of blood glucose concentration. This could potentially decrease device accuracy at either extreme. To improve this situation, future work my involve testing greater numbers of hypoglycemic and hyperglycemic patients, thus obtaining more representative calibration data.

Another issue was encountered when the finger placed upon the sensor was not perfectly stationary. Due to high sensitivity, this produced erratic readings. In this case, subjects were instructed to hold their finger absolutely still while waiting for the sensor readings to stabilize. Only after waiting for voltage pulse output to display a consistent peak value was the voltage measurement recorded. For practical applications, this design constraint must also be addressed.

5. CONCLUSION

This paper has presented the development of a non-invasive blood glucose sensor. Using a reflective optical sensor (TCRT1000) operating at a 950 nm wavelength, this device can measure the concentration of blood glucose level non-invasively, by simply placing a finger on the sensor. This instrument was tested across a range of blood glucose levels from 90 - 332 mg/dL, with the

corresponding output voltage signal displayed via an oscilloscope screen. The relationship between blood glucose concentration and output voltage was described by the equation $y = 252.95x^{-0.779}$, which demonstrated a coefficient of determination of 0.96 and root mean squared error of less than 11 mg/dL. Achieving this level of performance is highly promising, and lends support for further development of the non-invasive blood glucose sensor.

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