Faculty of Electronic and Computer Engineering

DEVELOPMENT AND ANALYSIS OF NEAR-INFRARED SPECTROSCOPY TECHNIQUE FOR NON-INVASIVE BLOOD GLUCOSE MONITORING SYSTEM

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Master of Science in Electronic Engineering

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2019

DECLARATION

I declare that this thesis entitled “Development and Analysis of Near-Infrared Spectroscopy Technique for Non-invasive Blood Glucose Monitoring System” is the result of my own research except as cited in the references. The thesis has not been accepted for any degree and is not concurrently submitted in the candidature of any other degree.

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Date :…….....……..….

APPROVAL

I hereby declare that I have read this thesis and in my opinion this thesis is sufficient in terms of scope and quality for the award of Master of Science in Electronic Engineering.

Signature :…….....……....….

Supervisor Name :…DR. WIRA HIDAYAT BIN MOHD SAAD….

Date :…….....….....

DEDICATION

For the sake of Allah, my Creator and my Master,

My great messenger, Muhammad S.A.W who taught us the purposes of life,

my beloved mother and father

ABSTRACT

Blood glucose monitoring is necessary for diabetes management therapy, where the common method used is an invasive glucose meter that involves finger prick for blood sample which can cause discomfort and skin injury. Painless monitoring of blood glucose would improve patient's quality of life, therefore the development and analysis of near-infrared (NIR) spectroscopy technique for non-invasive blood glucose monitoring system was proposed in this research. An appropriate conditional circuit for photodiode was constructed and 3D sensor casing was designed for output signal stability and noise elimination. The NIR light-emitting diode (LED) with wavelengths of 1050 nm, 1200 nm, 1300 nm, 1450 nm, and 1550 nm and Indium Gallium Arsenide (InGaAs) photodiode were employed in the in-vitro analysis and the Dextrose solution with different concentrations was used as samples. Based on the analysis on the result of the in-vitro experiment, the NIR LED with the wavelength of 1450 nm had the best coefficient of correlation (R^2) and it is used in the development of non-invasive blood monitoring device system. The in-vivo experiment utilises humans as subjects. The different area of the human body has a different absorption capability based on tissue composition and thickness. By considering that, three sensing areas, which are the finger, the area between the thumb and index finger, and earlobe, were selected for measurement. By referring to the measurement of the conventional invasive glucose meter, the earlobe area showed the best consistency of voltage output compared to other areas and this area was used to place the sensor prop for blood glucose measurement. A prototype of non-invasive blood glucose with the algorithm to convert voltage reading to glucose reading was developed based on the acquisition of the experiments that have been carried out. This prototype device has an LED indicator to alert the user about the condition of glucose level and Android application to monitor the blood glucose reading. In addition, this system of non-invasive blood glucose had also been developed with the temperature and motion parameters control for stability during the measurement. The Clarkson Error Grid (CEG) analysis was used to determine the accuracy of the measurement and the highest value of R^2 indicates a good correlation between the measurement of the proposed device system and conventional invasive glucose meter. Based on the tests performed, the algorithms constructed based on a single subject demonstrate a high reading accuracy The developed device system presented here has been proven to show a good correlation between NIR transmittance and blood glucose reading. However, as such an experimental device is not Food and Drug Administration (FDA) approved, it should only be used for academic or informative purposes, and should not be used for any medical decision-making process.

ABSTRAK

Pemantauan glukosa darah adalah satu keperluan kepada terapi pengurusan diabetes, dimana kaedah lazim yang digunakan adalah meter glukosa invasif yang melibatkan tusukan jarum pada jari untuk mendapatkan sampel darah yang boleh menyebabkan ketidakselesaan dan. kecederaan pada kulit. Pemantauan glukosa darah yang tidak menyakitkan akan meningkatkan kualiti hidup pesakit kencing manis dan oleh sebab itu, pembangunan dan analisis terhadap teknik spektroskopi inframerah dekat (NIR) untuk sistem pemantauan glukosa darah yang tidak invasif dicadangkan dalam kajian ini. Sebuah litar bersyarat yang sesuai untuk fotodiod dibina dan selongsong pengesan 3D direkabentuk untuk kestabilan isyarat keluaran dan penyingkiran bunyi hingar.. Diod pemancar cahaya (LED) NIR dengan jarak gelombang 1050 nm 1200 nm, 1300 nm, 1450 nm, dan 1550 nm dan fotodiod Indium Galium Arsenide (InGaAs) digunakan dalam analisis in-vitro dan larutan Dextrose dengan kepekatan yang berbeza digunakan sebagai sampel. Berdasarkan analisis keputusan eksperimen, NIR LED dengan panjang gelombang 1450 nm mempunyai pekali korelasi (R^2) terbaik dan ianya digunakan dalam pembangunan sistem peranti pengawasan darah yang tidak invasif. Subjek manusia digunakan dalam eksperimen in vivo sebagai sampel. Kawasan yang berbeza pada tubuh badan manusia mempunyai keupayaan penyerapan berbeza berdasarkan komposisi tisu dan ketebalan tisu. Setelah mengambil kira semua itu, tiga kawasan penderiaan, iaitu jari, kawasan antara jari ibu dan jari telunjuk, dan cuping telinga telah dipilih bagi pengukuran. Dengan merujuk kepada ukuran invasif meter glukosa konvensional, kawasan cuping telinga menunjukkan konsistensi terbaik voltan keluaran dan kawasan ini digunakan untuk meletakkan peralatan pengesan untuk pengukuran glukosa darah. Sebuah prototaip glukosa darah tidak invasif dengan algoritma untuk menukar bacaan voltan kepada bacaan glukosa dibangunkan berdasarkan hasil eksperimen yang telah dijalankan. Peranti prototaip ini mempunyai penunjuk LED untuk memberi amaran kepada pengguna mengenai keadaan aras glukosa dan aplikasi Android untuk memantau bacaan glukosa darah. Sebagai tambahan, sistem ini juga telah dibangunkan dengan kawalan suhu dan pergerakan parameter untuk kestabilan semasa ukuran. Analisis Grid Ralat Clarkson (CEG) digunakan untuk menentukan ketepatan pengukuran dan nilai R^2 yang tertinggi menunjukkan korelasi yang baik antara pengukuran sistem peranti yang dicadangkan dan meter glukosa invasif konvensional. Berdasarkan kepada ujian yang telah dijalankan, algoritma yang dibina berdasarkan subjek tunggal menunjukkan ketepatan bacaan yang tinggi. Sistem peranti yang dibangunkan telah terbukti menunjukkan korelasi yang baik antara kehantaran NIR dan glukosa darah. Walau bagaimanapun, kerana peranti percubaan itu tidak diluluskan oleh Pentadbiran Makanan dan Ubat-Ubatan (FDA), ia hanya boleh digunakan untuk tujuan akademik atau dapatan data, dan tidak boleh digunakan untuk proses membuat keputusan dalam perubatan.

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Special thanks to all my peers, my beloved late mother, father and sibling for their moral support and financial in completing this study. Lastly, thank you everyone who had been to the crucial parts of realization of this project. Not forgetting, my humble apology as it is beyond my reach personally mentioned those who are involved directly or indirectly one to one.

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LIST OF ABBREVIATIONS

CGMS	-	Continuous Glucose Monitoring System
NIR	-	Near-infrared
UI	-	User Interface
BLE	-	Bluetooth Low Energy
3D	-	Three-Dimensional
LED	-	Light-Emitting Diode
InGaAs	-	Indium Gallium Arsenide
WHO	-	World Health Organization
ISF	-	Interstitial Fluid
FDA	-	US Food and Drug Administration
CE Mark	-	European Commission
CO2	-	Carbon Dioxide
MIR	-	Mid-infrared
IR	-	Infrared
OCT	-	Optical Coherence Tomography
ConA	-	Concanavalin A
MHC	-	Metabolic Heat Conformation
Hb	-	Haemoglobin
SNR	-	Signal-to-Noise Ratio

Ge	-	Germanium
Si	-	Silicon
PbS	-	Lead Sulfide
Insb	-	Indium Antionide
PbSe	-	Lead Selenide
FGT	-	Fasting Glucose Test
OGTT	-	Oral Glucose Tolerance Test
RMSEP	-	Root-Mean Square Error of Prediction
R2	-	Prediction Correlation Coefficient
Ge	-	Germanium
Si	-	Silicon
PbS	-	Lead Sulfide
SEP	-	Standard Error of Predictions
EGA	-	Error Grid Analysis
PCB	-	Printed Circuit Board
PLA	-	Polylactide
DC	-	Direct Current
PTC	-	Positive Temperature Coefficient
NTC	-	Negative Temperature Coefficient
UV	-	Ultraviolet
IDE	-	Integrated Development Environment
SMBG	-	Self-Monitoring Blood Glucose
ADC	-	Analogue to Digital Converter
IC	-	Integrated Circuit

LIST OF PUBLICATIONS

The research papers produced and published during the course of this research are as follows:

1. Salam, N.A.B.A., bin Mohd Saad, W.H., and Salehuddin, F., Manap, Z.B., Karim, S.A. and Radzi, S.A., 2017. Comparative Study of Different Near-Infrared (NIR) Wavelengths on Glucose Concentration Detection. Journal of Telecommunication, Electronic and Computer Engineering (JTEC) 10(1), pp.2-6.
2. Saad, W.M., Salam, N.A., Salehuddin, F., Ali, M.A. and Karim, S.A., 2017. Study on Different Range of NIR Sensor Measurement for Different Concentration of Glucose Solution. International Journal of Human and Technology Interaction (IJHaTI), 1(1), pp.13-18.
3. Salam, N.A.B.A., bin Mohd Saad, W.H., Manap, Z.B. and Salehuddin, F., 2016. The Evolution of Non-invasive Blood Glucose Monitoring System for Personal Application. Journal of Telecommunication, Electronic and Computer Engineering (JTEC), 8(1), pp.59-65.

Attended conference:

1. International Conference on Telecommunication, Electronic and Computer Engineering (ICTEC) 2017
2. International Conference on Telecommunication, Electronic and Computer Engineering (ICTEC) 2015

CHAPTER 1

INTRODUCTION

1.1 Project background

Diabetes is described as a syndrome of metabolism diseases due to abnormal blood glucose levels in the body. Among Malaysians nowadays, diabetes has become one of the most common diseases (World Health Organization (WHO), 2016). Diabetes is a lifelong illness as the patient is fully dependent on medicines that should be taken on the advice of the doctor to help supply or improve insulin function in the body itself. It can also cause many other diseases that can lead to several complications to the patients. Diabetes is a condition where there is an abnormal level of glucose in the human blood. In the human organism, glucose is the main carrier of the energy and the recommended glucose level varies from 4.9 mmol/L to 5.9 mmol/L within two to three hours after a meal for a healthy individual (Frederick Chee and Tyrone Fernando, 2007). Normally, blood glucose level increases slightly after the meal is taken and the abnormal increases of glucose level in the blood may be caused by the body that loses the ability to produce sufficient insulin or the failure of the body to respond properly to the insulin that has been produced by the pancreas.

In the long term, diabetes can affect other health complications to the patients. Diabetes-related complications include damage to large and small blood vessels, which can lead to heart attack and stroke, and problems with the kidneys, eyes, feet, nerves, and skin as illustrated in Figure 1.1. The risk of most diabetes-related complications can be reduced

by regular screening to detect complications earlier. In order to have a regular screening every day, diabetic patient require a good blood glucose monitoring device.

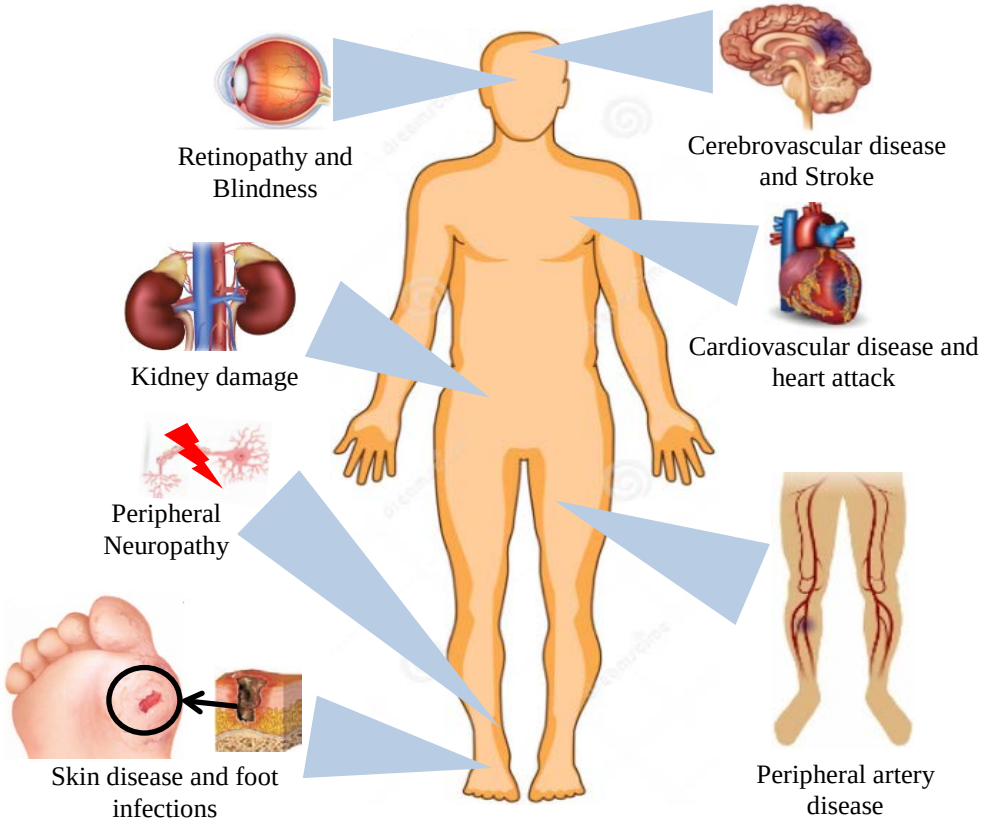


Figure 1.1: The major complication of diabetes to the body

Currently, people with diabetes face a complicated, painful process to measure their blood glucose levels; pricking their finger, extracting a drop of blood onto a test strip and putting it into a portable glucose meter. These processes need to be repeated up to eight times each day to enable effective control and management of diabetes. The introduction of glucose monitoring system for continuous monitoring throughout the day improves the problem faced by the patient (Sato et al., 2012). Some of the continuous glucose monitoring (CGM) system is also equipped with glucose pumps to continuously inject insulin into the body. The commercially available CGM system uses an invasive system

where a needle of the sensor is injected underneath the skin and left there for a week. This study aims to look at a method of using a non-invasive sensory technique specifically by using near-infrared (NIR) optoelectronics sensor.

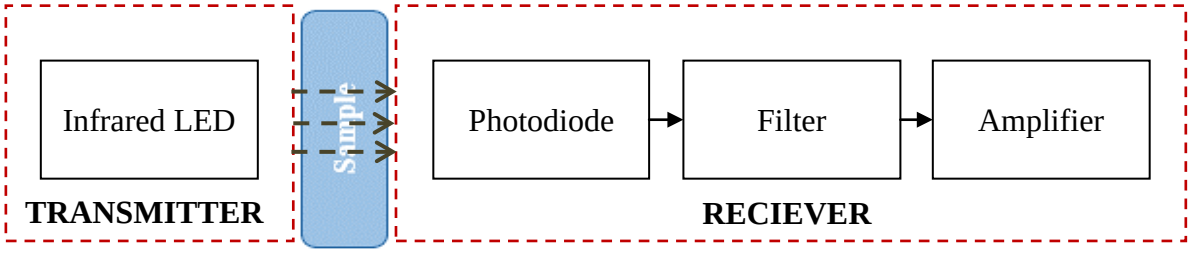


Figure 1.2: Block diagram of a simplified of light detector

Basically, NIR light-emitting diode (LED) is used as the light source and photodiode is used as a light detector to detect the transmitted light passing through the sample as depicted in Figure 1.2. The output from the detector is in the form of voltage reading. This voltage signal will go through a filter and the amplifier to remove the noise and improve the signal. The algorithm needs to be coded into the processing board to process the analogue signal to get the glucose concentration value.

In this study, a working prototype of continuous glucose monitoring system device is developed for sensor testing. The graphical user interface (UI) of the monitoring system is also designed based on an Android platform to make it easy for monitoring and logging the data. The readings can be reviewed through the UI system on the smartphone for further assessment process. The Bluetooth Low Energy (BLE) is used as a wireless communication medium between the sensor device and monitoring device. This communication is widely used in short distance communication between portable devices to a computer/smartphone and this technology also has a good power-saving.

By carrying out the study and development of NIR non-invasive blood glucose monitoring system, it will definitely bring the complete new ways to the diabetic patients to monitor their blood glucose condition and help the diabetic patients to have a detailed understanding of their individual diabetic situation and better daily insulin management for them. With the optimisation technique of low energy and high-speed data transmission, this can improve the practicality of such device for the portable application. In addition, the results of the experiment conducted can be used as a reference by other researchers in the same field.

1.2 Problem statement

Previous researches have tested the NIR sensor without a proper mount, which has caused noise in the output signal (Buda and M. Mohd. Addi, 2014). The sensor source light would be scattered to the surrounding and the receiver sensor could be exposed by the surrounding light during the reading measurement. To ensure that most of the NIR light signals are focused, a suitable sensor mounting casing for the application needs to be designed. As for the consistency of the output signal, the NIR sensor should be kept in a fixed position and this requires the error to be minimized during the experiment. Besides that, a suitable sensor mounting casing enables the user to handle it properly.

NIR Spectroscopy technique has wavelengths ranging from 700 nm until 2500 nm. Therefore, the selection of suitable wavelengths to be used for non-invasive blood glucose detection is difficult (Paul et al., 2012). This has thus led to a study to see the response between the different ranges of NIR wavelengths and different concentrations of glucose solution. In addition, the sensor's position also influences the output results, since different areas of the human body have different absorbing capabilities (Hotmartua et al., 2015) (Yadav et al., 2015). To further justify this, an experiment is conducted in order to

study the response of wavelengths against sensor placement area. There were three different areas of human body tested in order to choose the area with the best response.

The NIR is one of the optical methods for non-invasive blood glucose development. One analysis in order to find the clinical accuracy of NIR non-invasive blood glucose monitoring development is required to ensure this method is valid to be used (Yadav et al., 2015b). The invasive method of detecting glucose is currently the most common and accurate method used to measure blood glucose by comparison to any of the other existing non-invasive methods. Hence, the accuracy of the non-invasive technique of glucose reading needs to be compared with the reading of invasive glucose meter available in the market. A Clarke Error Grid is a standard indicator specifically used to determine the accuracy of glucose monitoring (Kamboh and Khan, 2013). However, the non-invasive method is incapable of giving the required accuracy compared to the invasive method. Due to that reason, this method is not suitable for clinical practices, nevertheless more appropriate for consumer use product.

1.3 Objective

The main goal of this research is to develop the NIR non-invasive blood glucose monitoring systems. Specifically, the objectives of the research are:

i. To develop the prototype of NIR non-invasive blood glucose monitoring system.
ii. To analyse the response of the NIR wavelength towards the concentration of glucose solutions and the target area on the subject for sensor placement.
iii. To investigate and validate the accuracy of near-infrared spectroscopy system developed for non-invasive blood glucose monitoring.

1.4 Scope of project

This scope of work is to design a non-invasive blood glucose method by using NIR spectroscopy technique. The project is limited based on the following explanation. This project uses different NIR wavelengths, which are 1050 nm, 1200 nm, 1300 nm, 1450 nm, and 1550 nm. NIR LEDs from Thorlabs (Thorlabs, 2007) and Indium Gallium Arsenide (InGaAs) photodiode from Hamatsu (Hamamatsu Photonics, 2015) are used as a light source and light detector. A Bluno microcontroller board is used as a processing unit and it is the Arduino microcontroller board integrated with BLE 4.0 module. This board is used to read the analogue data from the circuit and convert it into digital data for data logging. A casing for the sensor holder is designed and printed using a 3D printer. However, this works only for the laboratory experiment and prototype and not for the medical user. In the in-vitro experiment, Dextrose powder and distilled water are used to prepare the glucose concentration solutions to be used as a sample for the experiment. The human samples are used in the in-vivo experiment and that samples consist of more than eighty subjects of diabetic and non-diabetic patients. An algorithm to convert the analogue signal to the glucose reading was developed and loaded into the processor. Besides, the processor also programmed to alert the user about an abnormal condition of glucose level in blood. The results from the project are compared against an invasive glucose meter available in the market. In addition, a UI based on Android platforms is designed to display the data from the Bluno board by using Android Studio software.

1.5 Contribution of work

The goal of the thesis is to develop a non-invasive glucose monitoring system which patient can use on a daily basis at home for monitoring blood glucose condition during normal day-to-day activities. This research is based on NIR spectroscopy technique

and the analysis of NIR spectroscopy based on non-invasive blood glucose application can aid researchers who wish to perform optical spectroscopy. There are lots of research that have been done by the previous researchers based on this technique. However, the non-invasive method reading is inaccurate like the invasive method. This system is incapable of measuring accurate glucose concentration. For instance, its main purpose is to detect the sudden changes of glucose concentration in blood that can lead to hypoglycaemia or hyperglycaemia for diabetic patients and alarm them for medical attention or trigger insulin injection. This project was built by using a minimal budget, but the device would still be able to read the change of blood glucose level. Besides that, an algorithms model that requires user-friendly calibration was implemented. A calibration is needed for every different user to use this device. This work also focuses on developing a technique for a product that is convenient and can be worn by patient without developing other side effects or health hazards like skin irritation, allergic reaction or immune response. Lastly, this research is a major contribution to society and science, especially in the use of NIR technique for glucose concentration detection. Part of the results in this study had been published in several different indexed journal for public access.

1.6 Thesis organisation

This thesis involves chapters which are categorized as; Chapter 1, covering introduction that consists of the project background, problem statement, objectives of the project, scope of research, contribution of work and the key matters that lead to this research.

Chapter 2 features the review on diabetes and the development of blood glucose monitoring. The first subtopic describes the basic introduction of diabetes. The second subtopic explain the methods of blood glucose measurement. The third subtopic is about

the revolutionary history of blood glucose monitoring systems development from the starting generation until presents. Next, several technologies of non-invasive blood glucose monitoring are reviewed in detail, including the working process of the technique, advantage and disadvantage of that technology. The types of NIR detector and, the limitation and challenges of using the NIR absorption spectroscopy are discussed in Subchapter 2.5, alongside the fundamentals of NIR absorption spectroscopy. Furthermore, the subchapter presents the method of experiments and how the data of the experiment were analysed. Finally, the previous research on NIR spectroscopy technique for non-invasive blood glucose monitoring is critically reviewed in order to identify the advantage and limitation of the previous research.

Chapter 3 puts a lot of concern towards the realization of the research objectives which is the methodology of this project. The first stage focuses on design; it includes circuit design, 3D casing design, and software development. After that, the experimental part of the study is elaborated. There are two types of experiments carried out. The first part is in-vitro experiment and the second part is in-vivo experiment. The analysis part of the experiment is explained in Chapters 2 and 3, as the focus is more on the preparation of these two experiments. Lastly, a simple algorithm is developed to convert the analogue signal on the processor board to form value in glucose reading and help for data verification. The flowchart of the algorithm is presented in the last part of this chapter.

Chapter 4 presents the experiment result as well as discussion of the result. The result is divided based on in-vitro and in-vivo experiments. The in-vitro analysis result focuses on the relationship between the NIR wavelengths with the glucose concentrations. Meanwhile, the in-vivo analysis results are based on the few points such as sensor designs, and selection of the target area to place the sensor. This result is discussed in details and the final prototype is developed based on the acquired data in the experiments. The

algorithms used in the controller board are also further discussed in this chapter. The prototype system is verified by comparing the results with the invasive glucose meter present in the market. The results are discussed based on Clarke error grid analysis.

Chapter 5 provides a conclusion and suggestion of possible additions in the project for future improvement to this project.

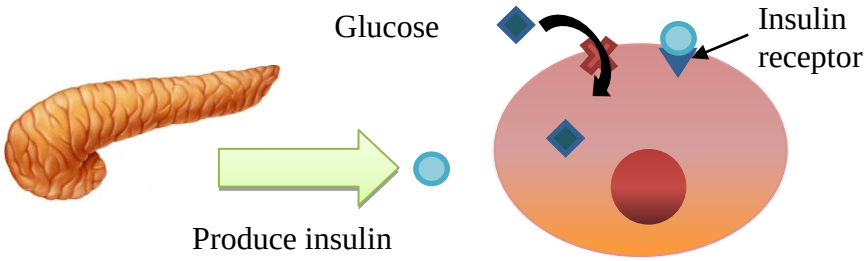
CHAPTER 2

LITERATURE REVIEW

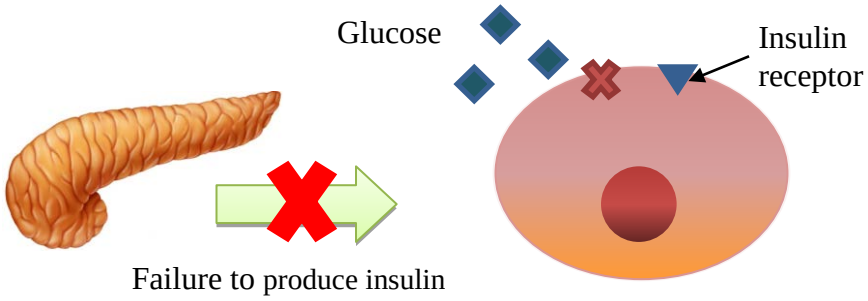
2.1 Introduction

Diabetes is one of chronic disease that occurs either when the body does not use insulin that is produced effectively or the pancreas does not function normally to produce enough insulin for the body. There are three types of common diabetes, which is type 1 diabetes also known as insulin-dependent, juvenile or childhood-onset diabetes. Type 1 diabetes is due to pancreas failure to produce insulin in the body. Type 2 diabetes known as non-insulin dependent or adult-onset diabetes. With type 2 diabetes, the pancreas usually produces some insulin, but either the amount of insulin produced is not enough for the body's needs, or the body's cells are resistant to it. The last type is gestational diabetes (GDM) and this high blood sugar is developing during pregnancy and usually disappears after giving birth (World Health Organization (WHO), 1999). Figure 2.1 illustrates diabetes mellitus in the human system.

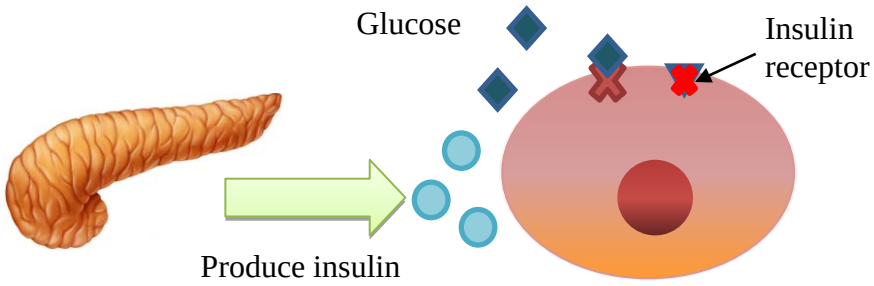
Uncontrolled diabetes and over time can cause high blood sugar (hyperglycaemia) or low blood sugar (hypoglycaemia). These conditions lead to serious complications and damage to many of the body systems such as heart attack, stroke, blindness, kidney failure, lower limb amputation, the nerves and blood vessel problem (World Health Organization (WHO), 2016). World Health Organization (WHO) recommended criteria for the diagnosis of diabetes and intermediate hyperglycaemia as shown in Table 2.1.



(a) Healthy condition and the system work normally



(b) Type 1 diabetes. Pancreas failure to produce insulin



(c) Type 2 diabetes. The pancreas produces insulin, but cells fail to respond to insulin properly

Figure 2.1: The human system based on three conditions; (a) Normal condition (b) Type 1 diabetes (c) Type 2 diabetes

Table 2.1: Recommended target blood glucose level ranges for non-diabetes, pre-diabetes and diabetes (Holt et al., 2011)

Category	Fasting value		2 hours after meal
	Minimum	Maximum	
Normal	70 mg/dL 4.0 mmol/L	100 mg/dL 6.0 mmol/L	< 140 mg/dL < 7.8 mmol/L
Pre-diabetes	101 mg/dL 6.1 mmol/L	126 mg/dL 6.9 mmol/L	140-200 mg/dL 7.8-11.1 mmol/L
Diabetes	> 126 mg/DdL > 7.0 mmol/L		> 200 mg/dL > 11.1 mmol/L

Diabetes is one of the main diseases that causes death worldwide. According to the Global Report on Diabetes by WHO, the number of adults worldwide suffering from diabetes has increased almost quadrupled in 2014 compared to 108 million in 1980, about 422 million adults (World Health Organization (WHO), 2016) Malaysia has the highest number of diabetes mellitus in Asia and among the highest in the world after Saudi Arabia. About 2.5 million adults in Malaysia aged 18 and over have diabetes. Figure 2.2 shows the trends and projections of diabetics among the Malaysian population age 18 years and above from the year 2006 until 2020 (Berita Harian, 2018).

One of the main goals of diabetes treatment is to control blood glucose level in the body within a specified range. Therefore, blood glucose monitoring device is important in helping diabetic patients to monitor their blood glucose level, manage insulin dose, controlling their food intake, and increase their daily activity. The blood glucose level in the body can be checked as often as needed or as recommended by a doctor by using self-blood glucose monitoring. By maintaining good blood glucose level, it can help diabetic

patients to reduce the chance of another complication caused by diabetes (Clarke and Foster, 2012).

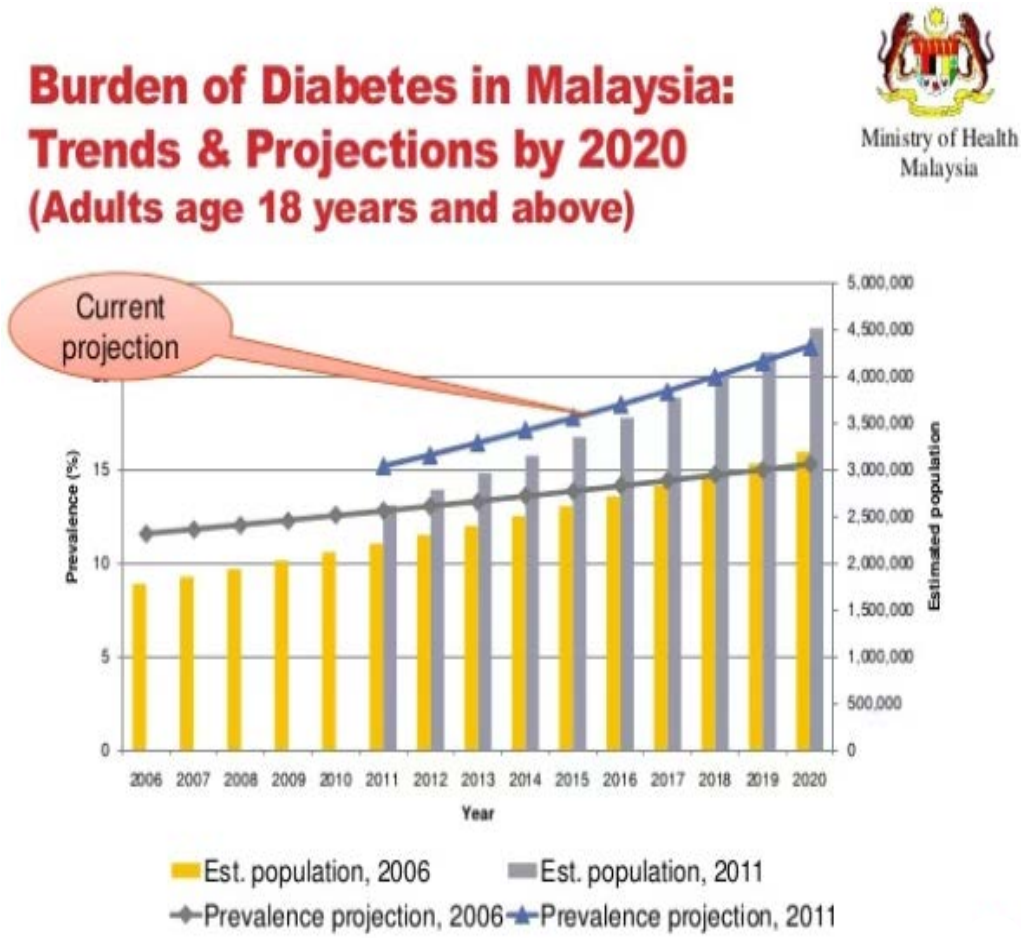


Figure 2.2: Trends and projections prevalence of diabetes in Malaysia by years 2020 (Ministry of Health Malaysia, 2011)

2.2 Blood glucose measurement methods

The blood glucose measuring is classified into three major methods which are invasive, non-invasive and minimally invasive or also known as interstitial as show in Figure 2.3.

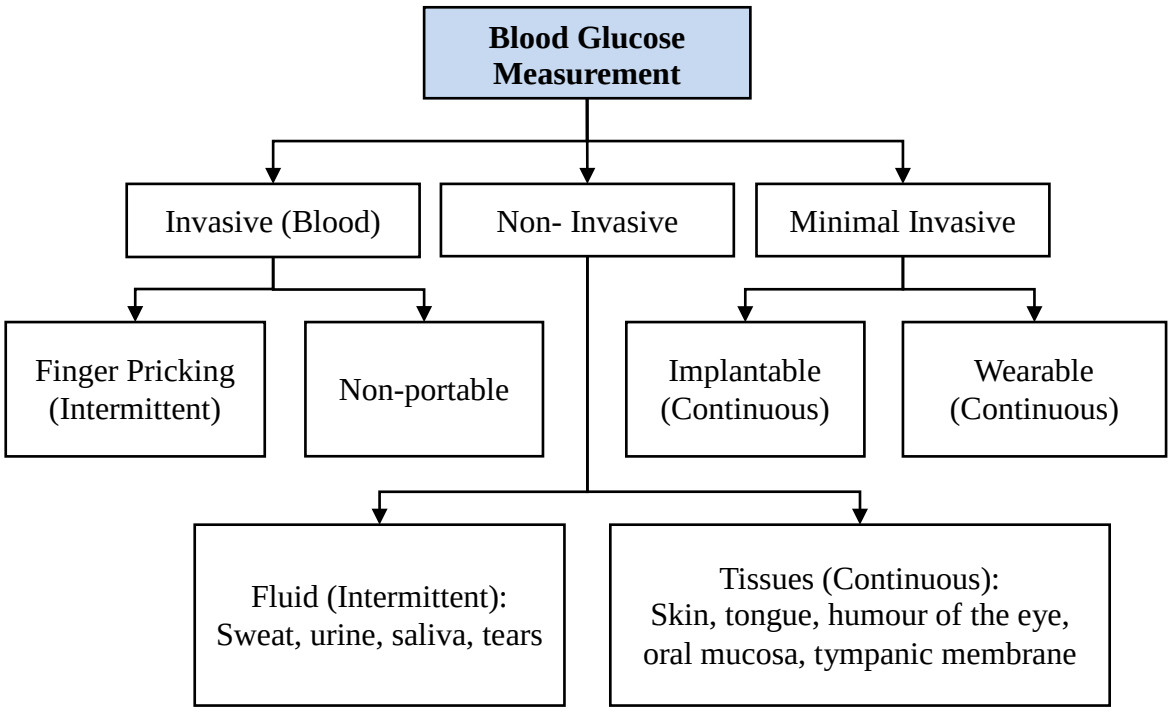


Figure 2.3: Overview of blood glucose measurement technologies (Ferrante do Amaral and Wolf, 2008)

The invasive method is also known as a traditional method of testing the blood glucose and it require a sample of blood. Therefore, this method involves finger pricking by a lancet (a small, sharp needle), putting a drop of blood on a test strip placed into a glucose meter and glucose meter readings are recorded as shown in Figure 2.4. This method most commonly used by patients due to the price of the device is cheaper for beginning than the device that uses partially invasive and completely non-invasive methods.

(a) (b) (c)

Figure 2.4: Blood glucose monitoring approach using a FreeStyle glucose meter; (a) Finger pricking (b) Blood test by glucose meter (c) Self record

The partially or minimally invasive method is developed by using subcutaneous sensors for the measurement of glucose concentration in interstitial fluid (ISF). Due to this approach, patients will suffer from limitations in terms of discomfort, require continuous calibration, and high susceptibility to bio fouling (accumulation of microorganisms) (Vashist, 2012). In-vivo non-invasive is a method for the measurement of glucose without taking blood samples or inserting a substance into the body. Therefore, minimally invasive and non-invasive approaches are gaining popularity in continuous monitoring of blood glucose concentration. These two methods offer many advantages over invasive methods for long time usage. These two methods are discussed in more detail in the subchapter of history of blood glucose monitoring systems development.

2.3 History of blood glucose monitoring systems development

The blood glucose monitoring systems have been introduced since the 70s. Figure 2.5 shows the chronology of the blood glucose monitoring from the first generation until the fourth generation (Klonoff, 2005). The development made to improve the way of measuring the blood glucose is depending on several parameters such as reading accuracy, techniques and the size of the device, and the technology of the embedded systems. This

development also gives effect to the user in term of financial, comfortability, and convenient because the use of needles has been reduced (Yoo and Lee, 2010). The impact of environment pollution will also be reduced since less of the test strip and needle used, less disposable material produced. Each of this generation will be briefly discussed in details on the next coming sections.

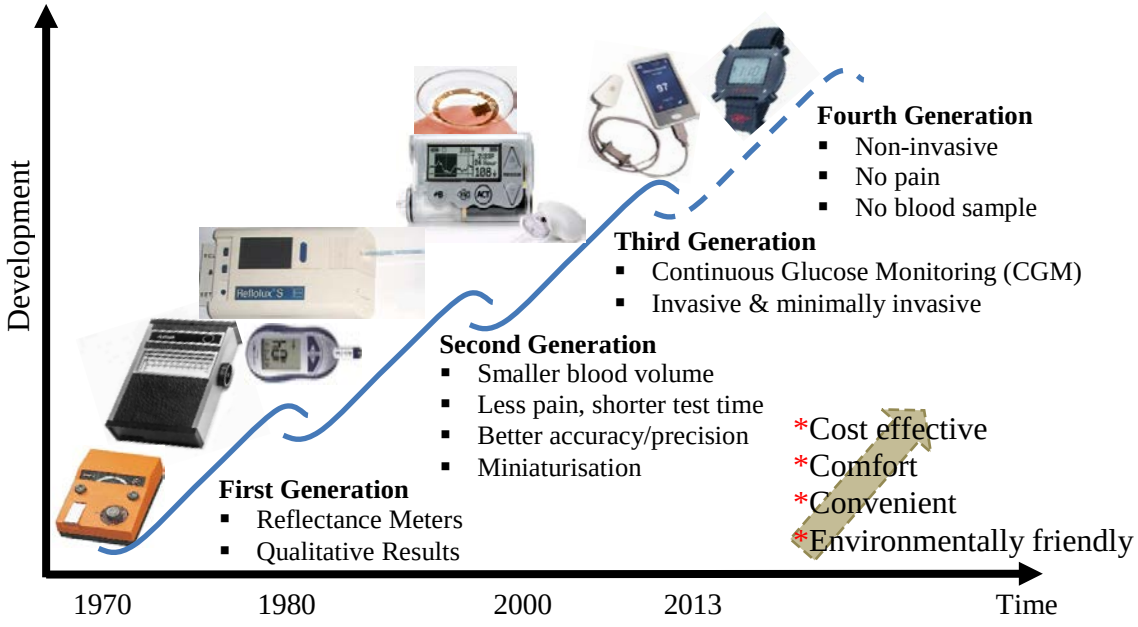


Figure 2.5: The revolution of the blood glucose monitoring device

2.3.1 First generation (invasive)

The first generation of blood glucose monitoring system has been introduced in 1971 by Anton Hubert (Tom) Clemens as an inventor of Ames Reflectance Meter as shown in Figure 2.6 (Newman and Turner, 2005). This glucose meter uses an enzyme test strip where needs the blood sample to be applied onto the test strip and then it washed away after the reading is taken. The blood glucose concentration level is displayed on the meter by a moving pointer on three analogue scales equivalent to 0-4 mmol/L, 4-10 mmol/L and 10-55 mmol/L blood glucose. This instrument is expensive at the time

when it was being introduced, relatively large in size and very heavy (approximately 1 kg). This device required a relatively big amount of blood to be placed in the sensor area approximately 3/8 inch x 1/4 inch. Since every time the blood glucose reading was taken, the blood placement need to be washed, causing this device to be more suitable to be used in the hospital by the physician rather than for personal home used (Clarke and Foster, 2012). This device became the reference design for subsequent reflectance colorimeters such as Eyetone (introduced in 1972) and the Ames Glucometer (introduced in 1971).

Figure 2.6: The first portable blood glucose meter; Ames Reflectance Meter. This image from (David Mendosa, 2005)

In the year of 1975, Yellow Spring Instrument commercialising the YSI 23A model of the glucose analyser as shown in Figure 2.7 (Newman and Turner, 2005) and this device is based on the enzyme catalysed process glucose detection method that has been developed by Clark and Ann Lyons since 1962 (Harper and Anderson, 2010). This device utilised the oxidation of glucose, and subsequently the oxidation of the hydrogen peroxide that formed during the initial response by glucose oxidase and enzyme horseradish

peroxidase respectively (Wang, 2008). In order to do the readings, this device require only 25 µL of whole blood sample yet it was able to improve the detection accuracy in comparison to Ames reflectance meter (Clark and Lyons, 1962). Even though a stationary model linked to the hospital and should be handled by a doctor, this developing technology had become the basis for state-of-the-art handheld devices of the second generation of the blood glucose monitoring, especially for personal home-monitoring.

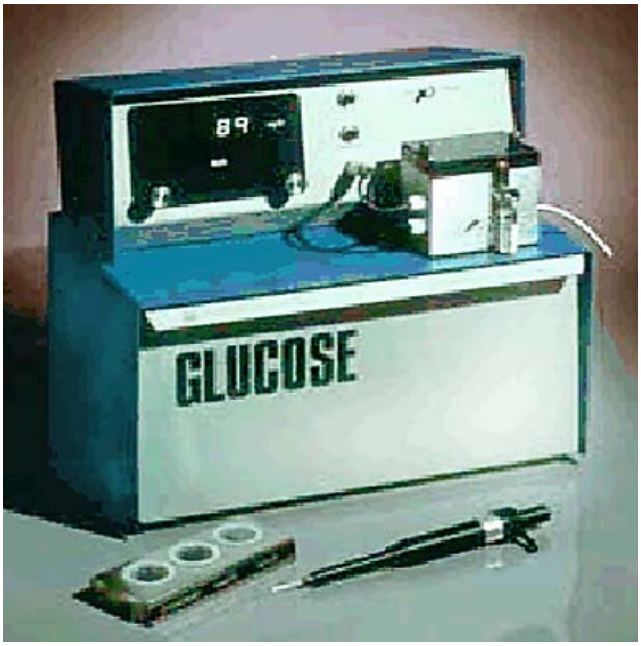


Figure 2.7: The YSI 23A model of the glucose analyser. This image from (Newman and Turner, 2005)

2.3.2 Second generation (invasive)

The first commercialised blood glucose meter for home application is ExacTech® strip produced by MediSense (later it became part Abbott Laboratories) in 1987 (Wang, 2001). This technology based on the first generation of technology that has been introduced by Clark and Ann Lyons as it is utilising an integrated electrochemical ferrocene-derivative mediator as an electron acceptor (Dimeski et al., 2010) (Harper and

Anderson, 2010). There is a lot of improvement to this device that has been done in terms of sample size, test time, glucose reading accuracy, device size, and as well as its functionality for every version that has been released up to until now. This technology widely being used until now in the development of invasive blood glucose monitoring device and it's run by several brands as can be seen in Figure 2.8 (Newman and Turner, 2005).

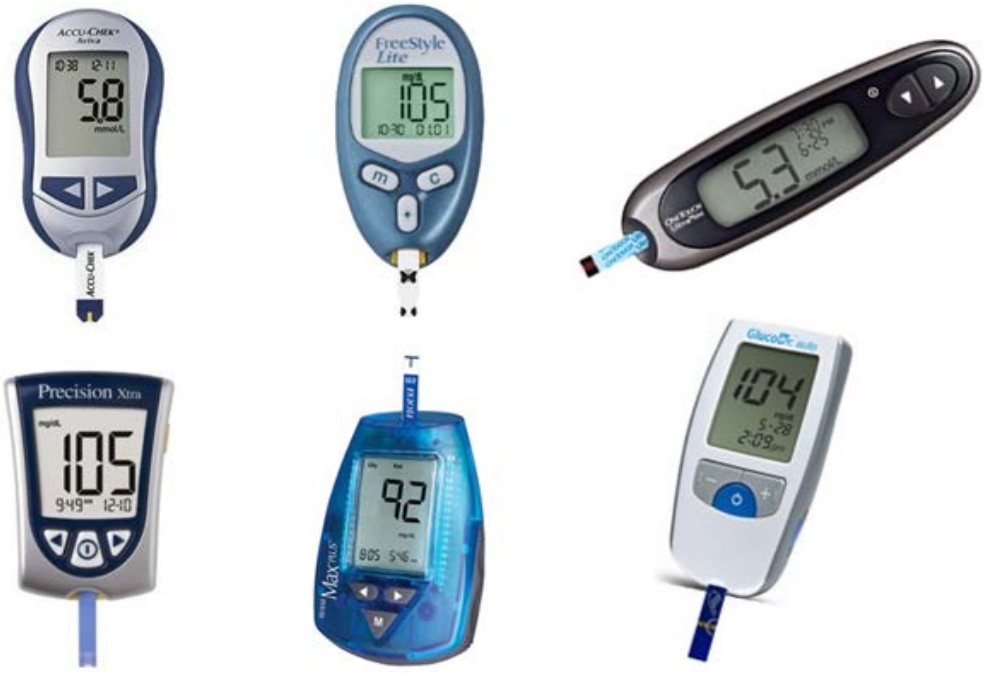


Figure 2.8: Conventional devices of blood glucose meter with different size and design (ACCU-CHEK, FreeStyle, ONETHOUCH, Precision, MaxPlus, GlucoDr.)

The invasive method needs user to spend some money to buy basic needs for blood tests such as needles and test strips as shown in Figure 2.9. The test trips need to be used in a certain time after the container is opened since it have an expiration date.

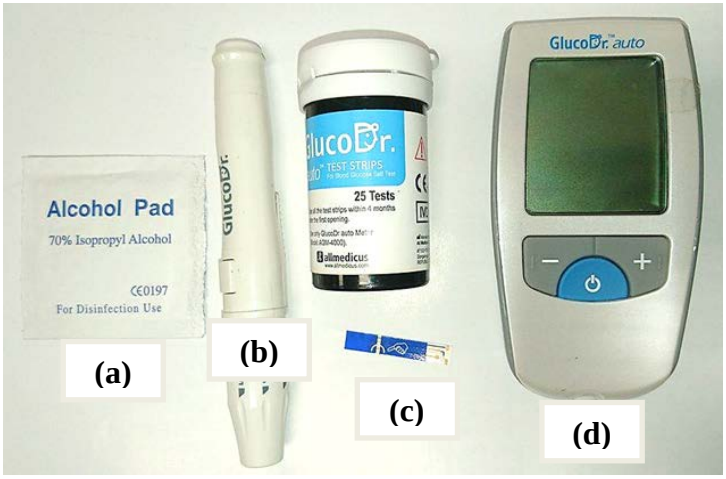


Figure 2.9: Blood glucose meter test kit for blood glucose test; (a) Alcohol Pad (b) Needle Pen (c) Test Strip (d) Test Meter

2.3.3 Third generation (minimally invasive)

The third generation of the glucose monitoring system is more toward the continuous glucose monitoring (CGM) system. This monitoring technology is possible due to the development by using subcutaneous sensors (micro-needle) that can be inserted underneath a skin as shown Figure 2.10 and can be left there for a few days without require to draw of blood or the supplement of medication.

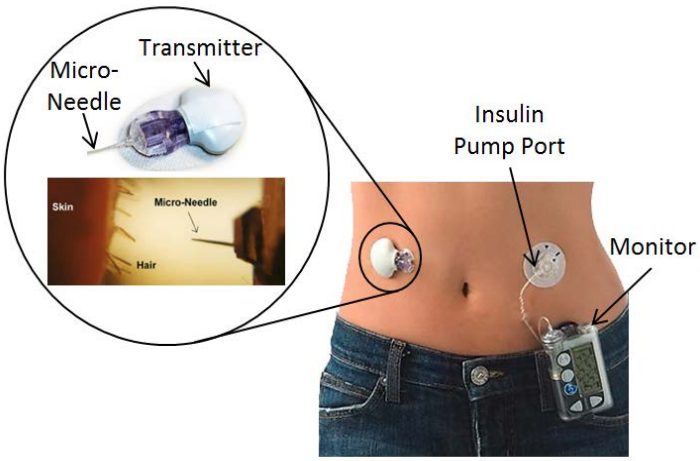


Figure 2.10: The minimally invasive (CGM) device with insulin pump

GlucoWatch biographer is the first CGM device approved by the US Food and Drug Administration (FDA) in 1991, but it is no longer been used. Figure 2.11 shows several types of commercially available CGM device that have been approved by the FDA or carry the European Commission (CE Mark) for Europe market, since the beginning of the twenty-first century (Thomas C. Blevin et al., 2010). This personal CGM device is typically used by diabetes patients and pregnant mother with gestational diabetes symptoms with a recommendation of their physician (Mclachlan and Neal, 2007).

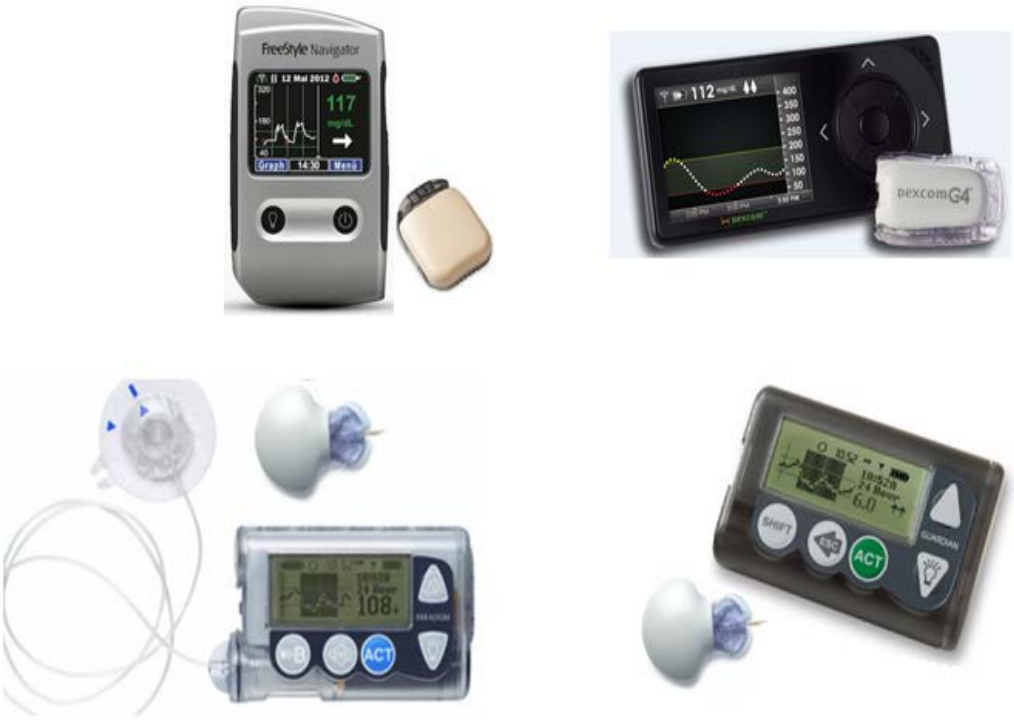


Figure 2.11: Brands and models of CGM device available in market

There are several personal CGM devices that are available with alarms to indicate the changes on reading. Besides, there are also CGM devices that attach together with insulin pump where the whole systems are able to function like pancreases in the body (Bequette, 2010). A personal CGM device is normally owned by a patient

individually. The patient can monitor their glucose level continuously in every minute by using this system. The insulin will be released by insulin pump into the blood circulation with the right dosage that required by the body based on the reading taken. This system is able to give a real-time glucose result that allows for immediate therapeutic adjustments continuously on the insulin intake (Keenan et al., 2009).

2.3.4 Fourth generation (non-invasive)

Nowadays, the latest development of the blood glucose monitoring device is more toward a non-invasive approach. This method is one option for painless blood glucose monitoring with substitute blood with other fluids that could contain glucose substance such as urine, saliva, sweat or tears (Krushinitskaya, 2012). However, continuous non-invasive blood glucose monitoring can only be achieved through direct measurement on the body tissues, like skin and eye (Ferrante do Amaral and Wolf, 2008). The continuous blood glucose concentration is measured by placing the sensors directly onto the human targeted area as demonstrated in Figure 2.12 without any sample of body liquid, ISF or a needle penetrating through the skin for reaching these fluids.

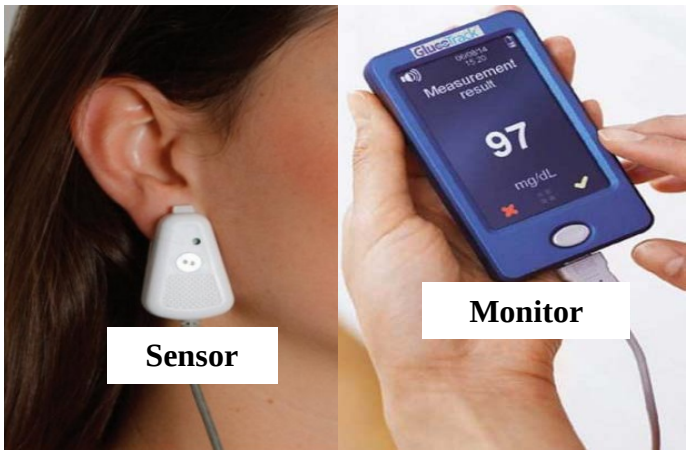


Figure 2.12: GlucoTrack™ non-invasive glucose monitoring device

The studies on the development of non-invasive technology have begun since 1957 and the works are still continuing up to the present. Many researchers have come up with different methodological ideas to develop non-invasive devices for the determination of blood glucose concentration. There are only a few numbers of non-invasive devices that have been produced using different technology as can be seen in Table 2.2 and only a few of them are approved for sale in the certain country by FDA or CE Mark (Vashist, 2013). Because of the method used to read the value of the blood glucose without directly in contact with the blood, most of the non-invasive blood glucose monitoring device is registered under consumer product and not under a medical product. In the next section, some of the available technologies of non-invasive glucose monitoring system development is discussed.

Table 2.2: Non-invasive glucose monitoring devices in the market (So et al., 2012)

Devices	Descriptions
	**Device:** CLEARPATH DS-120 **Company:** Freedom Meditech, Inc. **Year:** 2007 **Technology:** Fluorescent technology **Target area:** Eye **Status:** FDA approved and CE mark approved in December, 2013. **Advantages:** Easy to use and test results are given instantaneously in 6 seconds; does not require patients to fasting before the test. **Disadvantages:** Use as a diabetes screening test and monitoring tool for patients aged around 20-70 years who have a biological lens present. **Reference:** (Freedom Meditech, 2013)

Devices	Descriptions

	**Device:** GlucoWatch © Biographer **Company:** Animas Technologies (Cygnus Inc.) **Technology:** Reverse iontophoresis **Target area:** Wrist skin **Status:** FDA approved in August, 2002 and CE Mark approved. Consequently, this device was withdrawn from the market after 2008. **Advantages:** The measurement done every 10 minutes for up to 13 hours at a time. The fluctuations of skin temperature and perspiration were considered; has an alarm and trend indicators to inform the changes of measurement, event marking, data download, data analysis software and data storage. **Disadvantages:** Pricey; requires warm-up around 2-3 hours, difficulty in calibrating and use an invasive glucose meter for calibration; every 12 hours the disposable pad need to be replaced; movement, sweating or rapid temperature changes can affect the accuracy; skin irritation is the main weakness; the sweating can cause device shutdowns automatic, does not reliably detect hypoglycaemia but the detection is better at high glucose levels. **Reference:** (David Mendosa, 2007)
	**Device:** Pendra© **Company:** Biovotion AG (Solianis Monitoring AG; Pendragon Medical Ltd. Technology) **Technology:** Bioimpedance spectroscopy **Target area:** Wrist skin **Status:** CE Mark approved. **Advantages:** Have an alarm to inform the changes and hypoglycaemia; data download; data analysis software and data storage; self-correction for changes in impedance due to variations in temperature and long-lasting battery. **Disadvantages:** The device needs to be placed on the same skin area where it was calibrated; differences skin and underlying tissues require additional calibration and difficulty in calibrating; patient must rest for 1 hour for equilibrium before used for taking the measurement; every 24 hours the tape needs to be replaced; only have 35% correlation with invasive glucose meter. **Reference:** (Flacke, 2004)

Devices	Descriptions
	**Device:** NBM-200G **Company:** OrSense Ltd **Technology:** Occlusion near-infrared spectroscopy **Target area:** Fingertip skin **Status:** CE Mark approved. **Advantages:** Allows non-invasive haemoglobin and oxygen saturation; measuring time less than a minute, have an alarm, trend

	data analysis, data storage and integrated wireless telemetry; does not require frequent calibrations and easy in calibrating; 24 hours continuous glucose measurement. **Reference:** (Medgadget, 2007)
	**Device:** HG1-c **Company:** C8 MediSensors **Technology:** Raman spectroscopy **Target area:** Skin **Status:** CE Mark approved in 2012. **Advantages:** The measurement done every 5 minutes; transmit data continuously to a smartphone; can view 3 hours of instantaneous readings and the previous 4 months' reading for a retrospective view; alerts for hypo and hyperglycaemia; no requirement for constant recalibrating; compact, wearable and light-weight and water-resistant. **Reference:** (C8 MediSensors, 2012)
	**Device:** GlucoTrack™ **Company:** Integrity Applications Ltd. **Technology:** Thermal, ultrasound, electromagnetic **Target area:** Ear lobe **Status:** CE Mark approved in 2013. **Advantages:** High accuracy as it employs multiple technologies; user friendly; easy in calibrating; need calibration every 1 month; alerts for informing hypoglycaemia and hyperglycaemia, multi-user support, has data analysis software and data storage. **Disadvantages:** Requires individual calibration based on normal blood glucose meter and post-prandial blood glucose references. **Reference:** (GlucoTrack, 2016)

Devices	Descriptions
	**Device:** Combo Glucometer **Company:** Cnoga Medical, Israel **Year:** 2010 **Technology:** NIR spectroscopy **Target area:** Finger blood capillary **Status:** China Food and Drug Administration (CFDA) approved for China market in 2016. **Advantages:** Simple and easy to use. Connectivity by using USB, interface with PC, smart-phone and web. **Disadvantages:** Have minimal glucose reading levels. Requires a period of 1-2 weeks personalizes the device to get an accurate reading

	in the future. **Reference:** (Cnoga, 2015)

2.4 Technologies of non-invasive blood glucose monitoring

There are several different approaches to the non-invasive blood glucose monitoring technologies as shown in Figure 2.13. Non-invasive monitoring technologies can be characterized by two main approaches as follows; optical and transdermal (So et al., 2012) (Oliver et al., 2009). The glucose measurement can be done on the fluids produce by the body that is other than blood such as tears, sweat, saliva or urine. However, continuous glucose monitoring by using non-invasive approach can only be achieved by using direct measurement to the body tissues such as skin, tympanic membrane, cornea, tongue or oral mucosa (Poddar et al., 2008). By using a suitable transducer of blood glucose sensor, it is able to detect a weak signal indirectly from the blood through intervening tissues, such as skin, bone and fat.

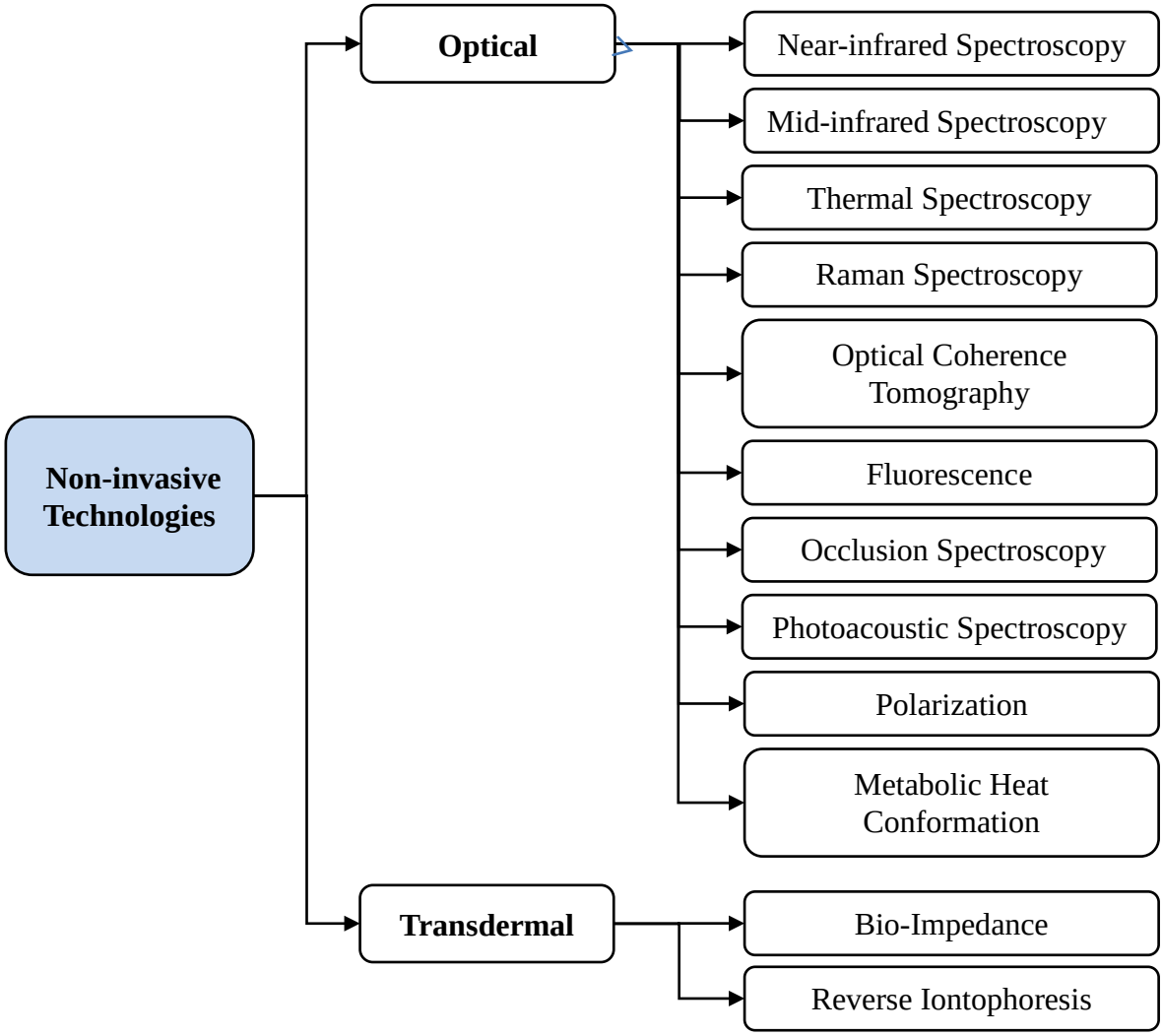


Figure 2.13: Overview on non-invasive glucose monitoring techniques

The optical spectroscopy technology is used by determining the properties of light which is either absorbed, transmitted or emitted as a function to measure blood glucose concentration (Guenter Gauglitz and Vo-Dinh, 2006). The detector will detect a small variation of the light produced after the light passes through the target area. Figure 2.14 shows the configurations used for optical spectroscopy light measurement.

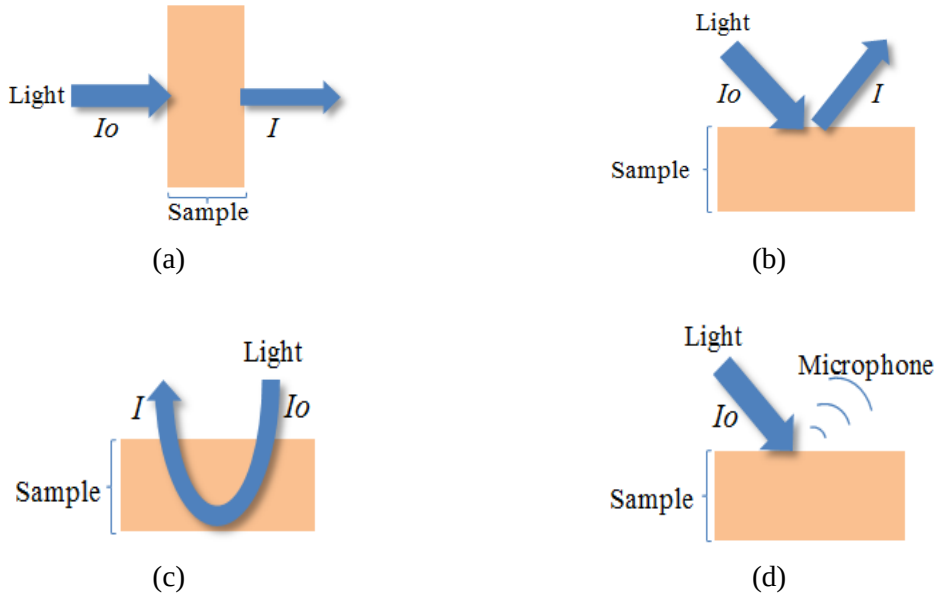


Figure 2.14: The diagram of different optical spectroscopy measurement configurations; (a) Transmission (b) Diffuse reflectance (c) Transflectance (d) Photoacoustic (Cunningham and Stenke, 2010)

The absorption of light can be described by the Beer-Lambert law. This law relates the attenuation of light to the properties of the material through which the light is travelling (Guenter Gauglitz and Vo-Dinh, 2006). The general Beer-Lambert law is usually written as shows in (2.1).

$$A = a_{\lambda} lc \tag{2.1}$$

Where A is absorbance, a_{λ} is a wavelength-dependent absorptivity coefficient, l is the path length of the sample with unit in cm and c is the concentration of the compound in solution with unit mol L^{-1}. While the equation in (2.2) is used when working in concentration units of molarity.

$$A = \varepsilon_\lambda lc \tag{2.2}$$

Where ε_λ is the molar absorptivity with units in mol^{-1} cm^{-1}. Experimental measurements are usually made in terms of transmittance (T), which is defined as in (2.3).

$$T = \frac{I}{I_o} \tag{2.3}$$

Where T is transmittance, *I* is the light intensity after it passes through the sample, and *Io* is the initial light intensity. Figure 2.14 illustrated the light intensity *I* and *Io*. The relation between *A* and *T* as in (2.4) and described in graph as shown in Figure 2.15.

$$A = \varepsilon_\lambda lc = \log \frac{1}{T} \tag{2.4}$$

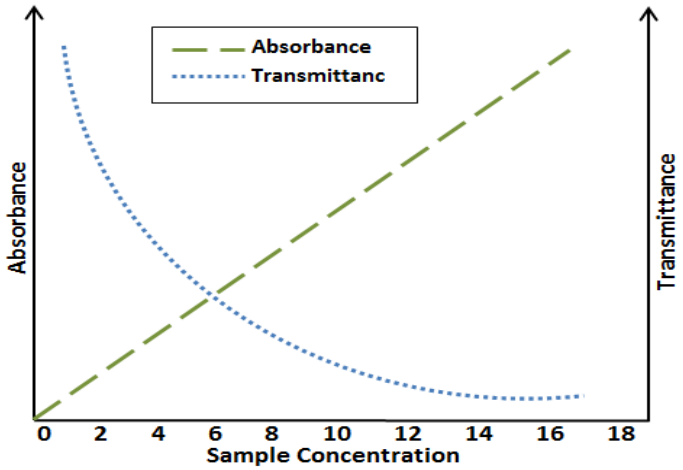


Figure 2.15: The graph of the relation between Absorbance (A) and Transmittance (T) toward sample concentration

This optical spectroscopy technology for non-invasive blood glucose monitoring consist of few important parts such as light source, detector for detecting the light intensity after it passes through the sample and signal processing signal processing to process the measurements that can be read easily by users. The basic block diagram is shown in Figure 2.16.

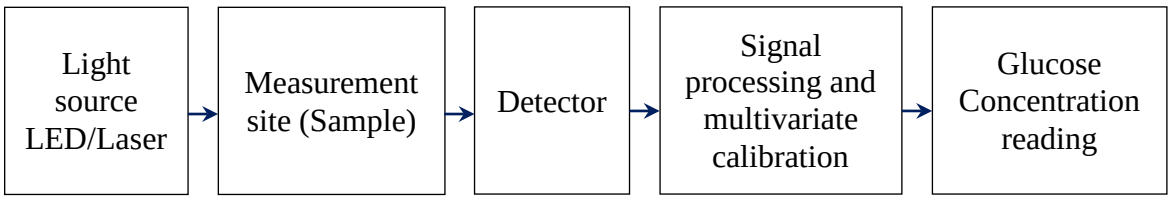


Figure 2.16: Basic system of optical non-invasive blood glucose monitoring

The transdermal technology uses a pair of the electrode that located on the skin. A small current pass through the skin and the detection are using the glucose oxidase catalytic system. Compared to transdermal technology, optical technology offers more investigative options.

2.4.1 Near-infrared spectroscopy

Near-infrared spectroscopy (NIR) is based on the collection of tissue absorption or emission spectrum by diffuse reflectance or transmittance with a spectrometer. This technique utilises a beam of light with the wavelength in the range of 750–2500 nm to estimate the blood glucose concentration in the tissues (1-100 mm deep) by measuring variations in the light intensity caused by transmission and reflectance in the tissue (Cunningham and Stenke, 2010) (Heinz W. Siesle et al., 2008). Table 2.3 shows different characteristics of NIR for different region of wavelength. The measurement sites

used for this technique are the area between the fingers (finger web) and finger cuticle, ear lobe, skin of the forearm, lip mucosa, oral mucosa, tongue, nasal septum, and arm skin.

Table 2.3: Characteristics in different wavelength region (Frederick Chee and Tyrone Fernando, 2007) (Yadav et al., 2015)

Wavelength (nm)	Characteristic
700-1300	- Higher orders of glucose overtone regions - Little glucose absorption - Low light absorption of water
1500-2500	- Highest glucose absorption - Does not get affected by excessive water attenuation - Relative minimum in water absorption spectrum - Highest absorption peaks for water, fat, and protein

This NIR technique has been used due to its cheaper cost, sensitivity, complexity, probability and selectivity. The glucose measurement may be disturbed by some physical and chemical parameters, such as variation in blood pressure, body temperature, skin hydration and albumin concentrations (Khalil, 1999). Errors can also occur due to environmental variations, such as changes in temperature, humidity, carbon dioxide and atmospheric pressure.

2.4.2 Mid-infrared spectroscopy

Mid-infrared spectroscopy (MIR) used a similar physical principle as NIR with a wavelength in the range of 2500 to 10000 nm. The advantage of MIR over NIR is that glucose-produced MIR bands, as well as other compounds, are sharper than NIR, which are often broad and weak. However, compared to NIR, MIR exhibits decreased scattering

and increased absorption due to the higher wavelengths. Therefore, light of MIR able to penetrate human tissue reach a few micrometers (μm) (Cunningham and Stenke, 2010). This limitation of the light penetration into the human tissue causing difficulty in getting the reading, especially used in the transmittance sensor arrangement (Ferrante do Amaral and Wolf, 2008). Besides that, the signal result has a present of the noise that affects from other molecules such as water and other non-glucose metabolites which modulate the magnitude of the absorption peak of glucose light, similar problems and confounding factors as of NIR (Oliver et al., 2009). This MIR technique is less studied than NIR technique for glucose measurements, probably due to the strong limitation in penetration.

2.4.3 Thermal emission spectroscopy

Thermal emission spectroscopy is based on the principle that the cutaneous microcirculation is dependent on the local glucose concentration. The infrared (IR) is used as a light source. The changes in the IR signal are generated in the human tissues as a result of glucose concentration changes (Ferrante do Amaral and Wolf, 2008). The light of IR radiation can be emitted by the human tissues and only the specific wavelength can be permissibly by using a special filter for glucose measurement to pass to a detector. This technique uses a similar concept as standard clinical tympanic membrane thermometers, with the addition of specific wavelengths for glucose human tissues. Therefore, the most significant sources of noise in this technology can be affected by variation of ambient temperature and sensitive to the motion (Oliver et al., 2009).

2.4.4 Raman spectroscopy

Raman spectroscopy based on scattering of single wavelength light. This technique applies the laser light of one wavelength, where the quantification and identification of blood glucose is judged by changes in the frequency of the light that results from inelastic scattering of the oscillation and rotation motions in the glucose molecule (Cunningham and Stenke, 2010). The advantage of Raman spectroscopy is that it has a high molecular specificity. It can reduce the interference of water compared to MIR or NIR spectroscopy due to weak diffusion index of water. However, this technique requires a powerful detector and complicated instrumentation because of the signal produced in this technique is extremely weak causing the noise to be very visible after an interaction with different tissue component (Oliver et al., 2009) (Norman Colthup, 2012).

2.4.5 Optical coherence tomography

The optical coherence tomography (OCT) is an optical signal acquisition uses a low coherence light source, an in-depth scanning system, a sampling device shown as in Figure 2.17. The skin is irradiated with coherent light (light in which the emitted photons are synchronized in time and space) and backscattered radiation is detected by the photodiode. The glucose concentrations in the dermis can be determined by using this technique as done by (Larin et al., 2002). The measurement sites used is a retina or skin, typically in the forearm (more specifically: in the ISF of the upper dermis of the skin). This technique provides advantages in signal-to-noise ratio (SNR), high resolution and depth penetration, because the inter-ferometric signal can be formed only within the coherence length of the source. This technique is not affected by urea, blood pressure, heart rate and haematocrit. However, the OCT technique can be sensitive to motion artifacts and

measured signal can be influenced by changes of several degrees of skin temperature (So et al., 2012).

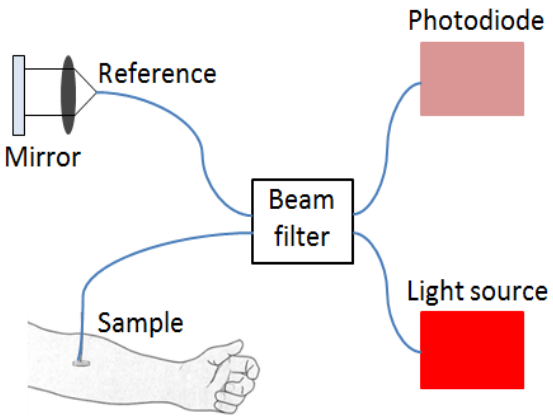


Figure 2.17: Schematic of OCT experiment set up by using arm

2.4.6 Fluorescence

Fluorescence technique is based on the changes in fluorescent light that is emitted from molecules in different states. Many fluorescence-based glucose sensors are based upon changes in fluorescence resonance energy transfer between a fluorescent donor and a receptor. Concanavalin A (ConA), is frequently used as the receptor molecule as shown in Figure 2.18, since it has four glucose binding sites. Commonly used competitive binders are dextran, α -methyl mannoside (Oliver et al., 2009). The advantage of this technique for bio-sensing is fast, extremely sensitive, reagent-less cause little or no damage to the host system (Pickup et al., 2005).

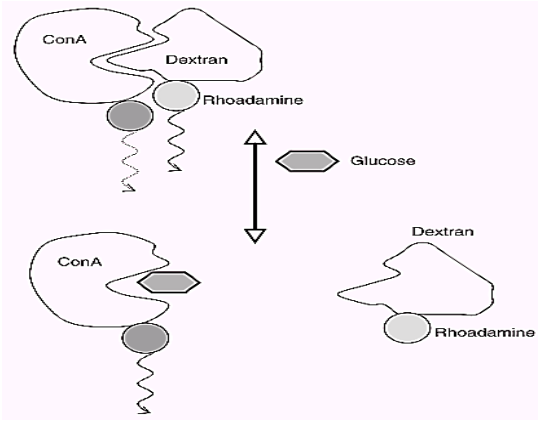


Figure 2.18: Fluorescence resonance energy transfer (FRET) fluorescence with ConA (Oliver et al., 2009)

Researchers, Ramachandram Badugu et al. (2003) have worked in development of fluorescence-based glucose sensing contact lenses. The contact lens based on polymer film has been developed for the glucose concentration detection in tears. This contact lens based sensor has been receiving a tremendous attention because it is disposable and portable. The colour of contact lens is changing according to the glucose concentration level. In addition, this hydro-gel soft lens is safe for daily wear. However, this technique is extremely sensitive. In tissues, the use of ultraviolet light could lead to strong scattering and fluorescence phenomena (Zhang et al., 2011).

2.4.7 Occlusion spectroscopy

The occlusion spectroscopy uses scattering technique and exploits pulsatile arterial flow. This technique uses a light source such as a multi-LED or laser diode matrix and light detector to detect the transmitted light from the sample. Finger's root is the best site for glucose detection in this technique (Amir et al., 2007). The light source used in this technique such as LED's is preferable in terms of cost and availability. The advantage of this technique is that, it measures arterial glucose but it is vulnerable to many intravascular

variables such as drug treatment, intrinsic erythrocyte aggregation, free fatty acid concentration and chylomicrons (Oliver et al., 2009). However, this LED-based technique required complex interpretation of spectral measurement result.

2.4.8 Polarimetry

Polarimetry is based on the rotation of polarized light by a transparent optically active substance. Polarized light is referred to linearly polarize with respect to the direction of propagation, and all waves vibrating in a single plane are termed plane parallel or plane-polarized. Light rotation is depending on several factor including pH, temperature, wavelength of light source, sample thickness and concentrations of the sample used (Oliver et al., 2009). In the field of pharmaceutical and nutritional industries, this technology has been used a long time ago to find out the level of the compounds in every mixture. The skin cannot be used as a research tissue, because it shows a high scattering effect, particularly in the stratum corneum since this technique is sensitive to the scattering properties of the tissue used, as scattering depolarizes light. This technology can be used to observe glucose from the aqueous humour of the eye, which shows a minimal scattering and absorption effect and it interacts with the degree of rotation of the polarization vector that is proportionated to the concentration of glucose. However, eye movement and motion artefacts are general sources of errors in this technique. Additionally, this method also has a time lag for a few minutes before a change in the blood glucose concentration can be observed from the eye (Baba et al., 2002).

2.4.9 Photoacoustic spectroscopy

Photoacoustic spectroscopy is based on the ultrasonic waves caused by the absorption of the light source to measure the glucose concentration. The solution sample is

excited by a light source, with a wavelength that is absorbed by a particular molecular in the sample. The light absorption causes microscopic localized heating in the medium, which causes an ultrasound pressure wave or sound is generated and it is detectable by a microphone (Oliver et al., 2009). The illustration of the photoacoustic spectroscopy is shown in Figure 2.19. The possible body area that can be used for measurement is the eyes (especially the eyes sclera), fingers and forearm, with the presence of glucose in the blood vessels by skin and tissues or both. The advantage of this technique because it uses optical radiation levels several orders of magnitude below the threshold of pain or tissue damage (MacKenzie et al., 1999). The light source wavelength that can be used vary in a wide interval (from ultraviolet to NIR) and provide higher sensitivity than traditional spectroscopy in the determination of glucose. However, there are some problems remain to be clarified for this technique, such as the effect of body water content and dehydration. This technique can be affected by the noise that is created from non-glucose blood components which need to be excluded from the measurements. Other than that, other factors that may affect cell membranes should also be evaluated.

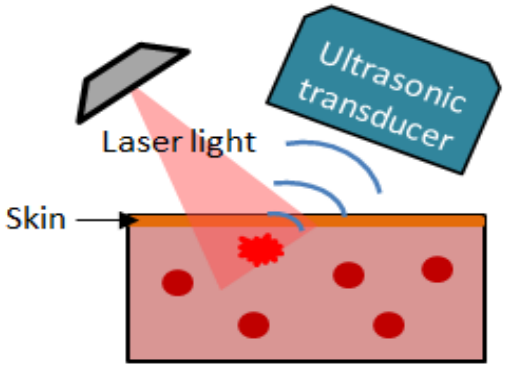


Figure 2.19: Basic setup of photoacoustic spectroscopy

2.4.10 Metabolic heat conformation

Metabolic heat conformation (MHC) is based on measurement of physiologic indices related to metabolic heat generation and local oxygen supply, which correspond to the glucose concentration in the local blood supply. The combinations of thermal and optical sensors are used to measure thermal generation, blood flow rate, haemoglobin (Hb) concentration, and oxyhaemoglobin concentration (Cho et al., 2004). The fingertips and skin is use as a measurement sites. This technique is feasible and low cost. However, this technique suffers from interference due to environmental parameters. The first MHC prototype as shown in Figure 2.20 has been produced by Hitachi company and it has a correlation coefficient (R^2) of 0.91 in laboratory conditions, but the company Hitachi intends to improve its performance in order to obtain sale approval (Ko et al., 2004).

Figure 2.20: Prototype using metabolic heat conformation blood sugar monitoring device from Hitachi

2.4.11 Bio-impedance spectroscopy

Bio-impedance spectroscopy technique is working by determining the dielectric properties of the skin tissue by using two electrodes. A small constant current is passed

between these electrodes and the change of the voltage is determined between these electrodes has been used for the determination of the glucose concentration level in blood (Oliver et al., 2009). The frequency range of 100 Hz to MHz is used to measure the dielectric spectrum. Variations in plasma glucose concentration drive to the decrease in Sodium concentration and increase in Potassium concentrations in red blood cells (Vashist, 2012). These variations cause changes in the red blood cell membrane potential, which can be estimated by determining the permittivity and conductivity of the cell membrane through the dielectric spectrum. The advantage of this technique is simple implementation and safe for the human used due to its non-invasive nature. This technique provides information about glucose in the vascular compartment. However, this technique has some limitations that could expose the user to potentially dangerous situations and there are errors that can happen due to variability, sweating and motion and the technique require calibration (Oliver et al., 2009).

2.4.12 Reverse iontophoresis

Reverse iontophoresis employs a low electric current between two electrodes to access the skin tissue. Electric potential is applied between the anode and cathode electrodes will cause charged and uncharged molecules to move across the dermis at a rate significantly greater than the passive permeability (Oliver et al., 2009). The uncharged molecules are carried along with the ions flow induced by electrolysis and being collected at the cathode where a glucose sensor is placed to get a direct glucose concentration measurement as shown in Figure 2.21. At physiological pH (~5.0-6.0), skin is negatively charged. Therefore, a positively charged ion will penetrate more easily across the skin than a comparably sized negative ion.

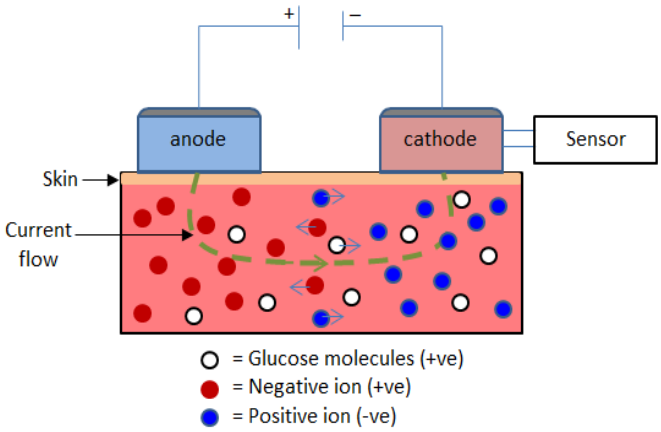


Figure 2.21: Schematic illustration of the principle of reverse iontophoresis showing an iontophoresis extraction device supplying a constant

Compared to other enzyme-based electrode sensor systems, this technique has some potential advantages. An oxygen supply is not a bound factor to glucose oxides that can cause the concentration of glucose increase. However, this technique tends to cause skin irritation and cannot be used if the subject is sweating significantly. This technique also requires a long time to be ready for calibration and it is a complicated process (So et al., 2012). The GlucoWatch® biographer as in Figure 2.22 uses this technique to record glucose reading through intact skin.

Figure 2.22: GlucoWatch® biographer using reverse iontophoresis technique

2.5 Fundamentals of near-infrared absorption spectroscopy

Near-infrared spectroscopy (NIR) is a spectroscopic method that uses the region of the electromagnetic spectrum from 750 to 2500 nm. The absorption of electromagnetic radiation in the NIR region is caused by overstone and combination vibrations. Polytomic exhibit many overtone combination vibrations, their spectral bands overlap and make typical NIR bands look very broad and featureless. However, NIR spectra contain molecular information of the sample, and this information can be extracted by using the chemometric method (use of mathematical and statistical methods to improve the understanding of chemical information and to correlate quality parameters or physical properties to analytical instrument data). A prerequisite for chemometric evaluations is the high quality of the collected spectral data. Therefore, resolution, wavelength precision, photometric precision and signal SNR is the important criteria for the selection of a NIR spectrometer (Guenter Gauglitz and Vo-Dinh, 2006).

NIR offers the greatest diversity of instrumentation principles, and the market for commercially available instruments is undergoing continuous change and grow, compared to others optical spectroscopic methods. NIR has a variety of applications, such as in agriculture, medical and pharmaceutical applications, food processing, polymer and plastics processing, environmental analysis, material recycling, and satellites or aircraft for remote sensing (Kolb et al., 2016). Commercial NIR spectrometers vary remarkably with depending on applications, cost, size and portability, and measurement time.

2.5.1 Detector

The detector of the NIR is also known as photodiode. There is no single detector that can cover the complete NIR range from 700 nm to 2500 nm. Therefore, there are

different types of detectors that have different range of detection. A list of detector types and their coverage ranges is given as in Table 2.4 (Guenter Gauglitz and Vo-Dinh, 2006).

Table 2.4: Types of near-infrared light detector

Types of detector	Wavelength ranges (nm)
Germanium (Ge)	600 to 1800
Silicon (Si)	400 to 1100
Indium Gallium Arsenide (InGaAs)	900 to 1700
Extended InGaAs	1100 to 2800
Lead Sulfide (PbS)	1100 to 3300
Indium Arsenide (InAs)	1500 to 3500
Indium Antimonide (InSb)	2000 to 4000
Lead Selenide (PbSe)	1100 to 4800

The NIR detector is commonly used trans-impedance amplifier in the circuit to amplify the light-dependent current. In its most simple form, a trans-impedance amplifier consists of an operational amplifier and a feedback resistor as shown in Figure 2.23. The current to be amplified is applied to the inverting input, causing the output voltage to change according to the equation in (2.5). The proper design for a single supply photodiode amplifier requires the consideration of many factors, such as stability and limitation of input voltage and output voltage. Furthermore, the effects of direct current (DC) error sources such as input bias current and input offset voltage are often ignored and this can disturb the response of the circuit (Caldwell, 2014).

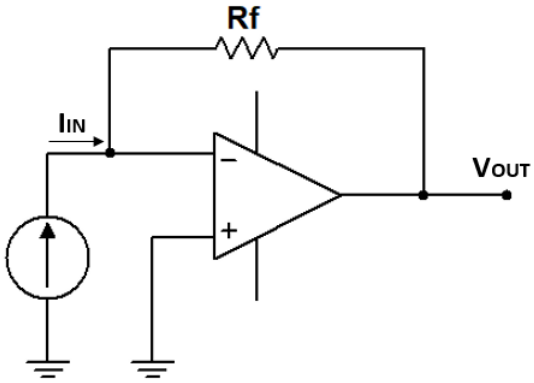


Figure 2.23: A basic op amp trans-impedance amplifier.

$$V_{OUT} = -I_{IN} R_F \tag{2.5}$$

2.5.1.1 Modes of detector operation

A photodiode can be operated in one of two modes: photoconductive (reverse bias) or photovoltaic (zero-bias). Mode selection depends upon the application's speed requirements and the amount of tolerable dark current (leakage current).

The basic circuit of photoconductive mode is shown as in Figure 2.24. An external reverse bias is applied in photoconductive mode. The measured current through the circuit is based on the illumination accepted by the photodiode; the measured output current is linearly proportional to the input optical power. This operating mode reduces the response time because the reverse bias increases the width of the depletion junction producing an increased responsively with a decrease in junction capacitance and produces a very linear response. Operating under these conditions also increase the dark current without much change in the photocurrent, the photocurrent is linearly proportional to the illumination, but this can be limited based upon the photodiode material. The output voltage change as in (2.6) (Osi Optoelectronics, 2009).

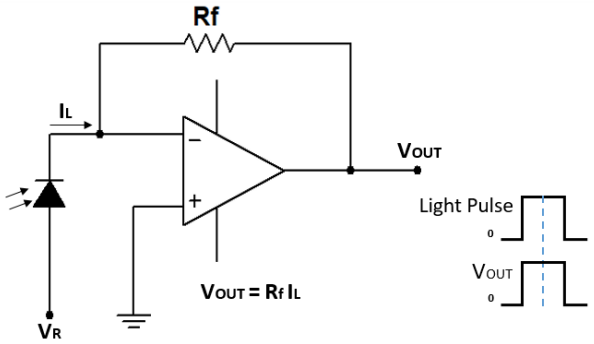


Figure 2.24: Connection of a photodiode to the trans-impedance amplifier in mode of photoconductive

$$V_{OUT} = I_L R_F \tag{2.6}$$

Figure 2.25 shows the basic circuit of the photovoltaic mode. In the photovoltaic mode the photodiode is zero bias. The photocurrent flow out of the sensor is limited than a voltage builds up. In this operating mode, the dark current is kept at a minimum. The voltage output of the photodiode follow in (2.7) (Osi Optoelectronics, 2009).

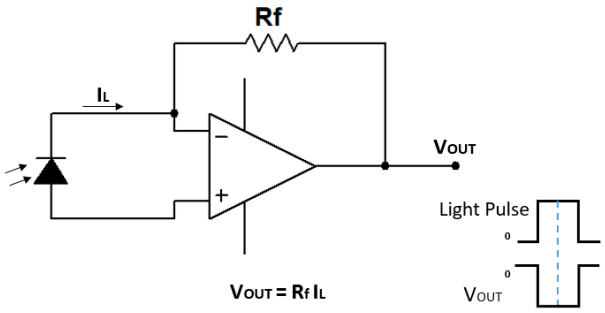


Figure 2.25: Connection of a photodiode to the trans-impedance amplifier in mode of photovoltaic

$$V_{OUT} = I_L R_F \tag{2.7}$$

2.5.1.2 Dark current

Dark current is leakage current that flow through the photodiode in the absence of light when bias voltage is applied to a photodiode. The dark current includes photocurrent generated by background radiation and the saturation current of the semiconductor junction. When operating in a photoconductive mode, there is a tendency for increasing of dark currents that change directly with temperature. The increasing of the dark current approximately doubles for every 10°C increase in temperature. The junction capacitance will decrease by applying a higher bias, but the value of dark current presents will increase.

Besides, the material and the active area size of the photodiode are also affected the dark current. Silicon devices generally produce low dark current compared to germanium devices which have high dark currents.

2.5.2 Main limitation and challenges

There are many groups of researcher have found that main reasons which limit the use of NIR spectroscopy for non-invasive blood glucose detection are because of the absorbed by other biological chromophores, interferences due to stratum corneum resulting in poor SNR, selection of proper wavelength, physiological factor, calibration issues, environmental factors, and thermal noise.

In order to reduce the interferences due to the stratum corneum, optimal wavelength selection is required. The effective optimal wavelength for diffuse reflectance measurements is considered from 1.3 to 2 mm (Delpy et al., 1988).

The selection of the measurement site also one of the important aspects that need to be considered for non-invasive blood glucose monitoring because it can affect the improvement of the SNR. The higher of SNR can help to improve the accuracy and sensitivity. In addition, this technique needs different calibration for a different user to

provide better results. The optical characteristic of tissues shows significant change for different individuals or different measurement sites. Since the skin thickness, colour, and intracellular fluid in tissue are different for every individual and measurement sites. This would be difficult to implement stable universal calibration models for all subjects.

Moreover, physiological factors which, depending on the time, such as blood pressure, blood flow, temperature and dynamic nature skin also can affect the glucose measurement and this can be a problem in long-term blood glucose measuring. Therefore, calibration is needed from time to time. The factors of environmental change such as temperature, atmospheric pressure, and humidity also can cause errors. But, if the measurement is taken under well-controlled physiological and environmental conditions then the repeatability of glucose spectra can be improved (Valery V. Tuchin, 2008).

Tenhunen et al. (1998) addressed the issue of thermal noise and baseline drift in NIR non-invasive blood glucose detection. The thermal noise is due to the photodetector and it is reduced by averaging the result. Repositioning of sensor probe for measurement can also cause baseline drift in the spectra as an error. The sensor prop should be designed so that such type of error can be avoided (Tenhunen et al., 1998).

2.6 Experimental and analysis methods

There are two types of experimental methods that are used in scientific studies to aid in comparing two competing explanations. The experimental methods are in-vitro and in-vivo. The differentiation of these two methods is based on the type of sample that is being used in the experiment.

2.6.1 In-vitro sensing experiment of glucose

The primary analysis of blood glucose based on the NIR is carried out through in-vitro experiments first before in-vivo measurements. In the in-vitro experiments, the experiment samples are prepared in the laboratory. The chemical components or the elements of human blood such as glucose, protein, lactate, lipids and amino acids can be used. These components can be mixed together to measure the effect of other composites in the detection of blood glucose (Arimoto et al., 2003) (Parab et al., 2010). The in-vitro experimental conditions is less complicated as compared to in-vivo measurements and also shows positive results. The benefit of in-vitro experiments is that the effect of critical physical and chemical parameters of the blood glucose samples is created can be monitored in a well-controlled environment.

2.6.2 In-vivo sensing experiment of glucose

In-vivo experiments may be performed on human subjects (diabetic patient or non-diabetic). Fasting glucose test (FGT) is carried out for diabetic patient subjects and oral glucose tolerance test (OGTT) is carried out for non-diabetic subjects (American Diabetes Association, 2014). The in-vivo monitoring can be affected by some factor such as temperature, pressure, site of measurement area, skin hydration and physiological parameters (Troy and Thennadil, 2001) (Liu et al., 2005). Human skin has multiple layers, i.e. epidermis, dermis and subcutaneous (hypodermis) layers such as shown in Figure 2.26. The outermost layer, which is the epidermis and it does not contain useful information. The middle layer dermis is assumed to be correlated with the blood glucose, as it carries most of the blood vessels and the subcutaneous is the innermost layer which has only been covered by fatty tissues. However, a large depth of the dermis layer can cause interference on the penetration of photons (Troy and Thennadil, 2001).

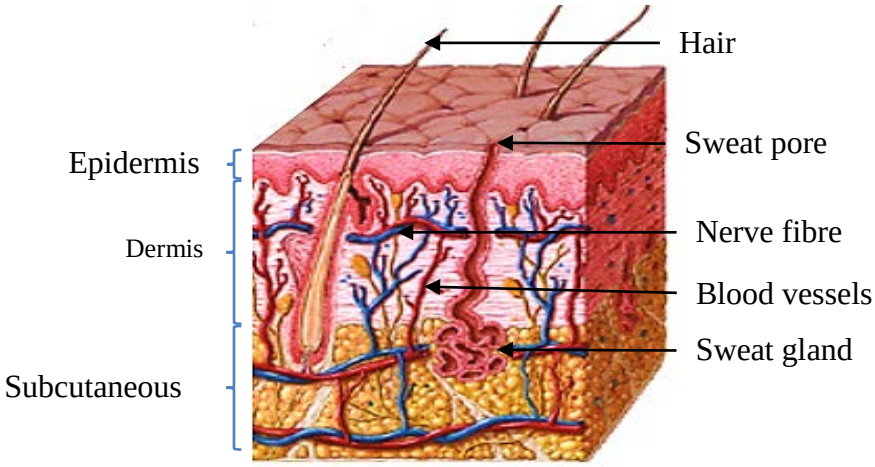


Figure 2.26: Skin structure

2.6.3 Analysis method

Selection of appropriate data analysis methods, specific compatible hardware and software is an important requirement to make sure all the information are interpreted well. The mathematical models are required to estimate the relationship between the output signal of NIR and concentration level of glucose samples used. Numerous multivariate analysis techniques have been used in literature such as partial least squares regressions, principal component regression, multiple linear regressions and artificial neural network to extract glucose related information from the overall spectra.

A calibration model is required to determine glucose concentrations corresponding between invasive and non-invasive methods. The results of non-invasive measurements are determined in the form of root-mean square error of prediction (RMSEP), correlation coefficient prediction (R^2) and standard error of predictions (SEP). A lower value of SEP and the higher value of R^2 indicates a better quality of results. Moreover to measure the clinical accuracy, Clarke error grid analysis (EGA) is used which was developed in the year 1989 by Clarke (Clarke et al., 1987). This Clarke EGA dividing the graph area into five regions and has a reference line as shown Figure 2.27. Points *A* are within 20% of the

reference method, showing clinical accurate area. Point in area *B* would not lead to inappropriate treatment. Points in area *C* would lead to unnecessary treatment, whereas the points in area *D* are potentially dangerous, failed to detect hypoglycaemia or hyperglycaemia events correctly. Points which are in area *E* would create confusion and hypoglycaemia will be treated as hyperglycaemia and vice versa.

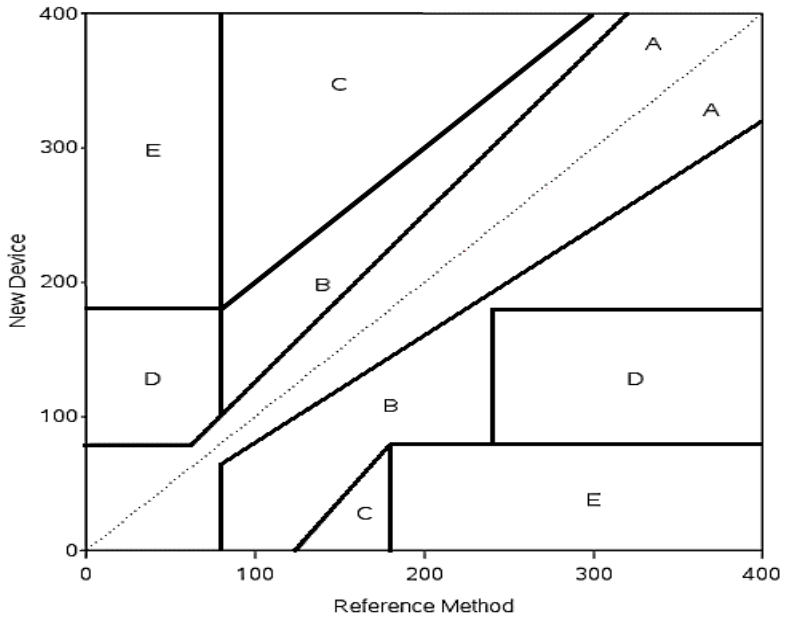


Figure 2.27: Clarke Error Grid Analysis

2.7 Relevant reviews of existing studies on NIR spectroscopy technique for non-invasive blood glucose measurement

The non-invasive blood glucose monitoring has attracted growing attention by many researchers in recent years due to their fast response, cost-effective, easy to use at home by potential users and environment-friendly. Non-invasive blood glucose monitoring also has high attention and demand by diabetics, as this technique is not painful compared to the invasive method that used a glucose meter. However, there are few issues on the proper accuracy and precision as invasive conventional techniques. There are many

features that can be considered and there are a few study to improve the development of non-invasive. The development of the non-invasive method is based mainly on the optical and dielectric properties.

There are two types of sensor configurations commonly used in the study that has been carried out using NIR techniques, namely transmittance and reflectance. Referring to the previous research works, there are many investigations and experimentation that have been conducted using these two configurations and there are various types of sensor layouts that have been used. There are research finding at (Yadav et al., 2014) show the simple sensor arrangement that used a single LED with a specific wavelength and light detector as shown in Figure 2.28.

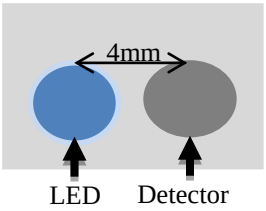


Figure 2.28: Basic reflectance sensor arrangement (Yadav et al., 2014)

In other findings, there are also sensor arrangement in ring-shaped (circular) for reflectance configuration done by research groups of Mohd Aziz (2015) and Guevara (2010). That differentiates the sensor layout in these three researches is the number of LEDs and the LEDs wavelength used as shown in Figure 2.29.

Figure 2.29: Ring-shape reflectance sensor arrangement

Transmittance configuration is the choice of most researchers and there are various sensor arrangement that had been used. The basic and simple arrangement used a single LED with a specific wavelength and light detector as shown in Figure 2.30 (Unnikrishna Menon et al., 2013). In addition, there are also researchers who uses more than one LED and detector as found in the work that has been carried out by (Zeng et al., 2013) and Figure 2.31 shown the sensor arrangement used.

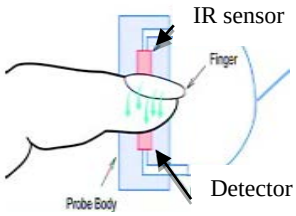


Figure 2.30: Basic transmittance sensor arrangement (Unnikrishna Menon et al., 2013)

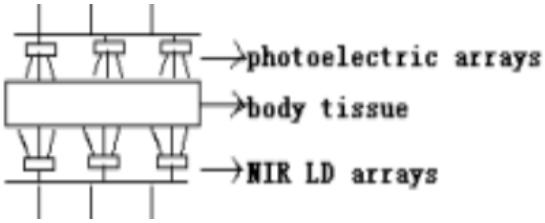
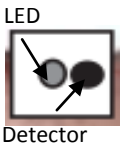
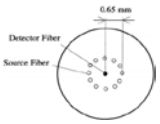
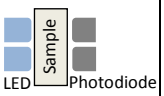
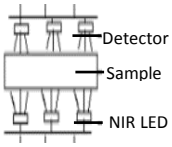
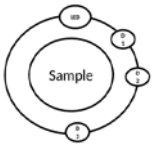


Figure 2.31: Multi sensor transmittance sensor arrangement (Zeng et al., 2013)

Table 2.5 shows the design comparison based on several previous studies that using the NIR technique in developing non-invasive blood glucose monitoring. The review shows that there is various wavelength range of NIR (780 nm to 2500 nm) that has been used. The in-vitro analysis use a sample of glucose solution and in-vivo analysis uses a human sample or blood sample.

Table 2.5: The design review on the previous researcher

Reference	Configuration	Wavelength	Detector	Sensor Structure	Analysis
(Yadav et al., 2014)	Diffuse reflectance	950 nm	Monolithic photodiode (OPT101)		In-vivo (Forearm)
(Narkhede et al., 2016)	Diffuse reflectance	950nm	Photo-transistor (PT333C)		In-vivo (Fingertip)
(Maruo et al., 2003)	Diffuse reflectance	1430-1850 nm	256 InGaAs photodiode		In-vitro In-vivo (skin)
(Yatim et al., 2014)	Diffuse reflectance	900-2500 nm	Spectrometer	-	In-vitro
(Buda and M. Mohd. Addi, 2014)	Transmittance	1550 nm	InGaAs photodiode (FGA10)		In-vitro In-vivo (finger)
(Chua et al., 2014)	Transmittance	1450 nm Red LED	Photodetector	-	In-vivo (Finger phantom)
(Hotmartua et al., 2015)	Transmittance	1300-1550 nm	InGaAs photodiode		In-vivo (earlobe)
(Zeng et al., 2013)	Transmittance	1400-1800 nm	Photodiode		In-vitro In-vivo (finger, earlobe)
(Guo et al., 2015)	Transmittance	820-1050 nm	Photoelectric sensor	-	In-vivo (fingertip)
(Yadav et al., 2015)	Transmission	950 nm	Photodiode		In-vitro In-vivo (arm, finger, earlobe)

A lot of development studies have been done in getting to improve the development of non-invasive blood glucose based on NIR technique. Therefore, there are previous research works have been found using a combination of various types of sensors or techniques that aimed to reduce the external effects and to increase the accuracy of blood glucose readings as shown in Table 2.6.

Table 2.6: The previous studies that used combination other technique in the development

Reference	Descriptions	Analysis
(Kamboh and Khan, 2013)	**Sensor configuration:** Transmittance **Measurement:** Blood glucose, haemoglobin, Oxy-haemoglobin and Tissue thickness **Sensor:** 2 NIR (1500 nm), IR, red and green LED, and photodiode (InGaAs) **Processor:** Programmable system-on-chip (PSoC-5LP) **Display:** LCD, android Application via Bluetooth. **Site measurement:** Earlobe	Clarke EGA = Zone A 75% Zone B 25 % Rp = 0.85
(Guevara and González, 2010)	**Sensor configuration:** Diffuse reflectance **Measurement:** Blood glucose **Sensors**: 6 multi-wavelength of NIR and photodetector and Electrical impedance **Site measurement:** Fingertip, wrist, flexi carpi.	Clarke EGA = Zone A 77.86% Zone B 22.14%
(Song et al., 2015)	**Sensor configuration:** Diffuse reflectance **Measurement:** Blood glucose **Sensor:** Multi-wavelength of NIR (850 nm, 950 nm, 1300 nm) and Electrical impedance **Site measurement:** Hand	Clarke EGA = Zone A 90% Zone B 10%

2.8 Summary

This chapter has presented the basic knowledge about diabetes, history of the blood glucose monitoring development, technologies of non-invasive blood glucose monitoring, fundamental of NIR absorption spectroscopy, experimental and analysis methods and

relevant reviews of existing studies on NIR spectroscopy technique for non-invasive blood glucose measurement. The basic knowledge of the non-invasive techniques need to be known before any technique is chosen to be used since there are various techniques that can be used. The NIR technique has been chosen to be used in this non-invasive development. Since this technique require low in cost, sensitivity, complexity, probability and, selectivity In order to understand the theory of near-infrared spectroscopy development, the basic knowledge of NIR, the detector and the limitation and challenges of NIR has to be understood before any design can be made. An addition, experimental types and appropriate analysis also need to be known to facilitate the data collection process. The experimental and analysis methodology are described in Chapter 3. As presented in the table of review in this chapter, there is the variety of NIR design that is being used before on purpose to see the output analysis and to improve the non-invasive development. NIR non-invasive blood glucose is very much demanded by the patients with diabetes because they do not require blood samples from the users and has no long-term impact on the users. This technique also cheap compared to other non-invasive technique.

CHAPTER 3

RESEARCH METHODOLOGY

3.1 Introduction

This thesis starts with study and research on blood glucose measurement which focused on the technique of non-invasive blood glucose measurement. The study is more dedicated on the design that used near-infrared (NIR) optical technique for blood glucose measurement as described in Chapter 2.

This chapter focuses on the methodology of the project. The proposed method, steps of every design in this project and its working principle to monitor the blood glucose level is explained. The design involves two parts; circuit sensor and sensor casing for the sensor holder. The basic principle of every circuit used in this project and any calculation that involved is presented in this chapter. Additionally, the involved components in the circuit design are also described. The dimension of the 3D design sensor casing is shown in this chapter. There are two types of sensor casing design and this casing is used during the experiment to hold the sensor. This project is divided into two analysis; in-vitro and in-vivo. The in-vitro analysis is using glucose concentration as a sample and the in-vivo analysis is used human as a sample. The procedures of these two analysis and method of sample preparation are explained in detail in this chapter. In addition, an algorithm to read the glucose level was developed. The data of this two analysis results are tabulated and discuss in details in Chapter 4.

3.2 Block diagram of the project

The development of NIR non-invasive blood glucose monitoring system is described in the block diagram as shown in Figure 3.1. This development begins with the first step of designing the circuits and sensor casing. There are several steps in circuit design and this circuit is designed based on its specification and function. The custom 3D casing was designed based on the requirement for experimental use as a sensor holder. Two types of experiments used to assist in data collection, which is in-vitro and in-vivo. An algorithm was developed to make the device functioning as blood glucose monitoring. The algorithm was developed based on data analysis. The conventional blood glucose measurement using glucose meter was used to verify the accuracy of the blood glucose measurement done using the device developed in this study. Lastly, a prototype device system has been developed with some additional sensors for stability during measurement and also as a reference. Besides, it is also equipped with the indicator for the alert about abnormal condition.

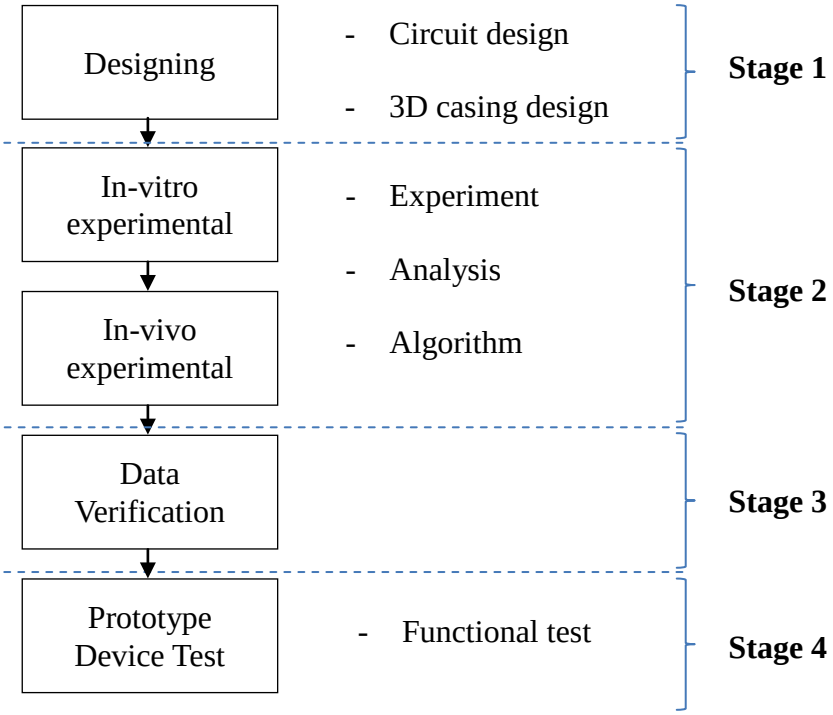


Figure 3.1: Illustrates the block diagram of the flow in the methodology part. The block diagram shows from starting of the first step this project proposed to finish

In general, there are four stages of conducting this study:

Stage 1: Components selection and designing

This stage is a design stage, there are two things that need to be designed at this stage: circuits and custom 3D casing. Indirectly, this stage also involves the study and selection of components to be used in the circuit that being designed. Besides, this stage also involves printed circuit board (PCB) fabrication process and 3D casing printing. The steps of designing are explained in this chapter.

Stage 2: Experimental and analysis

There are two types of experiment: in-vitro and in-vivo that are conducted at this stage. The in-vitro experiment is using samples of glucose solution that has been prepared

in the laboratory and the in-vivo experiment done by using a human as a subject. The experiment preparation also described in Chapter 3.

The program development is also done at this stage. This involves the development of algorithms that had been built into the microcontroller processor to ensure that the device built in this project can function as a blood glucose monitor. The analysis results from these two analysis are used to assist in the development of the mathematical model for the development of the algorithms for voltage measure to glucose concentration conversion. The flowchart of the development program is presented in Chapter 3.

Stage 3: Data verification

Data verification was done in this stage. The result from the analysis part in Chapter 3 is discussed in Chapter 4. As an invasive method using a glucose meter is the best way and accurate to measure the blood glucose, therefore the output reading from the device built is compared to the reading of the glucose meter to verify whether the output from the device built is valid and acceptable.

Stage 4: Prototype device testing

The last second stage is a functional test of the prototype system that has been developed. This prototype device used several combinations of the sensors. The purpose of the use of additional sensors is to control the other noise that can affect the measurement. This prototype was also developed with the indicator for user convenience. The function of the device is tested and the results presented as in Chapter 4.

3.3 Hardware design and development

The circuit and custom casing were designed in this section. The circuit design is divided into two parts; NIR LED and photodiode conditional circuit as the main circuit and thermistor circuit and vibration sensor circuit as the additional circuit. However, every circuit used has their own purpose to complete the full circuit system. The flowchart in Figure 3.2 shows the overall processes required in circuit design. The development of the circuit design starts with the comprehensive study for understanding the circuit design specification. The circuit was designed and simulated using Multisim software for PCB fabrication. Proteus 8 software is used to design the PCB layout for PCB fabrication. Detail circuit design and components used are explained in the subtopics of 3.3.1 until 3.3.4. All completed circuits are connected to the input port on the microcontroller used.

This casing was designed to make the experiment more manageable since this experiment uses a bunch of samples. There are two types of casing designed for two different experiments: in-vitro and in-vivo experiments. The SolidWorks software was used to design the casing while Cura software is used to prepare the model for 3D printing and the Ultimaker 3D printer was used to print the design. The material of PLA is used for 3D casing printing. Figure 3.3 shows the flowchart of 3D casing designing.

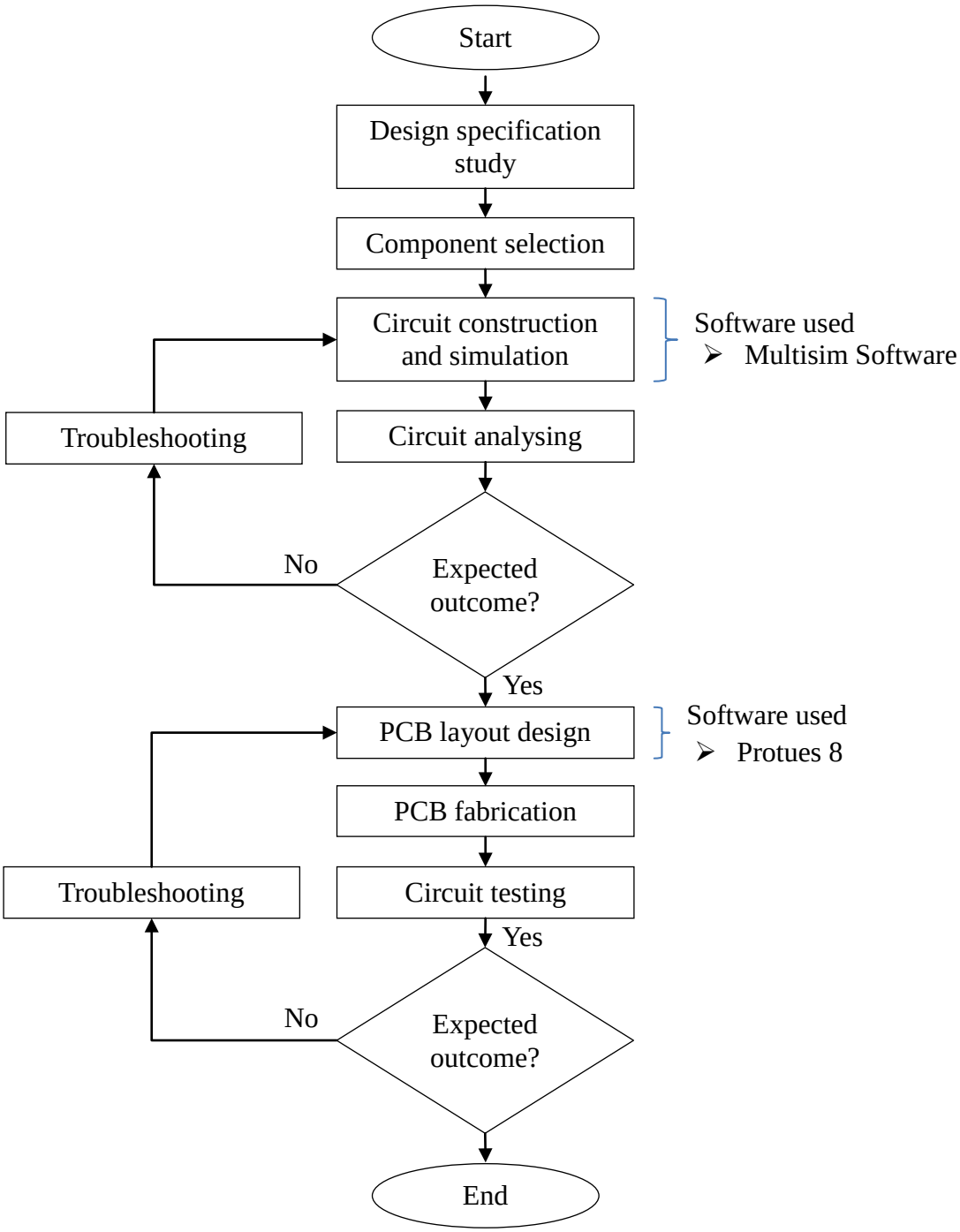


Figure 3.2: The flowchart of circuit designing

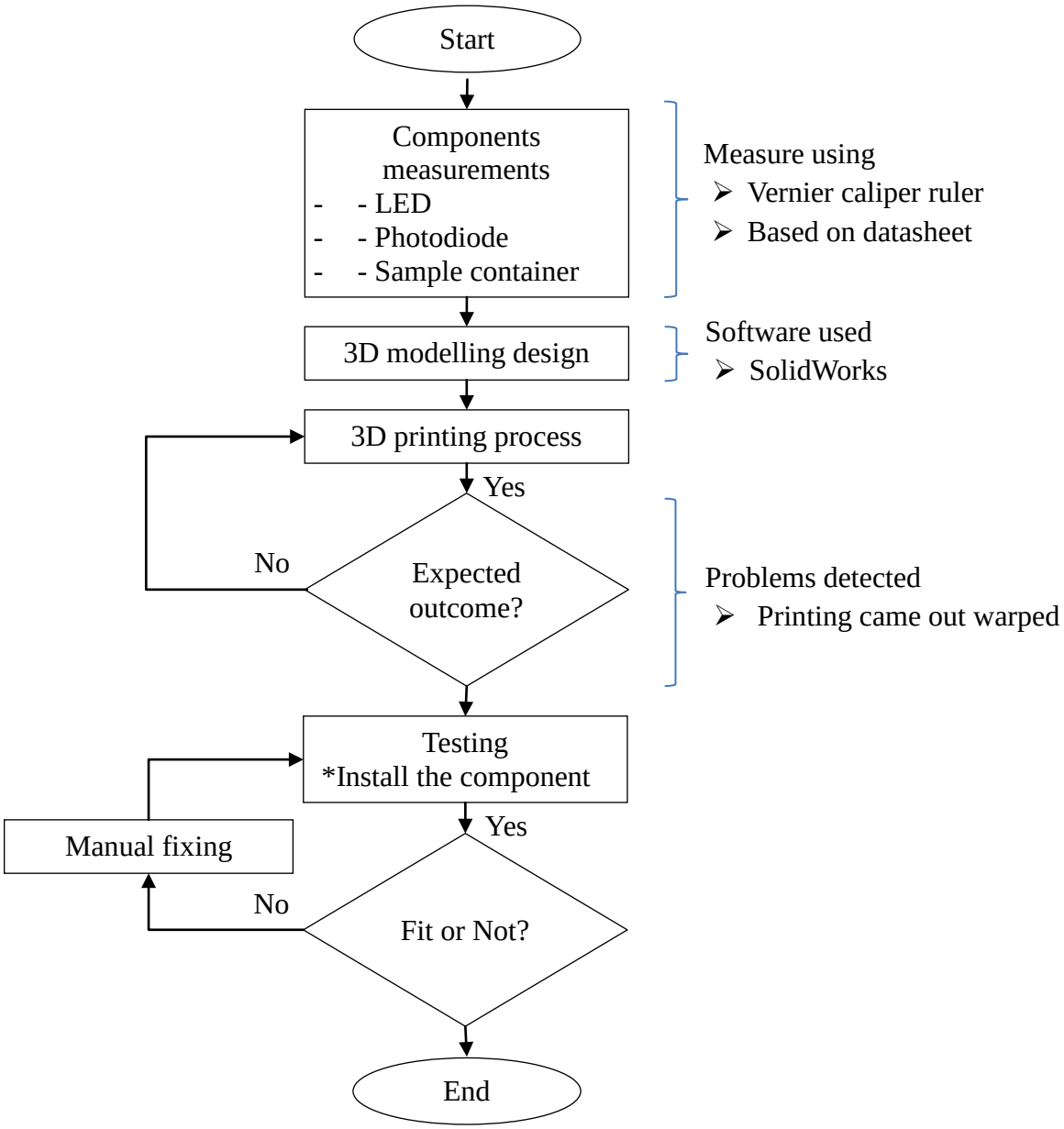


Figure 3.3: The flowchart of custom 3D casing design

3.3.1 Near-infrared and photodiode circuit design

The block diagram of the emitter and the detector conditional circuit is shown in Figure 3.4. This circuit is constructed for near-infrared (NIR) light-emitting diode (LED) as an emitter and photodiode as a detector.

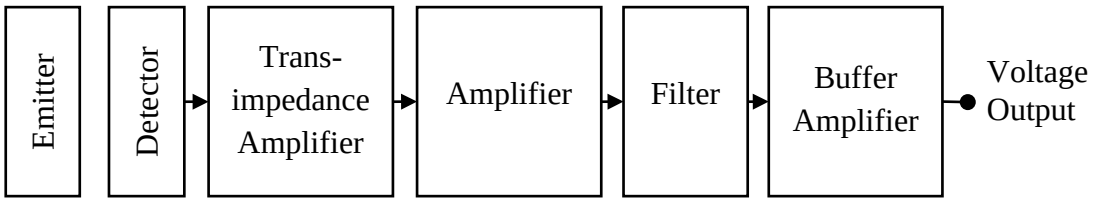


Figure 3.4: Block diagram of emitter and detector circuit

The conditional circuit for NIR LED circuit shown in Figure 3.5 and the NIR LED from Thorlabs as in Figure 3.6 is used as the light source. Table 3.1 shows the electrical specifications for the NIR LED used.

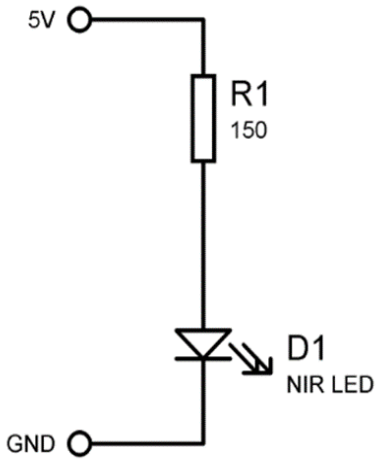


Figure 3.5: NIR conditional circuit

Figure 3.6: Near-infrared LED

Table 3.1: Near-infrared LED Thorlabs electrical specifications (Thorlabs, 2007)

Descriptions	Typical	Max
Power Dissipation		120 mW
Reverse Voltage		5.0 V
DC Forward Current		100 mA
Forward Voltage @ 20 mA	1.2 V	1.5 V
Reverse Current Vr=-5 V		10 µA
Pulsed Current (1 ms pulse with 10% duty cycle)		1000 mA
Operating Temperature		-30˚C to 100 ˚C

Since the maximum forward current that can flow through the NIR LED circuit is 100 mA and the maximum voltage is 5 V. Therefore, based on the (3.1), the minimum resistance that can be used in the circuit is 50 Ω. However, if the value of the resistor used is at minimum value or smaller than the value it should be, then if there are excess current generated exceed the current specification and this can cause LED burned and damaged.

$$R = \frac{V}{I}$$

$$R = \frac{5\text{V}}{100\text{mA}}$$

$$R = 50\Omega \tag{3.1}$$

For the LED brightness and to prevent LED from burning out if there is excess input voltage or over limitation current exist, 150 Ω values of resistance is used in the

circuit as shown in Figure 3.5. The (3.2) shows the current will through the LED is about 33.33 mA.

$$I = \frac{V}{R}$$

$$I = \frac{5\text{V}}{150\Omega}$$

$$I = 33.33\text{mA} \tag{3.2}$$

The range of the NIR wavelength is about 780 nm to 2500 nm. The range of wavelength used in this project is starting from 1050 nm to 1550 nm. Since conventional silicon photodiodes have limited spectral bandwidth, so they cannot be used for receiving NIR light, therefore other types of photodiodes must be considered. The photodiode type of Indium Gallium Arsenide (InGaAs) from Hamamatsu as shown in Figure 3.7 is chosen since it has spectral respond range from 900 nm to 1700 nm. This photodiode has high responded that able to receive the NIR light used in this design. Table 3.2 shows the specification of the photodiode chosen.

Figure 3.7: Package of Indium Gallium Arsenide (InGaAs) photodiode

Table 3.2: Hamamatsu InGaAs photodiode specifications (Hamamatsu Photonics, 2015)

Descriptions	Specifications
Model no.	G12180-010A
Photosensitive area	Ø 1 mm
Package category	TO-18
Spectral response range	900 to 1700 nm
Peak sensitivity wavelength (typ.)	1550 nm
Photosensitivity (typ.)	0.9 A/W
Dark current (max.)	4 nA
Cutoff frequency (typ.)	60 Mhz
Terminal capacitance (typ.)	55 pF

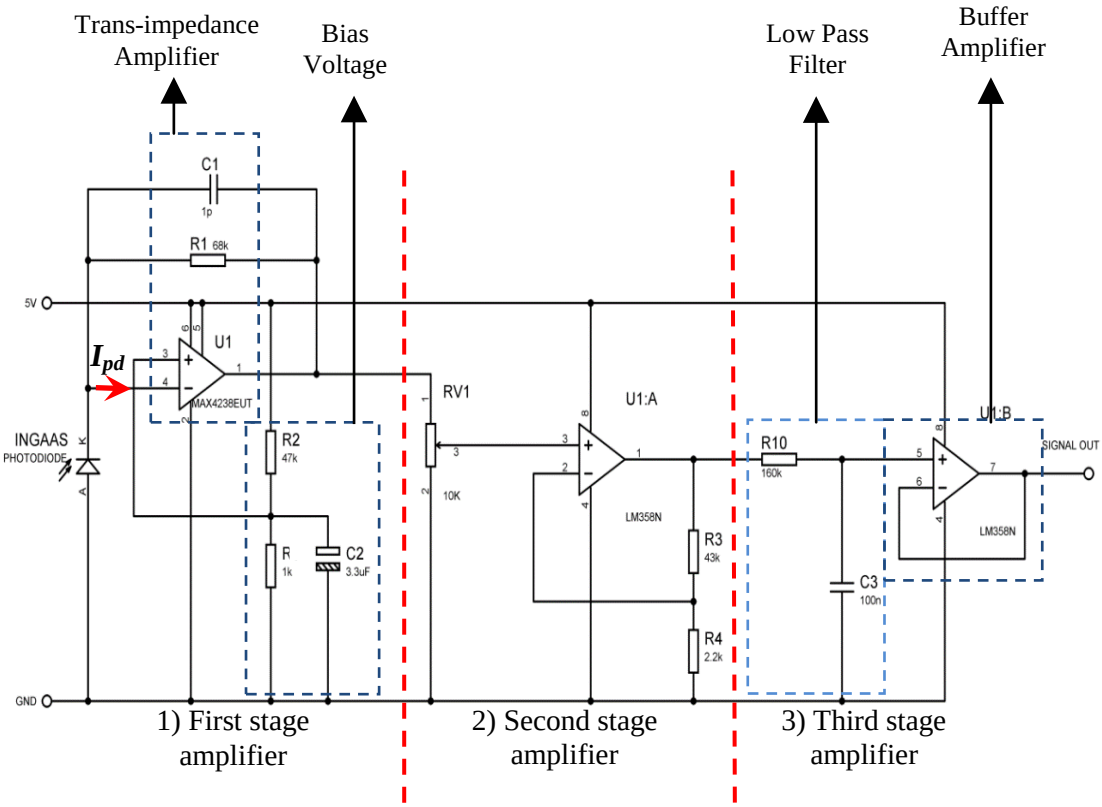


Figure 3.8: Photodiode circuit conditional (Hamamatsu Photonics, 2015)

Figure 3.8 shows the photodiode circuit conditional operation in photoconductive mode and the value of the output voltage is depending on the intensity of the light that receives by the photodiode. This circuit consists of three states of the amplifier, which is the trans-impedance amplifier, non-inverting amplifier and buffer amplifier. The trans-impedance amplifier is used to amplify the light-dependent current of the photodiode. The trans-impedance amplifier consists of op-amp, the feedback resistor (R_1) and capacitor (C_1). The MAX4238EUT was used in the first stage of amplifying and it is a type of current sense amplifier. The purpose of this amplifier was used to amplify a very small input from the current out from the photodiode which is almost zero (0.020µA). The bias voltage was also applied to the non-inverting input op-amp to prevent saturation at the negative power supply. Table 3.3 shows the descriptions of the amplifier used in the photodiode conditional circuit. The capacitor C_1 placed in parallel to R_1 was used for stability. The current to be amplified was applied to the inverting input and the output voltage from the first stage (V_{OUT1}) of an amplifier is dependent on the value of R_1, according to the (3.3).

$$V_{OUT} = i_{PD}R1 + V_B = i_{PD}R_1 + V_{CC}\left(\frac{R}{R+R_2}\right) \tag{3.3}$$

The gain of the non-inverting amplifier in second stage can be calculated from the formula given in (3.4). The common amplifier circuit has fixed level of gain. However, it is useful to be able to vary the gain. In the second stage of an amplifier, a potentiometer is used to vary the gain. The potentiometer can be adjusted to provide a necessary output voltage.

$$V_{OUT} = V_a \times \left(1 + \left(\frac{R_3}{R_4}\right)\right) \quad (3.4)$$

While V_a is the voltage output from the potentiometer. The potentiometer must be at the maximum percentage so that the value of *Va* is equal to value of V_{OUT}.

Table 3.3: Types of amplifier used in the photodiode circuit

Types of op-amp	Descriptions
MAX4238EUT	Current sense amplifiers or current shunt amplifier. This amplifier is used to amplify a very small input current and convert to a small voltage.
LM358N	High-gain, internally frequency compensated operational amplifiers which were designed specifically to operate from a single power supply over a wide range of voltages.

3.3.2 Thermistor sensor circuit

The thermistor was used to measure the human skin temperature. This reading was used only as the references skin temperature during the glucose measurement using the NIR sensor. There are two types of the thermistor, positive temperature coefficient (PTC) thermistor and negative temperature coefficient (NTC). The thermistor is thermally sensitive semiconductors whose temperature-dependent resistor with a non-linear mapping of resistance to the temperature that can be described by the Steinhart-Hart at (3.5) (Steinhart,1968). This equation was used in the microcontroller code to convert the analogue output from the thermistor circuit to the temperature reading. The thermistor is chosen based on its sensitivity, circuit simplicity, price, and accuracy is depending on calibration.

$$\frac{1}{T} = a + b\ln R + c(\ln R)^3 \tag{3.5}$$

Where T is the temperature in Kelvin, R is the measured resistance, and a, b, and c are constants provided by the thermistor manufacturer.

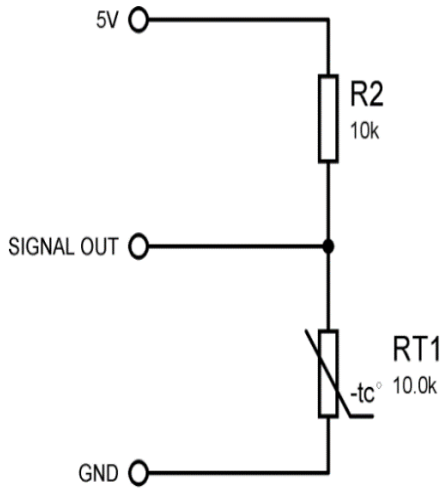


Figure 3.9: The negative temperature coefficient (NTC) thermistor circuit

The connection of the thermistor circuit with a resistance of 10 KΩ is shown in Figure 3.9. Generally, NTC thermistor is type is the most commonly used for temperature sensors as they can be used in virtually any type of equipment where need a measurement of temperature. The NTC, temperature is inversely proportional to the thermistor resistance when the resistance is decreasing, the temperature is rising. The thermistor resistance can be calculated by using potential divider as in (3.6). Table 3.4 shows the detail about the thermistor used.

$$\frac{R_2}{RT_1 + R_2} \times V_{CC} = V_{OUT} \tag{3.6}$$

Table 3.4: Thermistor specification and descriptions

Model	NTCLE100E3
Type	Negative Temperature Coefficient (NTC)
Resistance Load, R_L	10K ohm

3.3.3 Vibration sensor circuit

The vibration sensor was used as movement detection and to improve the stability of the measurement system. The measurement of the NIR sensor only can be made if the position of the sensor is in static. Since certain movement of the NIR sensor can affect the NIR sensor measurement.

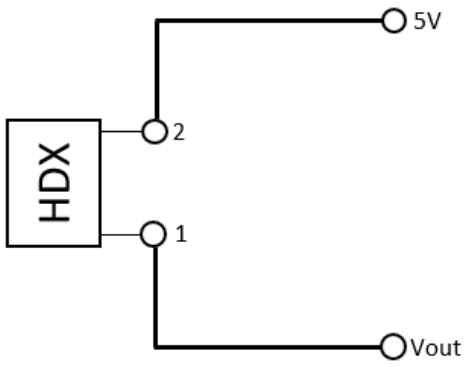


Figure 3.10: Vibration sensor circuit

HDX Vibration sensor was used and the circuit connection is simple as shown in Figure 3.10. The sensor output was connected to the analogue input port on the processor board. The function is based on the coding uploaded to the processor board. The HDX sensor can be placed in two positions as shown in Figure 3.11. Every different sensor position produces different analogue output. Therefore, the HDX sensor needs a calibration to find the analogue reading for the stationary position. The analogue output is used in the code that uploaded in the microcontroller.

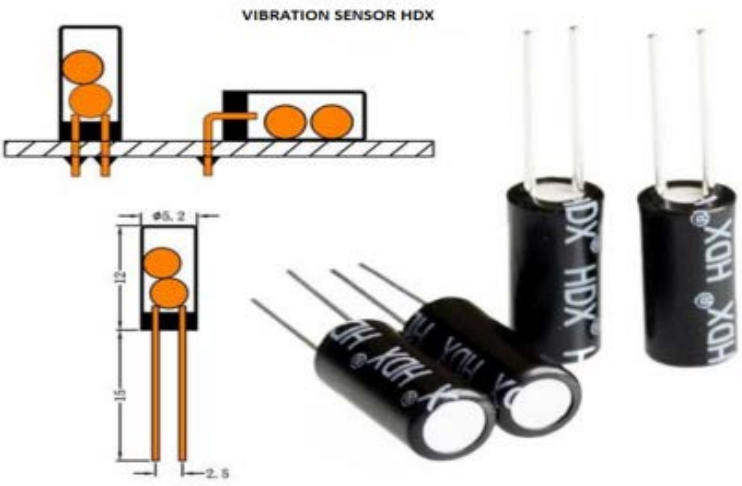


Figure 3.11: HDX vibration sensor available position

3.3.4 Schematic combination circuit

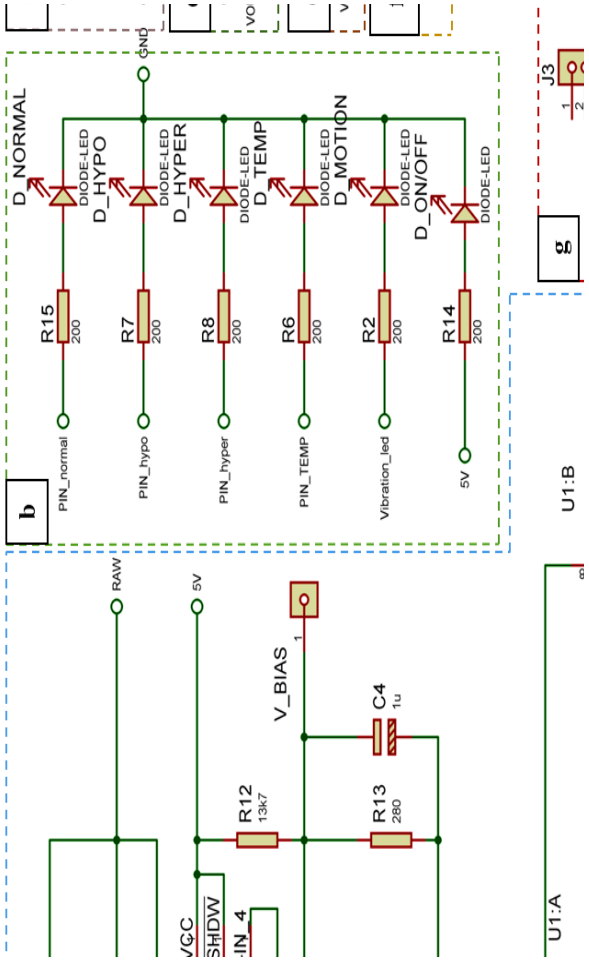


Figure 3.12: The combination of the all circuits with Arduino processing board port; (a) Photodiode conditional circuit, (b) LEDs indicator circuit, (c) NIR

The complete circuit is a combination of all circuits involved. By referring to the Figure 3.12, the full circuit consists of seven parts; (a) Photodiode conditional circuit, (b) LEDs indicator circuit, (c) NIR LED circuit, (d) Thermistor circuit, (e) Vibration sensor circuit, (f) Buzzer, (g) Arduino ports. The LEDs indicator is used to indicate user about on/off switch, vibration detection, change in the glucose reading (hypo or hyper) and temperature. Besides that, the buzzer was also used for the user to make them more alert on the abnormal condition that occurs. This combination of other sensors and indicator is to make the device work more efficient, stable and user-friendly. This circuit is constructed to be connected to the Bluno processing board.

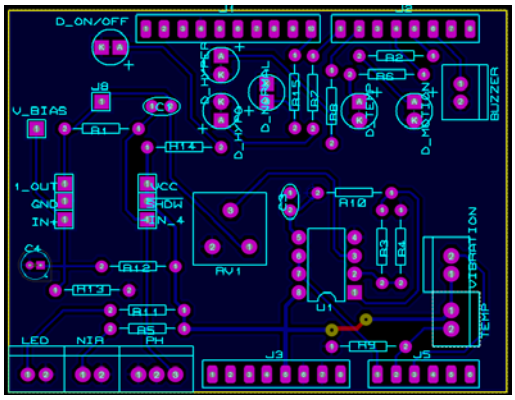


Figure 3.13: Proteus PCB layout view

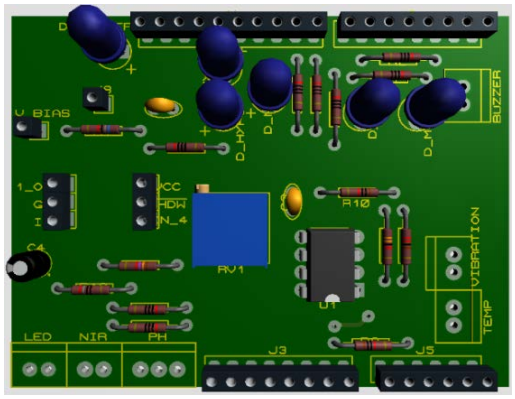
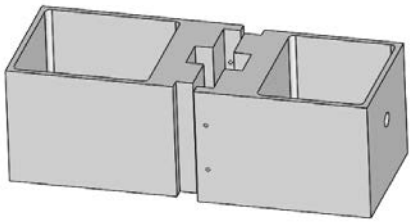


Figure 3.14: The 3D view of circuit board in Proteus software

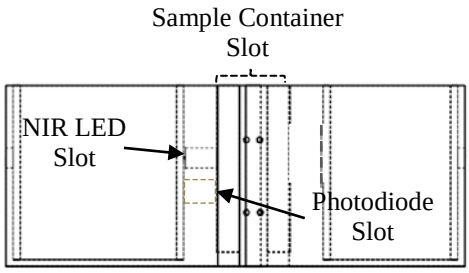
The PCB layout that is used in circuit fabrication is displayed as in Figure 3.13. The circuit fabrication process is done through several laboratory steps such as UV exposed process, developer process (removed unwanted photo resist), etching proses (removed unwanted copper) and drill the component holes. This PCB is used to reduce the complicated to the laborious process of point-to-point wiring. It also can reduce the failures at wire junctions and short circuits. The arrangement of the components can be seen in the 3D view of the PCB in the Proteus software as shown in Figure 3.14. The components can be placed on the PCB before soldering by referring this view.

3.3.5 Sensor casing design

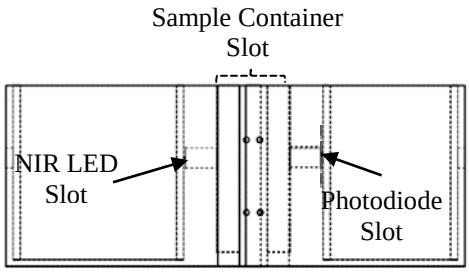
The NIR light and photodiode detector were placed in parallel and facing each other. The purpose of casing design is to eliminate the ambient noise such as unwanted light because this can affect the photodiode output signal.



(a)

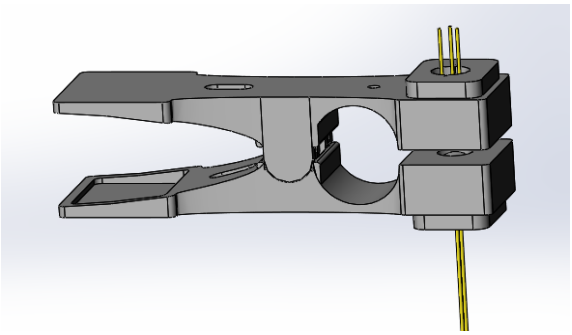


(b)



(c)

Figure 3.15: The casing design for in-vitro experiment used; (a) 3D view, (b) Transmittance sensor configuration, (c) Reflectance sensor configuration



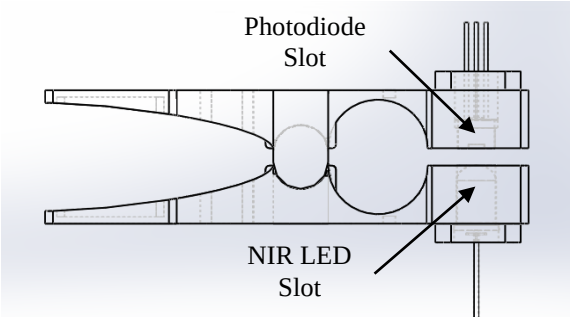


Figure 3.16: The clipper casing design for in-vivo experiment used

In the in-vitro experiment, the casing was designed to hold the sample container (cuvette) and to place the sensor and photodiode in a fix position as shown in Figure 3.15. There are two designs that had been make for two types of sensor configuration. In the in-vivo experiment, the casing was designed like a clipper as in Figure 3.16 to hold the sensor and photodiode in the fixed position on the subject. This casing also eases to place the sensor on the targeted area.

3.4 Glucose sample preparation

The glucose samples had been prepared by following a few steps. This sample prepared for use in the in-vitro experiments and this sample prepared with different concentrations. Powder type glucose was used for glucose concentration sample preparation. This powder is also known as Dextrose. The molecular formula of this powder is $C_6H_{12}O_6$.

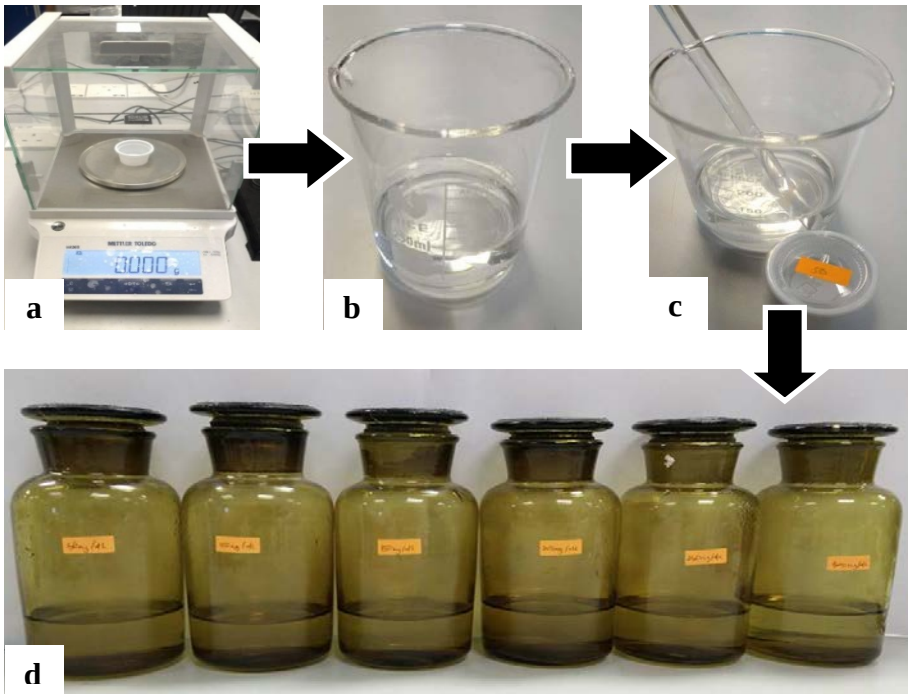


Figure 3.17: Step of glucose concentration sample preparation

The steps of sample preparation are shown in Figure 3.17. The sample preparation starts by weighing the glucose powder based on requirement need. Secondly, 100 mL of distil water iwas measured in a beaker. Then, the glucose powder which has been weighed with the 100 mL distil water is mixed in the beaker and the mixture is stired until dissolved. All the steps before are repeated for different weight of glucose powder (50 mg to 400 mg). The mixture solutions have been stored in the amber reagent bottles to avoid the solution from being affected by light from the unknown outside light and the amber container is labeled to avoid confusion. The sample of glucose concentration is prepared based on the equation of (3.7). Table 3.5 shows the nine different glucose concentrations which have been prepared for use in the in-vitro experiment.

$$\text{Concentrat ion}, \rho i = \frac{\text{Glucose } (\text{mg})}{\text{Water } (\text{mL})} \tag{3.7}$$

Table 3.5: The concentration of glucose solution samples

Glucose Powder (mg)	Distil Water	Concentrations	
		mg/dL	mmol/L
0	100 mL	0	0
50		50	2.775
100		100	5.55
150		150	8.325
200		200	11.1
250		250	13.875
300		300	16.65
350		350	19.425
400		400	22.2

3.5 In-vitro glucose concentration measurement experiment

The transmittance and reflectance of sensor configurations were used in the in-vitro experiment. This experiment conducted in purpose to observe the relationship between the output of various NIR wavelengths and the glucose concentrations based on these two configurations. The configuration with the best output observation was selected for use in the development of non-invasive blood glucose measurement. The experiment was conducted by using five different wavelengths of NIR LEDs; which is 1050 nm, 1200 nm, 1300 nm, 1450 nm, and 1550 nm. This range of wavelengths selected because of the high levels of glucose absorption and low light of absorption to water. Each LEDs were tested by using nine samples with different glucose concentrations as in Table 3.5 and Figure 3.18 shows the flowchart of the in-vitro experiment. The experiments of transmittance and reflectance were conducted by using the same procedure, only the sensor configuration used is different. The valid output voltage for this experiments is between 0V

to 2.5V. The troubleshooting is done if the output reading is not within the valid range. This may be because of circuit problems or sensor positional.

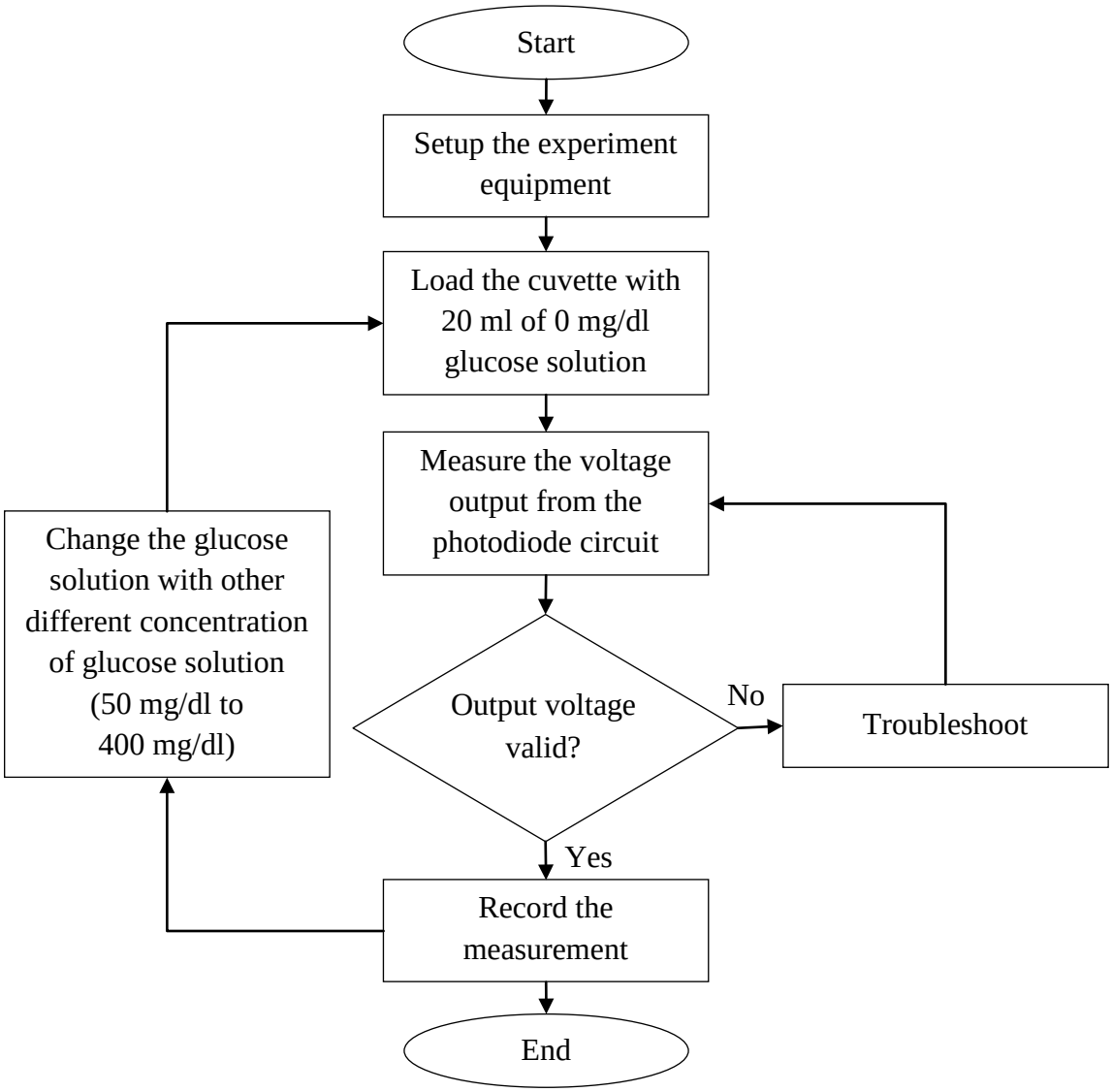


Figure 3.18: Flowchart of the in-vitro analysis

There is a custom casing designed for two different sensor configuration to use in the experiment as shown in Figure 3.15. The cuvette as in Figure 3.19 is used as a sample container to place the sample during the experiment. The cuvette was designed to hold

samples for spectroscopic experiments. A beam of light from NIR can pass through two clear sides and the absorbance value can be observed for analysis.

Figure 3.19: Cuvette fused quartz used for place the sample

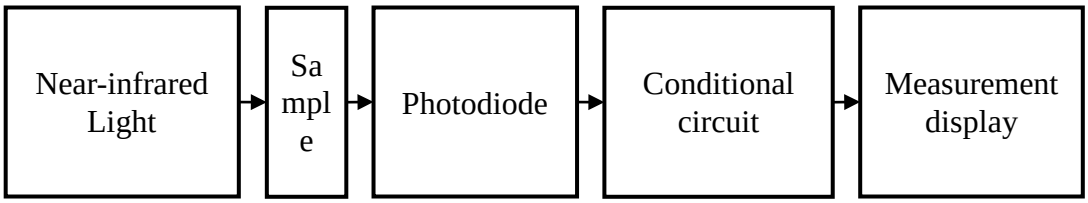


Figure 3.20: Block diagram of transmittance configuration experiment

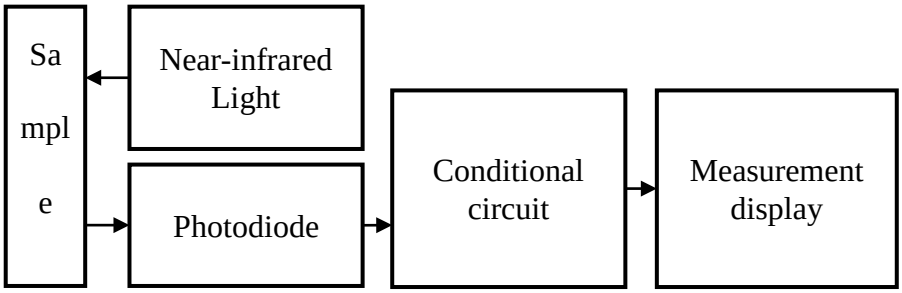


Figure 3.21: Block diagram of reflectance configuration experiment

Block diagram of the in-vitro experiment using a transmittance sensor configuration setup is shown as in Figure 3.20. The NIR light and photodiode was placed facing each other for light transmission. The distance between NIR light and the photodiode is about

12.60 ± 0.5 mm due to the size of cuvette used. The sensor distance used was fixed as it may affect the output measurement. Figure 3.21 shows the block diagram of the in-vitro experiment using a reflectance sensor configuration setup. The NIR light and photodiode is placed side by side for reflection. The distance between NIR light and the photodiode is about 4 ± 0.5 mm.

3.6 In-vivo NIR non-invasive glucose measurement experiment

Based on the analysis results of the in-vitro experiment in subtopic 4.2, the NIR light source with a wavelength of 1450 nm with the sensor configuration of transmittance was used in the in-vivo experiment. Block diagram of in-vivo experimental setup is shown as in Figure 3.22. The in-vivo analysis is conducted by using a human as a subject. There are two types of mathematical models are developed. The first by using samples from a single subject and the second are using samples from multiple subjects. A clipper as in Figure 3.16 is used to place the sensor at the targeted area on the subject.

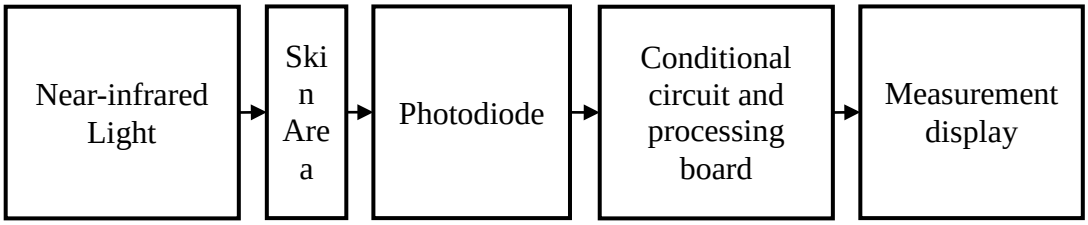
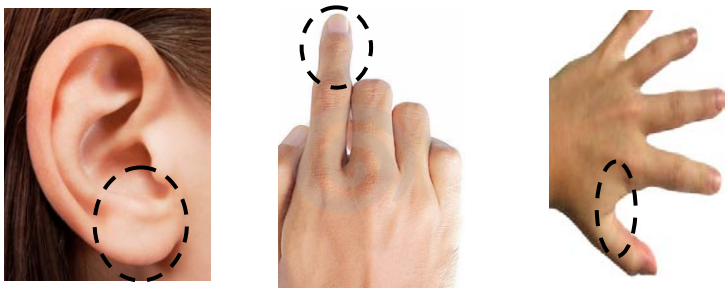


Figure 3.22: Block diagram of in-vivo analysis

3.6.1 Targeted areas for NIR non-invasive experiment

There are three different areas were used in this experiment, which are fingertips, earlobe, and the area between the thumb and index finger as shown in Figure 3.23. The purpose of this experiment is to observe the response of the output sensor based on the

different areas and the area with the best output responses was considered for use as a target area for the final prototype design.



(a) (b) (c)

Figure 3.23: The targeted areas used in the experiment; (a) Earlobe (b) Finger (c) Between thumb and index finger

Ten subjects were used as samples in this experiment. The subjects were chosen based on fair skin tone and age of 25 – 30 years old and with normal body mass index (BMI). The measurement was taken in two conditions, before meals (fasting) and two hours after a meal. The flowchart for this experiment is shown in Figure 3.24. The measurement by using a conventional glucose meter was also taken as a reference. The voltage output of this experiment is 0V to 2.5V based on the circuit that was designed before. The troubleshooting is done if the output reading is not within the valid range. This may be because of circuit problems or sensor positional.

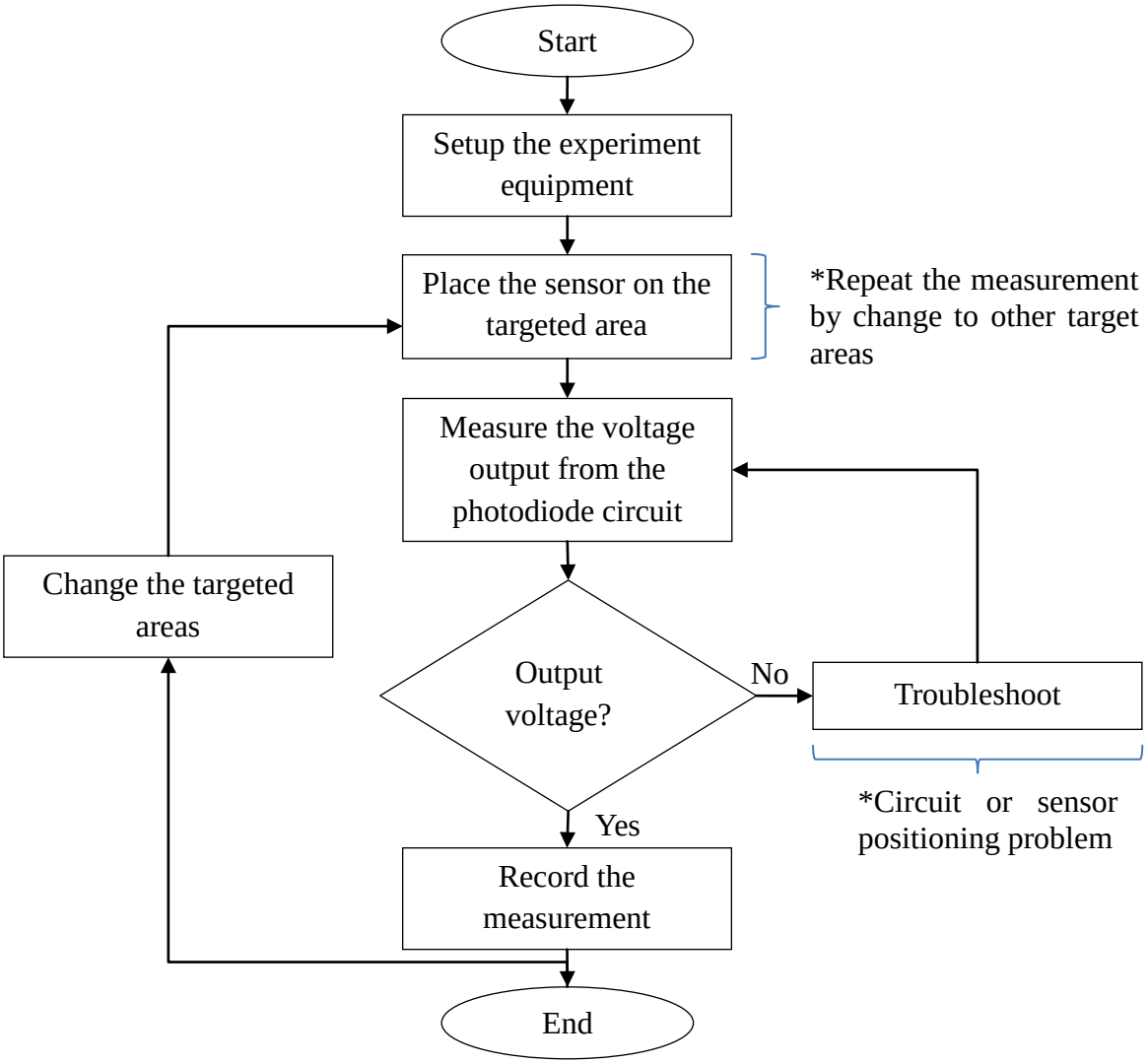


Figure 3.24: The flowchart of the in-vivo analysis using the prototype device

3.6.2 Development of algorithms for NIR non-invasive glucose measurement

There were two types of algorithms developed. The first type of algorithm is based on a single subject used as a sample, while the second type algorithm is developed using multiple subject samples. There were more than one hundred human subjects with multiple genders used in this experiment. Subjects were chosen based on skin tone (fair tone), age (about 25 – 55 years old) and health conditions (normal or diabetic). The algorithms were developed based on the readings taken using both the prototype device and conventional glucose meter at the same time. The prototype device reading was taken in voltage (V),

whereas the conventional glucose meter reading was taken in mmol/L. The sensor prop of the NIR prototype device was placed on the earlobe for measurements, considering that earlobes have good voltage change by comparison to other target areas based on prior analyses. Based on the recorded readings, a graph of conventional glucose readings against prototype device readings was plotted and the line of linear regression equation was carried out using (3.8)–(3.9). The equation line of linear regression was used for algorithm development in the microcontroller. The glucose level in blood can be predicted in mmol/L by using the prototype device based on the linear regression equation (3.9).

$$m = \frac{y_2 - y_1}{x_2 + x_1} \tag{3.8}$$

Where y represent the conventional glucose meter reading (mmol/L), x represent the prototype device reading (V).

$$y = mx + c \tag{3.9}$$

Where m represents the gradient and c represents the y intercept.

3.6.2.1 Single subject algorithm development

The algorithm was developed based on a single subject. Ten readings were taken for every two hours a day, starting from 6.00 a.m until 12.00 a.m. and subject conducted daily activities as usual. Based on the readings recorded, a graph was plotted and the equation line of linear regression from the graph was used in the algorithm.

3.6.2.2 Multiple subjects algorithm development

There were thirty samples used in this algorithm development. The subjects were composed of twenty two non-diabetic subjects and eight diabetic subjects, aged around 25 to 55 years old with fair skin tone. A graph was plotted based on the readings to find the equation line of linear regression for algorithms development.

3.6.3 The invasive blood glucose measurement using glucose meter

The conventional personal invasive glucose meter is a medical device that had been used by a diabetic patient to measure the concentration of blood glucose using blood sample and it can be handled by the patient itself at home. This meter required a small amount of blood sample to be placed on the test strip, but little or too much blood volume may cause a failure for meter read it. The reading is displayed in the units of glucose concentration which is mg/dL or mmol/L. The equipment for the glucose meter use consists of blood glucose meter, test strips, lancet/lancet device, and alcohol swabs as shown in Figure 3.25. There are few of precaution steps that need to be followed in the process of measuring blood glucose level using a glucose meter and the steps are described in Figure 3.26.

Figure 3.25: Invasive glucose meter equipment

Figure 3.26: Steps of blood glucose measurement using a glucose meter.

3.7 Prototype device testing and data verification

The final prototype was tested to more than hundred subjects (no-diabetic and diabetic subjects), age range about 25 – 55 years old. There are two algorithms was tested in these experiments and each algorithms applied different mathematical model. The subjects were requested to sit quietly in a chair and the sensor clipper is placed on the same target area for every subject. The glucose reading was taken after the device give a stable reading. The glucose reading from the prototype device was verified based on the

conventional method which by using an invasive glucose meter. The readings measured from both the prototype device and conventional glucose meter were taken simultaneously, and both were taken in mmol/L. The Clarke EGA graph was plotted for every test to determine the accuracy of glucose measurement using the prototype device developed.

The algorithm applied the mathematical model of a single subject was further tested in three ways. The first was using the same single subject used to develop the algorithm before, and the second way was using a different single subject. The reading was taken for every two hours from 6.00 a.m until 12.00 a.m for three days and the third way was by using multiple subjects. The third way was to use multiple subjects; whereby seventy healthy subjects and ten diabetic subjects were used to test the algorithms and all the subjects selected had similar skin tone (fair tone).

The mathematical model of multiple subject was apply to the algorithm was tested in two ways. The first way was by using a single subject and the readings were taken for every two hours from 6.00 a.m until 12.00 a.m for three days. The second way was by using multiple subjects to test the algorithms. The subjects consist of seventy healthy subjects and ten diabetic subjects aged between 25 to 55 years old with similar fair skin tone.

3.8 Development of prototype non-invasive blood glucose measurement experiment

The development of non-invasive blood glucose measurement system is a combination of hardware and software development. The main focus of the software programming development is based on the microcontroller. The Bluno V1.4 microcontroller board was used as shown in Figure 3.27. This board is based on Atmega328 chip, has 14 digital input and output pins and Bluetooth Low Energy (BLE).

The Bluno was used as the controller for the device as it is an open source which is easy to code and upload to the input/output (I/O) board. The open source Arduino code is known as integrated development environment (IDE).

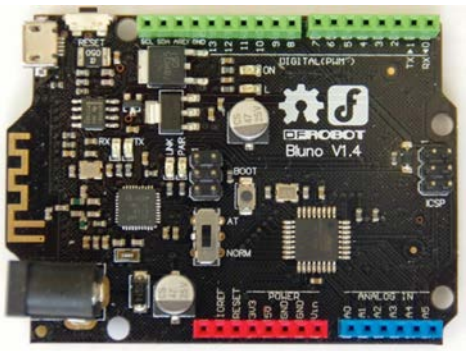


Figure 3.27: The Bluno microcontroller board

The block diagram of the circuit connection to the microcontroller board was demonstrated as in Figure 3.28. Bluno board was used to control the whole process. The microcontroller supplies voltages, 5V to bias all the sensors used.

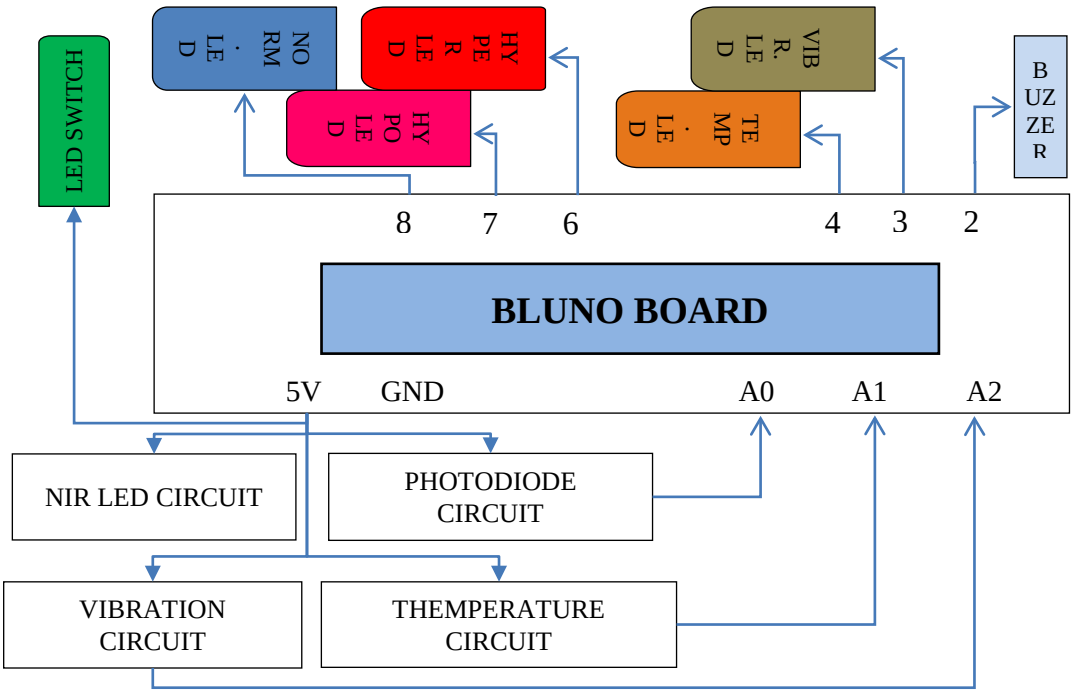


Figure 3.28: Block diagram connection on microcontroller board

The microcontroller has been coded with an algorithm to make every sensor used function well in this system. The flowchart of the microcontroller system is shown in Figure 3.29 until Figure 3.31. The system consists of four parts and there are LEDs for all parts as an indicator. The first part is switching LED indicator. This LED is turned on when there is a voltage supply on the processor board and when the device is ready making the measurement. The second part is temperature measurement. The temperature sensor is used as skin temperature monitoring, since the NIR sensor can be affected by the temperature. The temperature LED light is turned on when the temperature reading is in unstable condition and this is only used to inform user about their skin temperature. This information is very helpful and this measurement will become a guideline to the user whether to make a measurement straight away or to wait until the skin temperature is at a normal state.

The next part is the vibration measurement. This vibration sensor used to detect the sensor prop movement during the measurement. The glucose reading can be affected by the heavy movement. When there are movement detected, the vibration LED's light is turned on and the glucose will not be read until there are no movement detected. The last part is glucose measurement. There are LEDs indicator and buzzer used to inform user about hypoglycaemia (low glucose) and hyperglycaemia (high glucose) condition. The glucose reading is based on the linear equation of the linear regression that has been coded in the microcontroller processing board used.

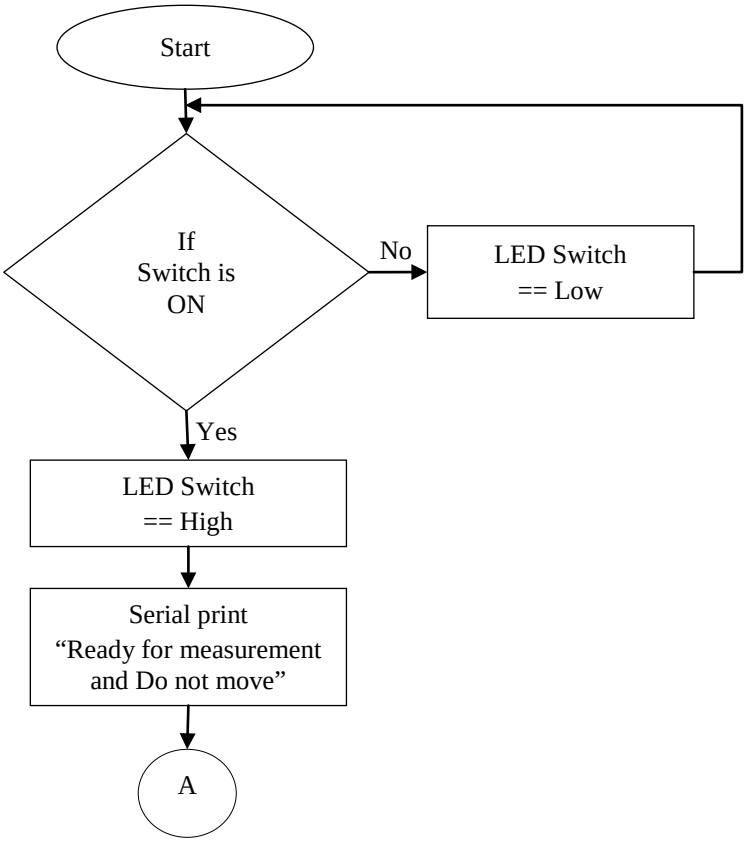


Figure 3.29: The flowchart of the microcontroller system (switching part)

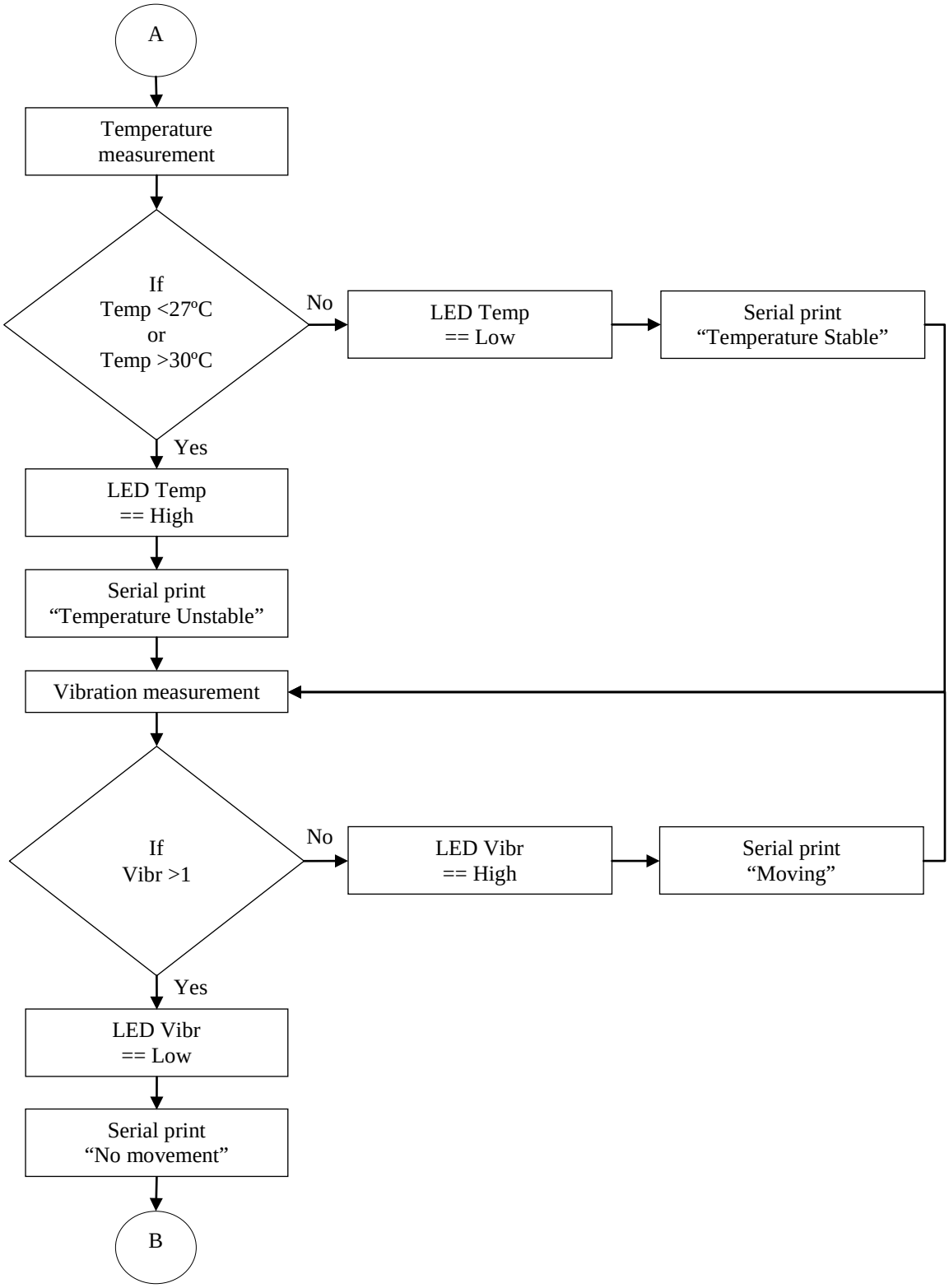


Figure 3.30: The continuity of the microcontroller system flowchart (temperature and movement part)

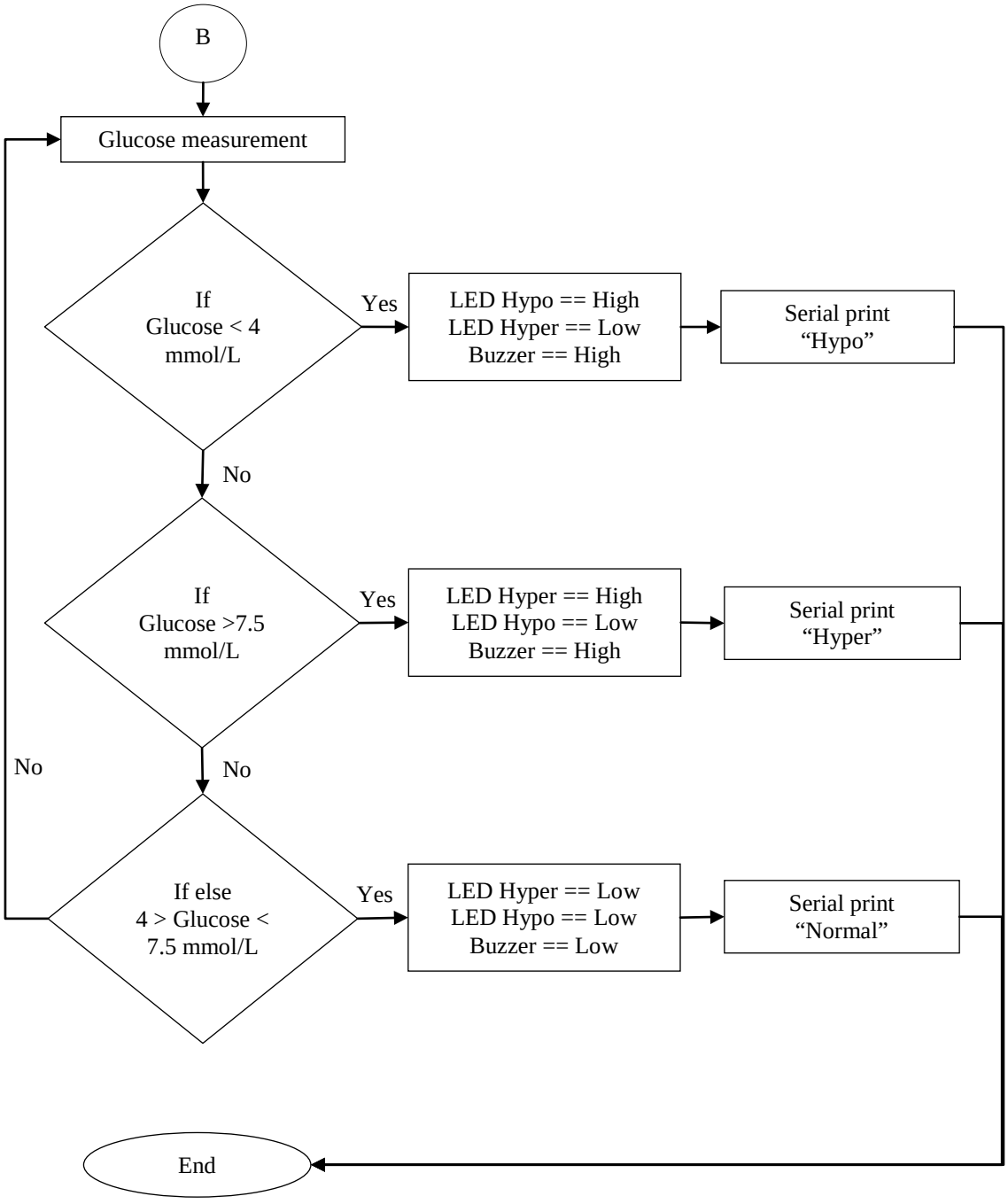


Figure 3.31: The continuity of the microcontroller system flowchart (glucose and LED indicator)

3.9 Summary

The design procedure for development of NIR non-invasive blood glucose monitoring has been described in every detail in this chapter. The mathematical model and algorithm design for the development of a prototype system were also discussed in detail. Besides, the procedures of all conducted experiments are explained clearly. The results of the experiments are presented and discussed in Chapter 4. In addition, the final design and functions of the prototype are also presented in Chapter 4.

CHAPTER 4

RESULT AND DISCUSSION

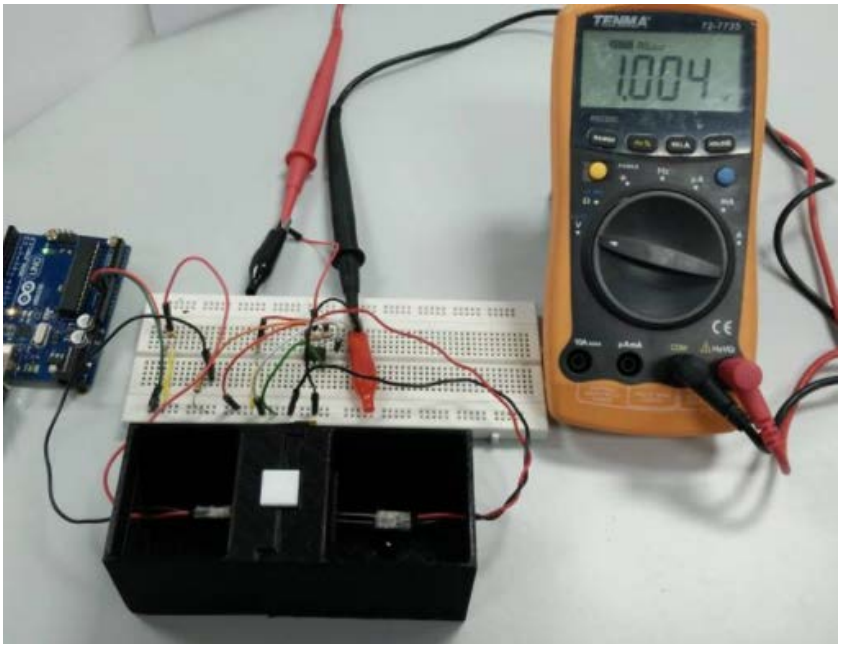
4.1 Introduction

In this chapter, the focuses are on the experimental outputs and analysis of the overall system. The results are recorded and discussed in detail. Tables and graphs are used to facilitate the compilation of results and discussions. The mathematical model that is used in the development of blood glucose monitoring algorithms is also derived from the graph plotted in this chapter. In addition, a comparison between the conventional method of self-monitoring blood glucose (SMBG) and the proposed device is explained in this chapter. Besides that, the NIR non-invasive blood glucose prototype device system is also explained in this chapter such as how the device is functions.

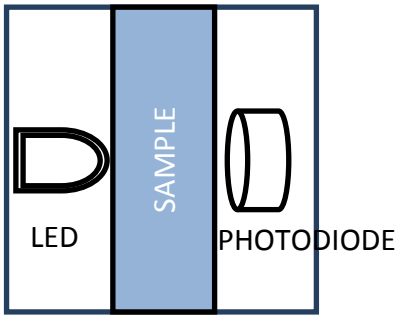
4.2 In-vitro experiments

The in-vitro experimental setup is shown as in Figure 4.1(a). The black box casing designed using a 3D printer is to hold the sensors and the sample container during the experiment is conducted. There are five different wavelengths (1050 nm, 1200 nm, 1300 nm, 1450 nm, and 1550 nm) and based on equation (3.7), nine different concentrations of glucose sample are used in these experiments as shown in Table 3.5. Two different sensor configurations are used in these experiments; transmittance and reflectance configuration as shown in Figure 4.1(b) and (c). In-vitro experiment setup is designed to investigate the attenuation of NIR light by glucose concentration. In addition, the experiments also conducted to find one of the best wavelengths among the tested five

wavelengths that can be absorbed well by glucose solution and the best sensor configuration to be used in the development of non-invasive blood glucose monitoring prototype that has been proposed.



(a)



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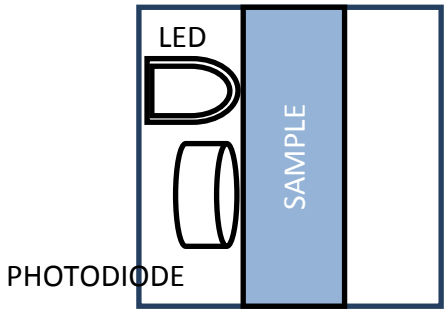


(c)

Figure 4.1: In-vitro experiment; (a) In-vitro experiment setup (b) Transmittance sensor configuration (c) Reflectance sensor configuration

4.2.1 Transmittance sensor configuration respond

The transmittance sensor configuration experimental setup is shown as in Figure 4.1. The voltage measurements using the transmittance sensor configuration are

recorded as shown in Table 4.1. The range of voltage outputs are increasing with the increase of the light source wavelength used and this happen due to the increase of the penetration light through the sample. While, the output voltages changes decrease in millivolt (mV) for every increment of glucose concentrations. Since the level of sample concentration can affect the light absorbance by the sample and the transmitted light.

Table 4.1: The output voltages of NIR transmittance configuration sensor based on different glucose concentrations

	Output Voltage (V)				
Wavelength / **Concentration (mg/dL)**	1050nm	1200nm	1300nm	1450nm	1550nm
0	0.9345	1.0550	1.1553	1.3440	1.6005
50	0.9300	1.0540	1.1557	1.3435	1.600
100	0.9295	1.0490	1.1475	1.3425	1.5920
150	0.9295	1.0465	1.1470	1.3405	1.5855
200	0.9270	1.0380	1.1430	1.3395	1.5800
250	0.9230	1.0330	1.1435	1.3355	1.5800
300	0.9215	1.0300	1.1415	1.3340	1.5780
350	0.9130	1.0180	1.1400	1.3300	1.5725
400	0.9075	0.9915	1.1390	1.3280	1.5725

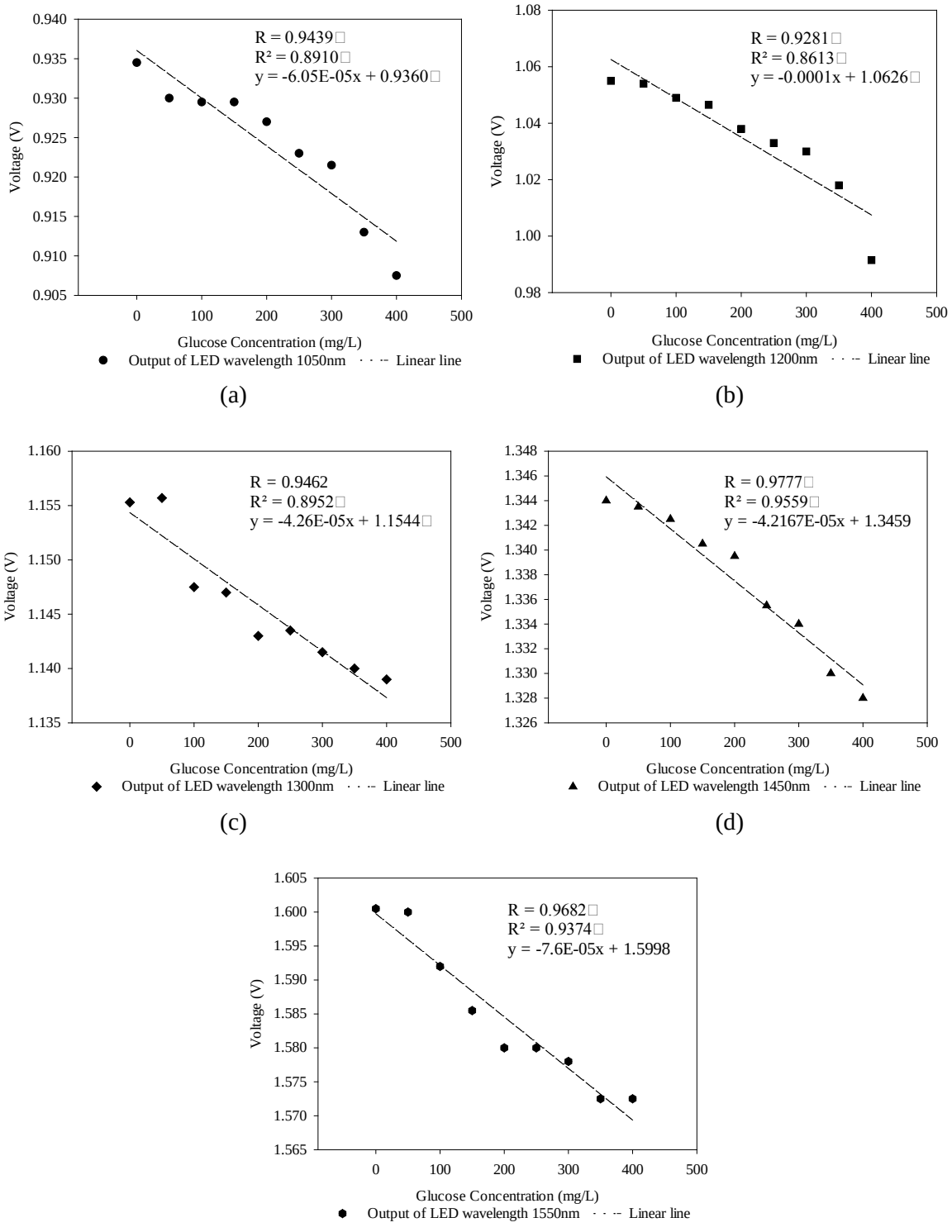


Figure 4.2: The relationship between voltage outputs of NIR transmittance configuration and glucose concentrations based on wavelengths; (a) 1050 nm (b) 1200 nm (c) 1300 nm (d) 1450 nm (e) 1550 nm

The graphs of relationships between voltage output of the NIR transmittance configuration and glucose concentrations are inversely proportional as presented in Figure 4.2 and it is plotted according to the range of NIR wavelengths. By referring to the graphs plotted, it shows that the output voltages of the photodiode are decreased when the glucose concentrations are increased; which shows that the light detected by the photodiode is decreased when the glucose solution is more concentrated. The line of linear regression equations and prediction correlation coefficient (R^2) values for each of the graphs are tabulated in Table 4.2. The line of linear regression equation is calculated based on the given equation in (3.9). The R^2 value is the regression value which indicates the correlation coefficient between two variables. NIR LED with a wavelength of 1450 nm has the best value of R^2 compared to other wavelengths, which is 0.9559. That means that there is a good correlation between LED wavelength 1450 nm and the glucose concentrations.

Table 4.2: Line of linear regression equations and prediction correlation coefficient (R^2) regressions based on graph Figure 4.2

Wavelengths (nm)	**Equations line of linear regression**	**Prediction correlation coefficient (R^2)**
1050	y = -6E-05x + 0.936	0.8910
1200	y = -0.0001x + 1.0626	0.8613
1300	y = -4E-05x + 1.1544	0.8952
1450	y = -4E-05x + 1.3459	0.9559
1550	y = -8E-05x + 1.5998	0.9374

4.2.2 Reflectance sensor configuration respond

The reflectance sensor configuration experiment used the same setup as shown in Figure 4.1 and the only change made was the black box casing to suit the LED and photodiode since this experiment used reflectance sensor configuration. Table 4.3 shows

the voltage output of five different wavelengths based on different glucose concentrations. The voltage output of photodiode in this sensor configuration is smaller than the voltage output by transmittance sensor configurations. The output voltage changes decrease in millivolt (mV) for every increment of glucose concentrations.

Table 4.3: The output voltages of NIR reflectance configuration sensor based on different glucose concentrations

	Output Voltage (V)				
Wavelength / **Concentration (mg/dL)**	1050nm	1200nm	1300nm	1450nm	1550nm
0	0.1535	0.1763	0.8685	0.9595	1.0235
50	0.1535	0.1762	0.8650	0.9560	1.0210
100	0.1532	0.1762	0.8630	0.9555	1.0215
150	0.1517	0.1754	0.8470	0.9495	1.0205
200	0.1516	0.1756	0.8445	0.9410	1.0180
250	0.1513	0.1750	0.8420	0.9355	1.0115
300	0.1508	0.1749	0.8110	0.9345	1.0115
350	0.1482	0.1746	0.7820	0.9340	1.0070
400	0.1447	0.1747	0.7675	0.9295	1.0055

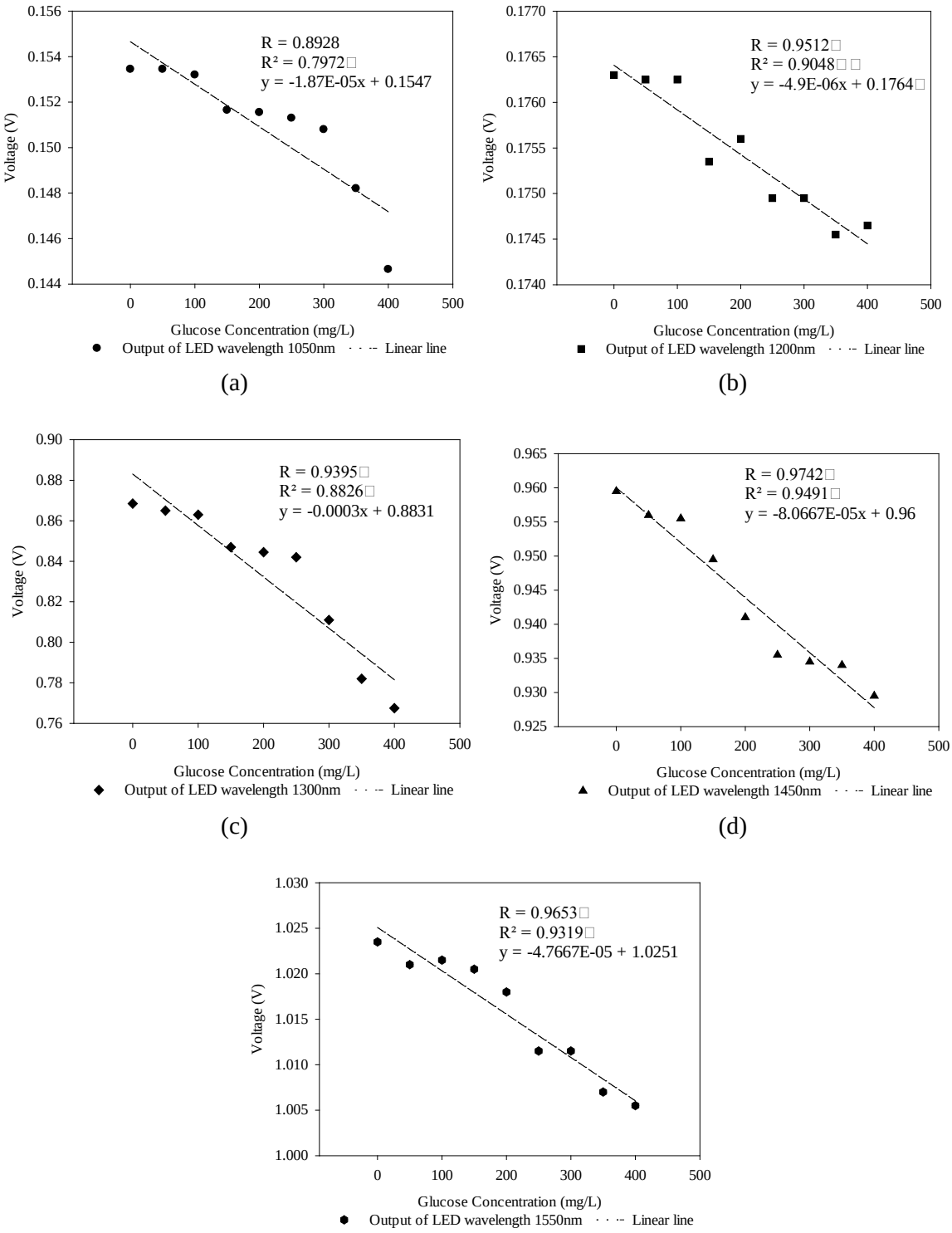


Figure 4.3: The relationship between voltage outputs of NIR reflectance configuration and glucose concentrations based on wavelengths; (a) 1050 nm (b) 1200 nm (c) 1300 nm (d) 1450 nm (e) 1550 nm

Figure 4.3 shows the graphs of the relationship between voltage outputs of NIR reflectance configuration and glucose concentrations also inversely proportional as transmittance outputs; and it is plotted according to the voltage output recorded in Table 4.3. Based on the graphs plotted, it shows that the output voltages are decreased when the glucose concentrations are increased; which indicates that the light detected by the photodiode is decreased when the glucose solution is more concentrated and this due to the Beer-Lambert law. The linear equation and prediction correlation coefficient (R^2) for every graph were calculated and tabulated in Table 4.4 and it can be concluded that the wavelength with the best R^2 of 0.9491 is 1450 nm.

Table 4.4: Line of linear regression equations and prediction correlation coefficient (R^2) regressions based on graph Figure 4.3

Wavelengths (nm)	**Equations line of linear regression**	**Prediction correlation coefficient (R^2)**
1050	y = -1.87E-05x + 0.1547	0.7972
1200	y = -4.9E-06x + 0.1764	0.9048
1300	y = -0.0003x + 0.8831	0.8826
1450	y = -8.0667E-05x + 0.96	0.9491
1550	y = -4.7667E-05 + 1.0251	0.9319

Based on the data recorded in Table 4.1 and 4.3, there is an overlapping of output voltage between different glucose concentrations. This may be due to the small differences in glucose concentrations which caused difficulty for sensors to accurately differentiate these measurements. In addition, the voltage difference for every glucose concentration was very small and thus difficult to be read by the multimeter.

The wavelength of 1450 nm was chosen out of other wavelengths tested since it has the best R^2 regression value in both experiments; which signifies better absorption of

glucose concentration solution in this NIR wavelength. Two different sensor configurations setup of reflectance and transmittance were investigated. Based on the R^2 regression values, it was proven that the transmittance configuration is a better option than the reflectance configuration for the development of the non-invasive blood glucose measurement. According to research conducted by Jeon et al. (2006), the author also recommended the transmittance configuration setup for the application by comparison to reflectance configuration setup, due to the fact that the reflectance analysis does not show good glucose absorption features. The reflected light may not have enough glucose information since a major portion of the detected light have short optical path length. In addition, prediction also becomes more dependent on medium scattering rather than glucose concentration, compared with transmission measurement.

4.3 Results and discussions of in-vivo NIR non-invasive glucose measurement experiment

(a)

(b)

Figure 4.4: Experiment setup; (a) In-vivo experiment setup (b) Clipper casing for sensor holder

The in-vivo experiment was used to test the NIR non-invasive blood glucose monitoring prototype. Based on the results from the in-vitro analysis before, the NIR LED of 1450 nm wavelength and the transmittance sensor configuration were used in the prototype system. Figure 4.4(a) shows the in-vivo experiment setup and clipper casing as in Figure 4.4(b) was used to hold the sensor during the experiment. There were two types of experiments conducted in this part. The first experiment was conducted based on three different target areas which were a finger, the area between the thumb and the index finger, and the earlobe. The purpose of this experiment is to identify the best area among these three target areas to place the sensor prop for the non-invasive blood glucose measurement. On the other hand, the second experiment was conducted to develop a mathematical model that will be used in the non-invasive blood glucose conversion algorithm. The selected subjects based on the specified characteristics which are age of 25 – 30 years old, have normal body mass index (BMI) and with fair skin tone. These characteristics are intended to control the physical differences of for each subjects The skin tone is identify based on skin undertone spectrum as shown in Figure 4.5.

Figure 4.5: The skin undertone spectrum (Sison, 2017)

4.3.1 Targeted areas for NIR non-invasive experiment

In this experiment, ten non-diabetic subjects were considered as samples for this test. Informed consents were obtained from all the participants prior to the conductance of the test. The output voltage readings were taken twice for each subject on the three aforementioned different areas; and glucose meter readings were also taken as reference. The first reading was taken before meal while the second reading was taken one hour after the meal. This was done in order to obtain a variety of output voltage ranges. The sensor prop was placed on the targeted areas simultaneously. The conventional home-use invasive glucose meter was used to test the subject's glucose level which was subsequently used as references. The output voltages for the three target areas are recorded in Table 4.5.

Table 4.5: The measurements of glucose level before and after the meal based on three target areas

	Voltage (V)						**(mmol/L)**	
Areas / **Samples**	**Finger**		**Bet. thumb and index finger**		**Earlobe**		**Glucose meter**	
	Before meal	**After meal**	**Before meal**	**After meal**	**Before meal**	**After meal**	**Before meal**	**After meal**
1	1.163	0.975	1.412	1.307	1.392	1.168	6.1	7.3
2	1.131	1.106	1.396	1.284	1.386	1.098	6.5	7.7
3	1.166	0.956	1.415	1.302	1.366	1.138	6.6	7.6
4	1.243	1.071	1.403	1.129	1.431	1.156	5.8	7.3
5	1.156	0.972	1.396	1.192	1.388	1.171	6.0	7.2
6	1.088	0.893	1.372	1.247	1.289	1.065	6.6	8.0
7	1.152	0.975	1.409	1.287	1.456	1.177	5.8	7.2
8	1.239	1.002	1.395	1.180	1.472	1.172	5.6	7.3
9	1.130	0.970	1.418	1.276	1.379	1.051	6.2	8.1
10	1.161	0.990	1.418	1.311	1.368	1.184	6.4	7.2

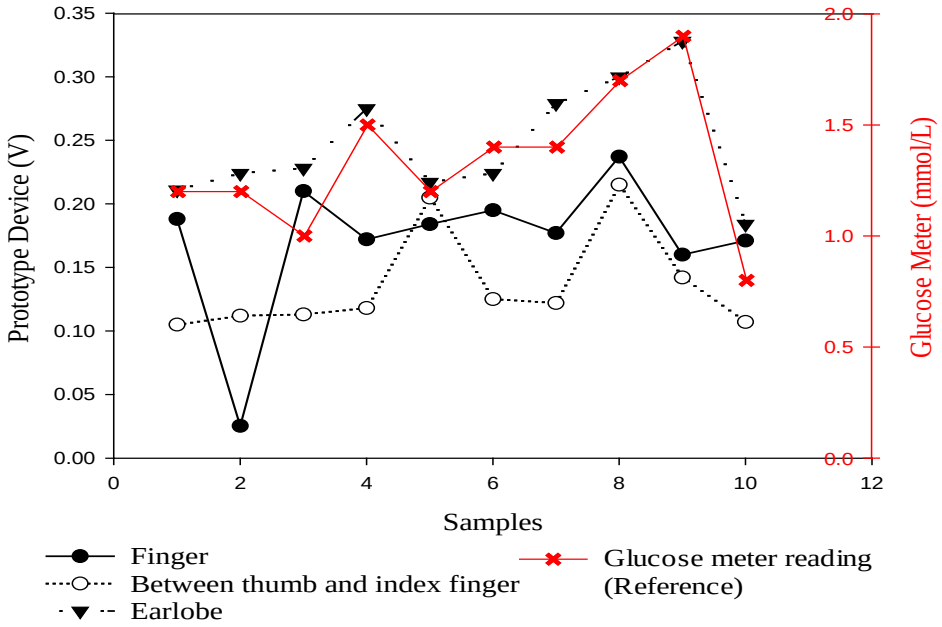


Figure 4.6: The change pattern before and after the meal based on the targeted area

The output voltage of photodiode and the glucose meter readings based on the three different target areas are recorded in Table 4.5. From the table, it is shown that after the meal, the glucose meter reading increases while the output voltage of photodiode decreases. This is due to lack of NIR light signal detected by the photodiode when the glucose concentration in the blood was increasing. Figure 4.6 depicts the changes in the output voltage pattern taken before and after the meal. Maximum changes of output voltage can be observed in the case of earlobe compared with the finger area, and the area between thumb and the index finger. The change in the reading of glucose meters was used as a reference because glucose meter is the most accurate tool for reading blood glucose levels. From that, the earlobe area shows that it has the most similar pattern to the glucose meter graph pattern compared to the other two areas. Therefore, the earlobe was chosen as the best area to place the sensor prop of prototyping for non-invasive blood glucose. This phenomenon is due to the absence of bone tissue and also due to its relatively small

thickness, since the thickness of the sample will affect the light intensity after it passes through the sample by referring to the Beer-Lambert law as in equation (2.4). The earlobe area was used by Kamboh and Khan, (2013) for non-invasive blood glucose monitoring using the NIR spectroscopy. Besides, the earlobe was also suggested by Yadav et al. (2015) based on the comparative study of different measurement sites using NIR.

4.3.2 Algorithms development for non-invasive glucose detection

These experiments were conducted to develop a mathematical model to be used in the algorithms of the non-invasive blood monitoring. The algorithms are programmed in the microcontroller used in this project. There were two types of mathematical models that have been developed. The first model uses a sample from a single subject, whereas the second model uses a sample of multiple subjects.

Table 4.6: The measurements of the prototype output voltage and invasive glucose meter based on samples from a single subject

Reading Time	Prototype Device (V)	Glucose Meter (mmol/L)
6:00 am	1.423	4.9
8:00 am	1.389	5.6
10:00 am	1.189	6.1
12:00 pm	1.107	6.3
2:00 pm	1.03	6.5
4:00 pm	0.983	6.6
6:00 pm	1.151	6.2
8:00 pm	1.026	6.5
10:00 pm	1.323	5.8
12:00 am	1.372	5.7

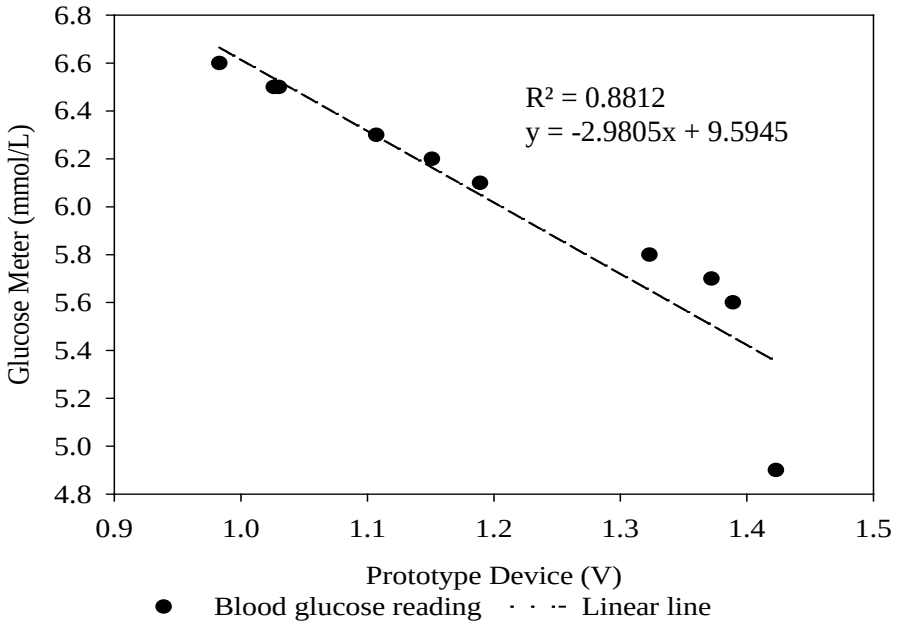


Figure 4.7: Line of linear regression graph of output glucose meter against output voltage non-invasive prototype device based on samples from single subject

The measurements for the single subject were taken ten times throughout the day starting at 6.00 a.m until 12.00 a.m by using both the prototype device and the conventional invasive glucose meter as shown in Table 4.6. The changes in glucose readings in the blood may be caused by the food intake or activities performed by the subject. Figure 4.7 shows a graph with the regression line plotted based on readings recorded from the single subject samples. The blood glucose level readings by the invasive glucose meter are inversely proportional to outputs voltage of the prototype device. The glucose meter readings decrease when the voltage outputs of the prototype device are increased. This makes the regression line run down through the data. The R^2 value and equation are generated from the linear regression line. The value of R^2 is 0.8812, this is a fairly decent model to use. The equation as shown in (4.1) is used as a mathematical model

in the development of the algorithm to predict the value of the glucose level by using the prototype device.

$$y = -2.9805\,x + 9.5945 \quad (4.1)$$

Where y represent the predicted glucose level and x represent output voltage from the prototype device.

Table 4.7: The measurements of the prototype output voltage and invasive glucose meter based on samples from multiple subjects

Samples	Prototype Device (V)	Glucose Meter (mmol/L)
1	1.392	5.7
2	1.277	6.3
3	1.382	5.8
4	1.285	6.1
5	1.212	6.6
6	1.359	5.6
7	1.389	5.2
8	1.41	4.9
9	1.102	8.2
10	1.069	7.7
11	1.414	4.2
12	1.418	3.7
13	1.405	4.8
14	1.092	8.4
15	0.982	9.2
16	0.978	11.2

17	1.419	3.9
18	1.189	6.1
19	0.963	9.1
20	1.388	5.4
21	1.392	4.9
22	1.157	6.8
23	1.229	6.4
24	1.115	7.1
25	0.989	9.4
26	1.212	6.5
27	1.412	4.1
28	1.391	5.3
29	0.981	8.7
30	1.352	5.1

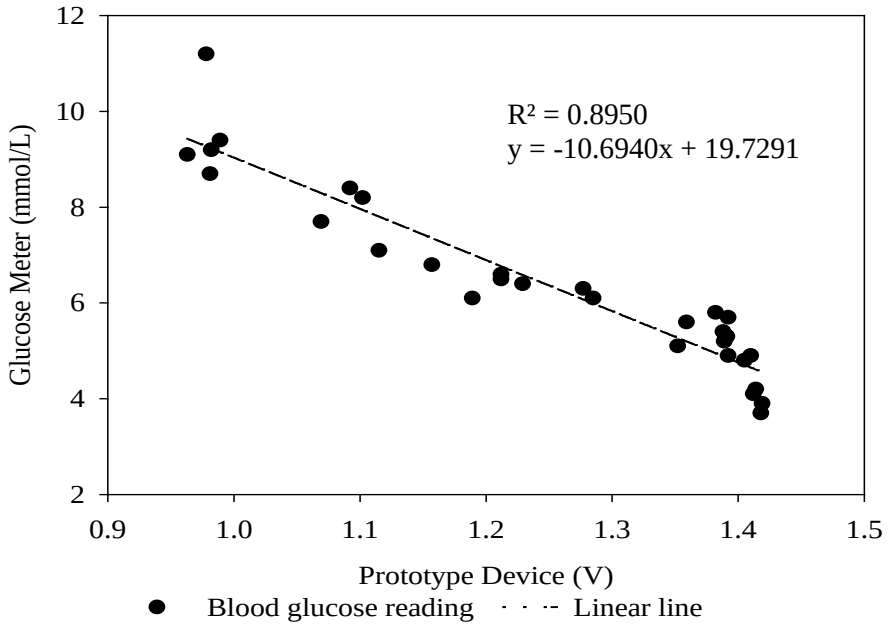


Figure 4.8: Line of linear regression graph of output glucose meter against output voltage non-invasive prototype device based on samples from multiple subjects

There were thirty subjects used as samples for mathematical model development. The measurement was taken once for every subject by using the prototype device and conventional invasive glucose meter. Table 4.7 shows the recorded data. Different subjects gave different readings, but they were of the same range. A graph with regression line that is plotted based on readings recorded from the multiple subject samples is shown in Figure 4.8. The blood glucose level readings by the invasive glucose meter are inversely proportional to output voltage of the prototype device. The regression line is running down through the data and generates the value of R^2 and the linear equation. The value of R^2 is 0.8950, this is a fairly decent model to use. The equation as shown in (4.2) is used as a mathematical model in the development of the algorithm to predict the value of glucose level by using the prototype device.

$$y = -10.694x + 19.7291 \quad (4.12)$$

Where y represent the predicted glucose level and x represent output voltage from the prototype device.

4.4 Glucose prediction accuracy based on non-invasive prototype device (non-invasive) and glucose meter (invasive)

This experiment was conducted in order to determine the accuracy and data validation based on two different mathematical models used in the algorithms. The algorithms were tested using two different mathematical model in equations (4.1) and (4.2). Figure 4.9 shows the example of the algorithm code by using the mathematical model generated and the output terminal. Based on the code, the ***PDvoltage*** represents the input voltage from the sensor prop whereas ***g*** represents the glucose level. The input of the

microcontroller was in 10-bit analogue to digital converter (ADC), and since the equation uses voltage values, the ADC values therefore should be converted to voltage too.

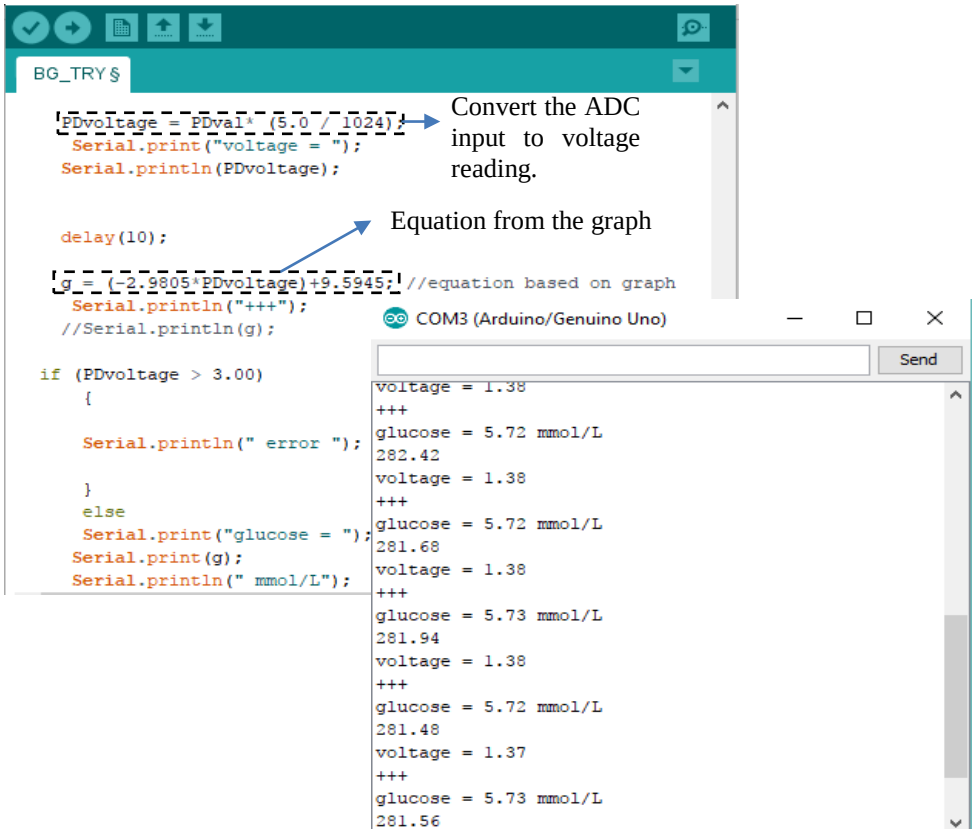


Figure 4.9: The glucose measurement algorithm coded in Arduino microcontroller

To determine the accuracy of the prototype device, the predicted measurements of non-invasive blood glucose prototype device were compared against the measurements of conventional invasive glucose meter. The percentage error between both the prototype device and invasive glucose meter readings were then calculated by using equation (4.3).

$$\text{Percentage Error (\%)} = \frac{Prototype(\ mmol/L) - GlucoseMet\ er\ (mmol/L)}{GlucoseMet\ er(mmol/L)} \text{x}100 \qquad (4.3)$$

The Clarke Error Grid Analysis (EGA) is the standard specifically used to determine the accuracy of glucose monitoring devices. The y-axis represents the values read by the prototype device, while the x-axis represents the values recorded by the conventional invasive blood glucose meter device, for the same patient, at the same time. Over hundred test points were taken on eighty patients to complete this experiments.

Three experiments were carried out for analysis the algorithm (developed using mathematical model of single subject samples). The first experiment used samples from the same single subject, the ones used in the mathematical model development. On the other hand, the second experiment used a different single subject than before. There are thirty points plotted for each graph as shown in Figure 4.10 and Figure 4.11. The third experiment involved multiple subjects as samples. Eighty subjects were used in this experiment and Figure 4.12 shows the data plotted. Besides, two experiments were conducted by using the algorithm (developed using mathematical model of multiple subjects). The first experiment used samples from a single subject to test the algorithms. Thirty data were recorded and a graph is plotted as in Figure 4.13. Lastly, the samples from multiple subjects were used in this experiment and Figure 4.14 shows the graph plotted with eighty data recorded during the experiment.

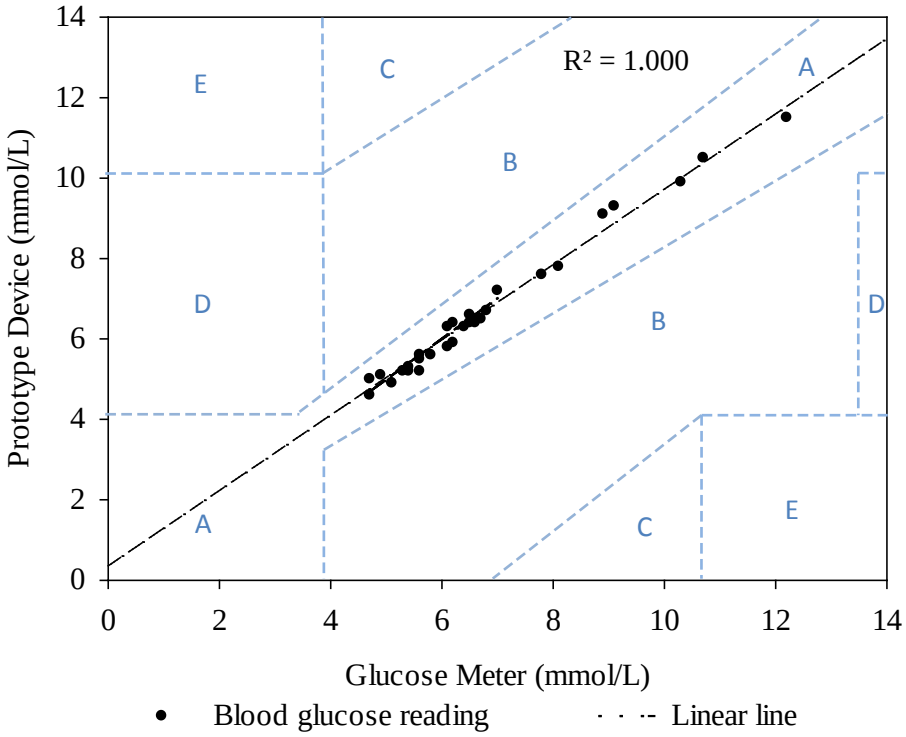


Figure 4.10: Clarke Error Grid of single subject algorithm tested by same single subject

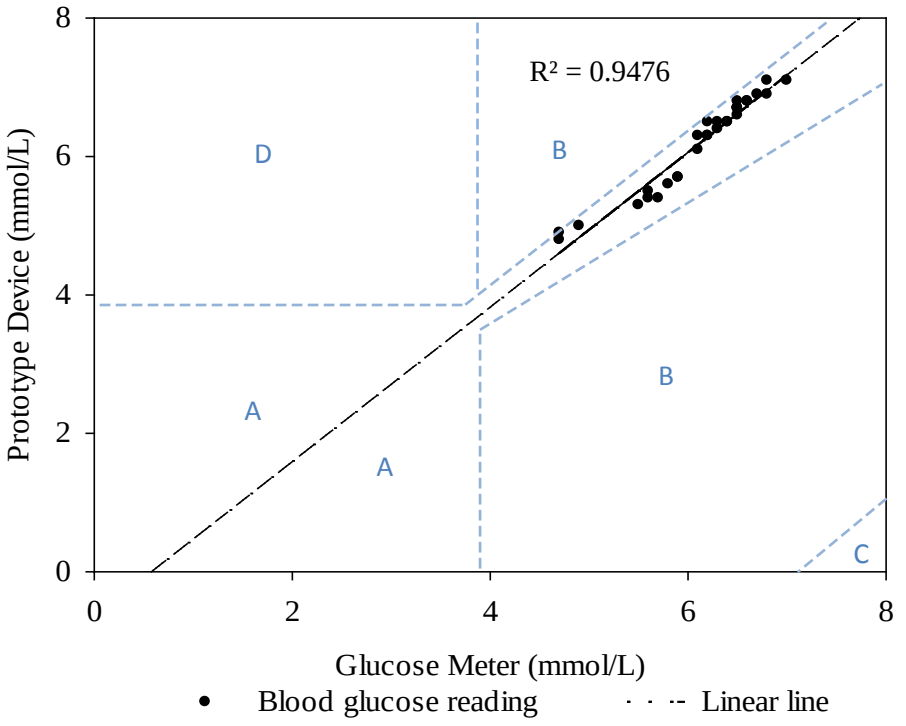


Figure 4.11: Clarke Error Grid of single subject algorithm tested by different single subject

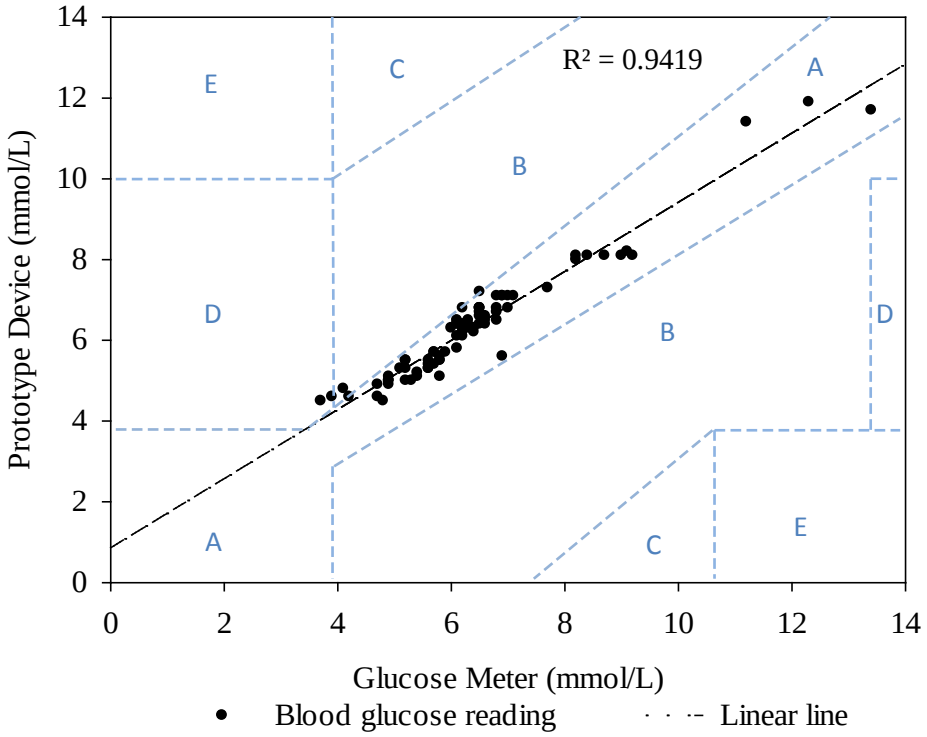


Figure 4.12: Clarke Error Grid of multiple subject algorithm tested by multiple subjects

There are three graphs were plotted based on algorithms which were derived from the mathematical model (developed using mathematical model of single subject). From the graphs, the highest value of correlation coefficient between measurements from the non-invasive prototype device and glucose meter is equal to 1.00 when the algorithm was tasted with the same subject used for mathematic model development. This was due to the minimum noise occurrence during the measurement. Since the same subject was used for mathematical model development and test, the subjects would possess similar physical characteristics such as skin tone and skin thickness. When different single-subject samples were used, the value of correlation coefficient was 0.9476; which shows a good relationship between measurements from the non-invasive prototype device and the reference glucose meter. In addition, based on the error grid in Figure 4.10 and

Figure 4.11, almost 100% off the data points lie in region *A* only. When the algorithms were tested by using multiple subjects as samples, around 90% of the data points lie in region *A*; whereas all the remaining points lie in region *B* as shown in Figure 4.12. However, the measurement of the prototype device and glucose meter show a good relationship based on the high value of the correlation coefficient, which was equal to 0.9419. The highest percentage error was 13.51% when the test was done using the samples from multiple subjects, as shown in the table in Appendix A. This occurs when the reading of blood glucose at a low level.

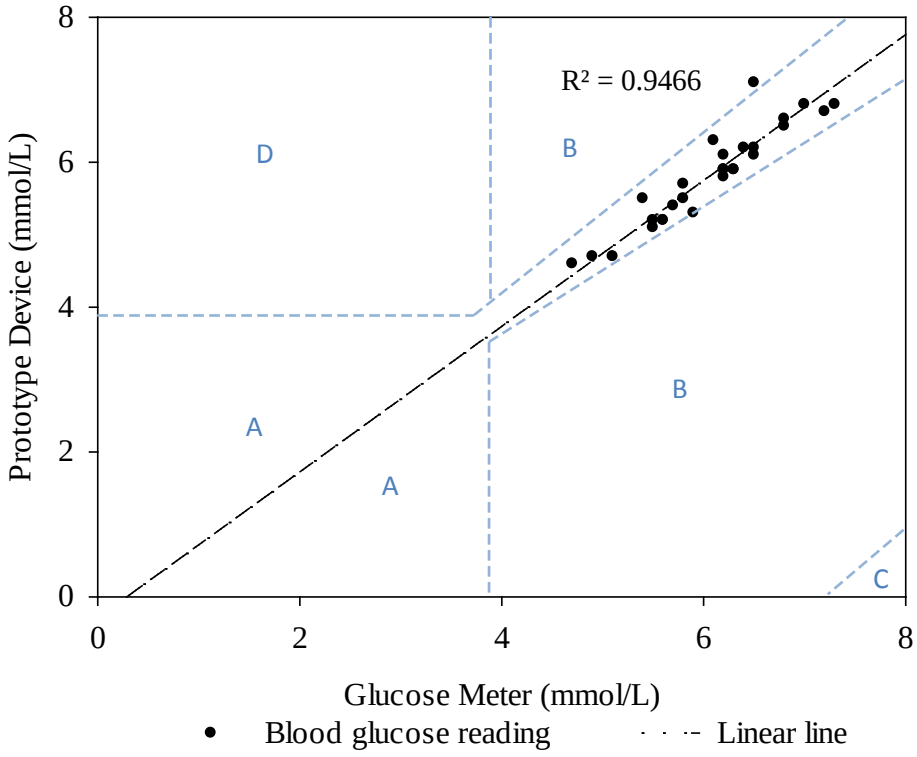


Figure 4.13: Clarke Error Grid of multiple subject algorithm tested by single subject

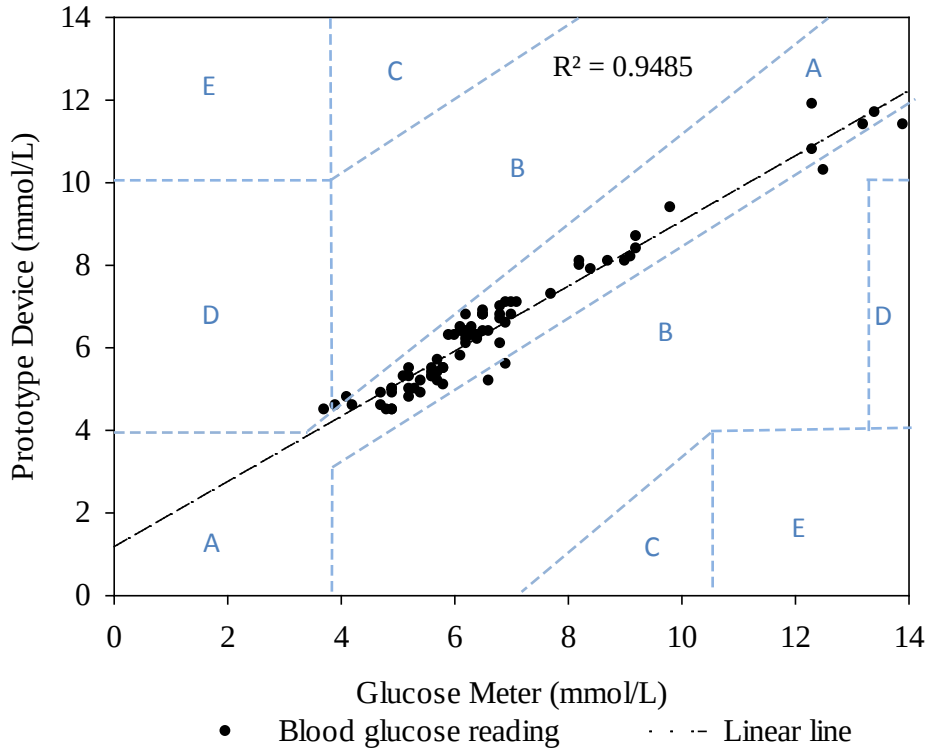


Figure 4.14: Clarke Error Grid of multiple subject algorithm tested by multiple subjects

The correlation coefficient values for the algorithm (developed using mathematical model of multiple subjects) were 0.9466; and 97% of the data points lie in region *A* and only one point lie in region *B* when tested by using samples from a single subject as shown in Figure 4.13. Meanwhile, when the algorithm was tested with samples from multiple subjects, the value of the correlation coefficient was equal to 0.9485, which was very good and around 10% of the data points lie in region *B*, nevertheless this would not lead to inappropriate treatment. Based on the percentage of errors, there was an error rate of up to 16.54% when the test done by using the samples from multiple subjects as shown in the table in Appendix B. Most the high reading error occurs when blood glucose readings are high and low as can see in the graph plotted and data table in appendix. Based on this situation, not every subject can use the same algorithm since each subject has different

physical features. One of the physical parameters that affect the glucose measurement is the earlobe tissue thickness since earlobe thickness could be different for each person. Tissue thickness determines the 'path length' of NIR, so a greater path length would result in lower NIR transmittance. Table 4.8 shows the summary of this experiments results.

Table 4.8: Summary of the experiments results for algorithms testing

Algorithm development	**Experiments (tested with)**	**Clarke Error Grid**	**Highest Percentage Error**
Single subject	Same single subject	Region A= 100%	5.35%
	Different single subject	Region A=100%	5.26%
	Multiple subjects	Region A= 97% Region B= 3%	13.51%
Multiple subjects	Single subject	Region A= 97% Region B= 3%	10.16%
	Multiple subjects	Region A= 90% Region B=10%	16.54%

4.5 NIR non-invasive blood glucose monitoring prototype system

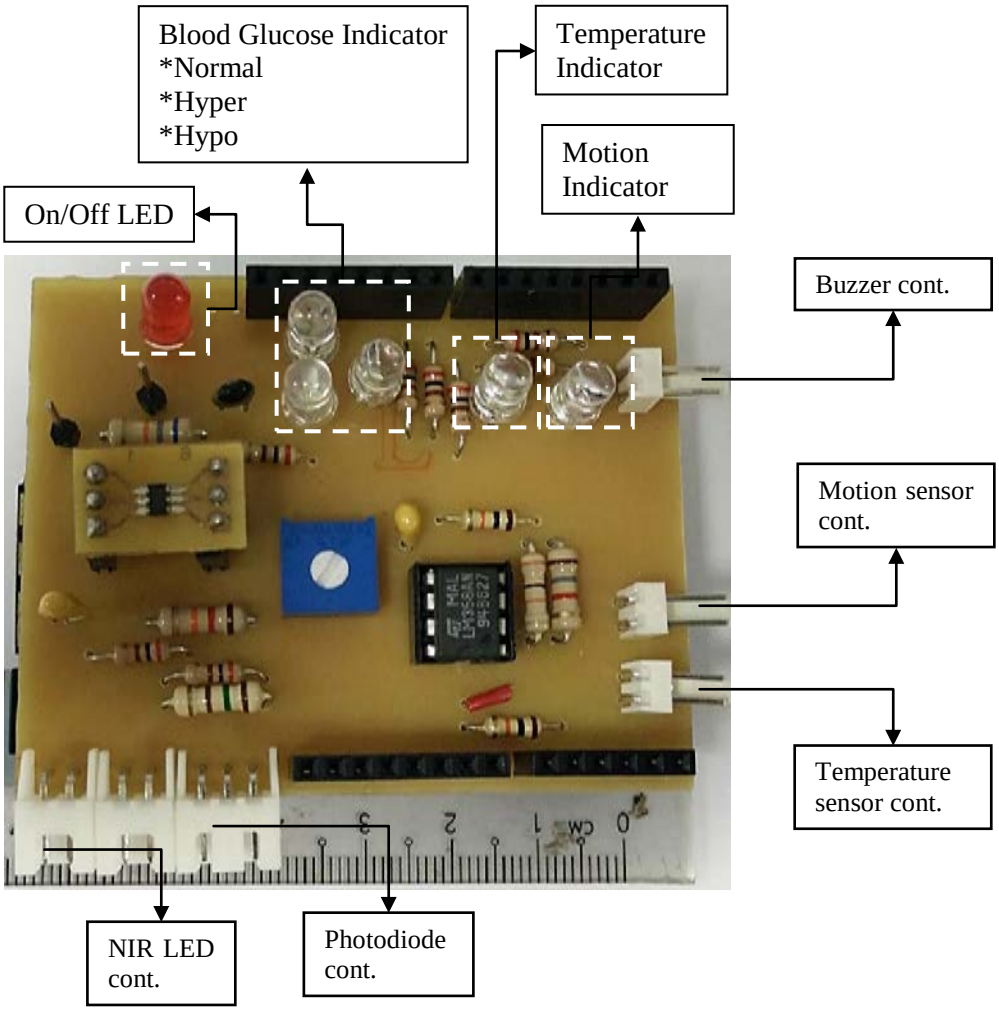


Figure 4.15: The prototype of PCB board of the NIR non-invasive blood glucose monitoring

Figure 4.15 shows the PCB board of the circuit used and it consists of several circuit combination of sensors circuit. The board was printed based on the Bluno Arduino processing board size as shown in Figure 4.16. From the presented figure, there are four sensors connector on it and LEDs are used as an indicator to indicate the user on several conditions. A microcontroller is coded to make the system function as needed.

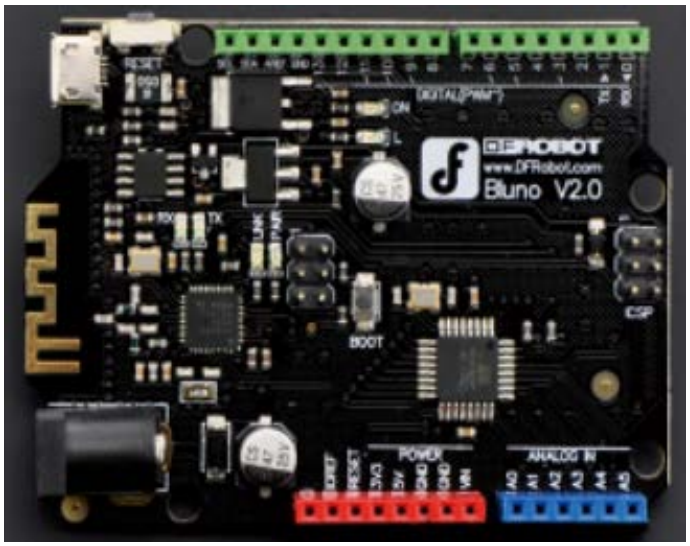


Figure 4.16: Bluno microcontroller processing board

Figure 4.17: The overall prototype system of NIR non-invasive blood glucose monitoring

The non-invasive blood glucose monitoring prototype device is shown as in Figure 4.17. The indicator LED will alert the user if there is an abnormal condition of blood glucose level. The abnormal glucose level occurs when the glucose reading is at

>4.0 mmol/L or >11.1 mmol/L. A different colour of LED is used to differentiate between hypoglycaemia and hyperglycaemia condition. The final glucose value can be read on the Android phone using Bluetooth connectivity. The Bluno microcontroller is built with Bluetooth Low Energy (BLE). This BLE technology is real-time low energy communication and it ideal for prototyping platform for both software and hardware developers. The Universal Asynchronous Receiver Transmitter (UART) of Bluno microcontroller is connected to the Bluetooth device.

4.5.1 Temperature sensor calibration

The normal skin temperature is taken as the reference and control parameter during the non-invasive blood glucose measurement. Since the NIR sensor can be affected by the higher temperature. Therefore, a temperature sensor is built in this project. The equation Steinhart-Hart as shown in (4.3) is used in the microcontroller code for temperature measurement.

$$\frac{1}{T} = a + b\ln(R) + c(\ln(R))^3 \tag{4.3}$$

Where T is the temperature (in Kelvin), R is the resistance at T (in ohms) and a, b, and c are the Steinhart-Hart parameter.

The temperature sensor response is being tested for calibration. The temperature sensor was placed on the skin of the target area. This measurement is recorded every five seconds for 90 seconds and the measurement is repeated for three times. The room temperature while the experiment is carried out is 22°C and the skin temperature is 33°C was measured by using a thermometer.

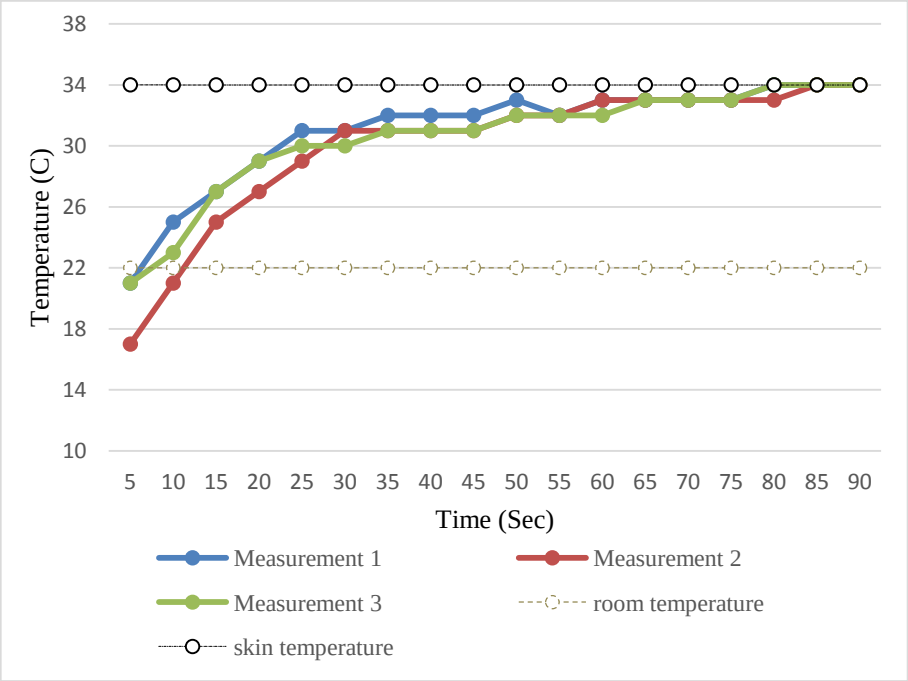


Figure 4.18: The measurement test of skin temperature

The graph of the measurement is shown on Figure 4.18. It shows that the maximum temperature measured is 33°C which is the skin temperature of the body. For all of the repeated measurements, it shows a similar response of the temperature increment and gets saturated when it reaches a stable skin temperature. The temperature sensor takes about 60 seconds to reach a stable temperature reading for these three experiments. The environmental temperature can influence the measurement.

4.5.2 Prototype device function

The prototype device function is explained in this subtopic. Based on Figure 4.19, it shows that the green LED motion indicator is light up when the movement of sensor prop is detector and blood glucose measurement is not occurring. When there is no sensor prop movement detected the blood glucose measurement is occurring. The white LED is light

up as in Figure 4.20 when the blood glucose is in the normal range (4.0 to 7.8 mmol/L). Figure 4.21 shows the orange LED is light up to indicate the hyperglycaemia condition (>7.9 mmol/L) and the blue LED as shown in Figure 4.22 is to indicate the hypoglycaemia condition (<4.0 mmol/L). Besides, the buzzer is turned on in the hyper and hypoglycaemia condition.

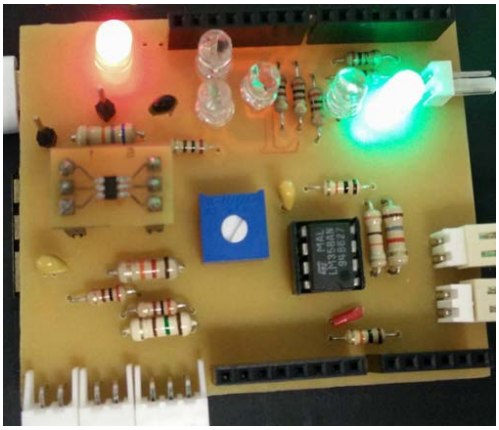


Figure 4.19: The motion indicator LED

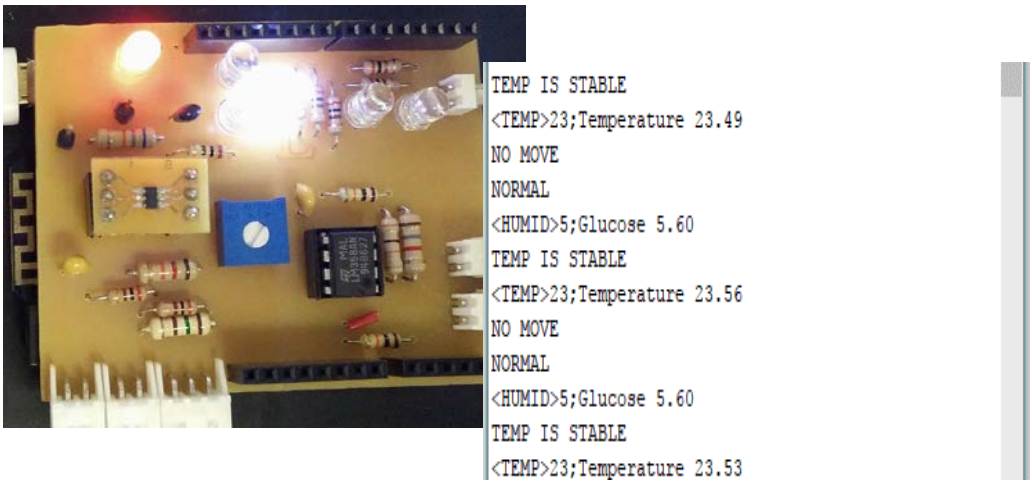


Figure 4.20: The normal blood glucose indicator LED

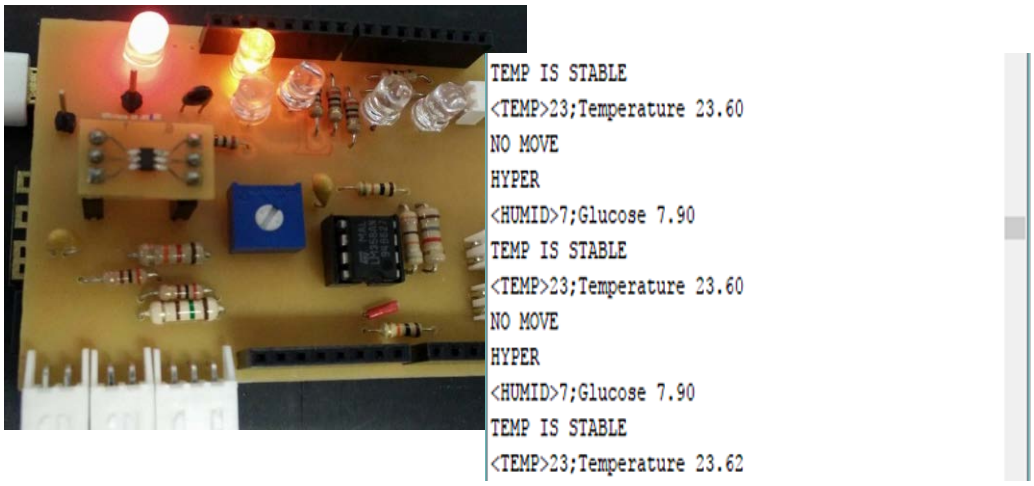


Figure 4.21: The hyperglycaemia blood glucose indicator LED

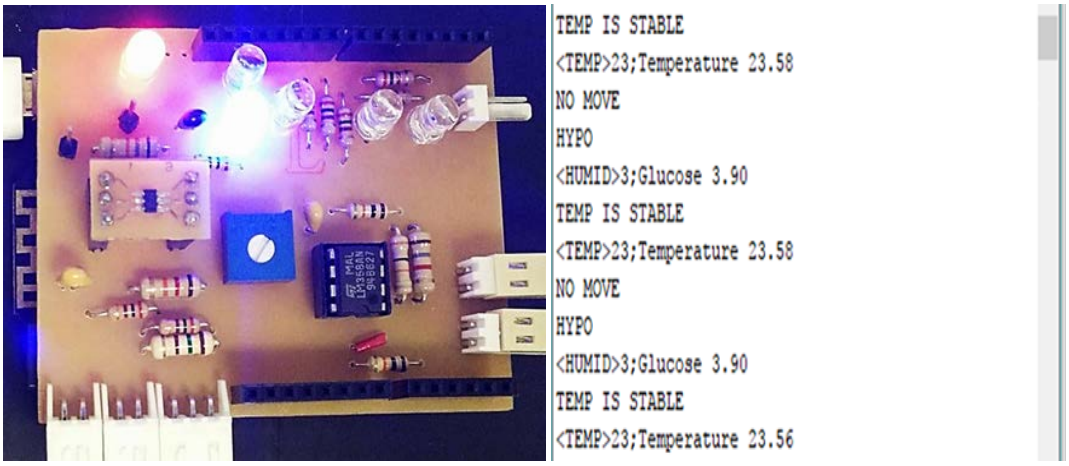


Figure 4.22: The hypoglycaemia blood glucose indicator LED

4.6 Summary

In this chapter, the analysis and steps of NIR non-invasive blood glucose monitoring development were presented. This non-invasive blood glucose prototype device can detect hyperglycaemia and hypoglycaemia blood glucose conditions without using blood sample or finger pricks. Although the device has difficulties in reading blood sugar level above 12 mmol/L, it can still warn the condition of hyperglycaemia to the user due to the fact that the maximum blood sugar level for the diabetic is greater than 11.1 mmol/L.

This device also can be easily adapted to provide continuous blood glucose monitoring. Besides, there are indicators that have been used on the prototype to alert the users about their blood glucose condition simply by reading the blood glucose measurement using Arduino application via Bluetooth connection. Based on the tests that have been conducted, every single user of this device needs their personal calibration since there is a differential in the physical conditions which affect the blood glucose measurements. Other than that, there are two additional sensors intended to control multiple parameters that will interfere in the measurement of NIR such as the skin temperature and sensor movement.

CHAPTER 5

CONCLUSION AND FUTURE WORK

5.1 Conclusion

In conclusion, this project aims for the development and analysis of near-infrared (NIR) spectroscopy technique for the non-invasive blood glucose monitoring system. Near-Infrared Spectroscopy was chosen due to its sensitivity, selectivity, low cost, and portability. The first objective of this thesis which is to develop the prototype of NIR non-invasive blood glucose monitoring system for personal use was answered. This development consists of a few steps. The first step is to choose the components which are the near-infrared LED and photodiode. The photodiode that was chosen must be able to cover the NIR wavelength used. Therefore the InGaAs type of photodiode was chosen since it is able to cover the NIR wavelengths from 700 nm to 2500 nm. The function of the prototype system is explained in Chapter 4. This prototype device has LED indicator to alert the user about an abnormal condition, and Android application with BLE connection to monitor the blood glucose measurement. This prototype development also involves the algorithm development for glucose measurement. Besides, the system of non-invasive blood glucose was also developed with an addition of temperature and motion parameters control for stability during the measurement. The developed device is low cost and is able to detect hypoglycaemia and hyperglycaemia conditions.

The second objective of this thesis is to investigate the response of different NIR wavelength lights to different concentration of glucose solutions. The wavelength of 1050 nm, 1200 nm, 1300 nm, 1450 nm, and 1550 nm were tested and analysed. In between

this five wavelength, 1450 nm was found to have a good response to the different glucose concentrations and the R^2 value obtained was 0.9559, which was the highest value compared to others. In addition, the transmittance and reflectance configuration was tested to find suitable sensor configurations for non-invasive blood glucose. The transmittance configuration showed the best response to different glucose concentrations. From the analyses, it is recommended to use the near-infrared wavelength of 1450 nm and sensor configuration of transmittance as a possible means to measure or predict glucose concentrations. Based on three target areas that were tested, the earlobe area shows the best consistency of voltage output besides being the best area used to place the sensor prop for blood glucose measurement compared to the other areas.

The last objective of this thesis is to investigate and validate the accuracy of near-infrared spectroscopy system developed in this project for non-invasive blood glucose monitoring. There are two algorithms which used two different mathematical models. The aforementioned mathematical models were developed by using samples from a single subject and multiple subjects. The Clarke Error Grid analysis was used to test the accuracy and the analysis shows that 100% of the data points lie in region *A* when the test was done by using single subject as a samples. Meanwhile, 90% of the data points lie in region *A* and the remaining in region *B* when samples from multiple subjects were used during the test. This shows that every user needs to have their own algorithms calibration to increase the accuracy of the NIR measurement. The results in Chapter 4 shows that this NIR blood glucose monitoring system development is a promising approach for non-invasive blood glucose. This development device presented here is only a proof of concept, showing a good correlation between NIR transmittance and blood glucose concentration. However, in such cases whereby an experimental device is not FDA approved, it could also be used

under consumer electronics device application, but not for any medical-related decision making.

5.2 Future work

Due to the lack of equipments, financial and available resources, some ideas of the improvement were not implemented. Presented here are suggestions and recommendations for the further development,future implementations or extensions of this project. The multi-sensor of NIR and the photodiode with a large active area can be used to improve the output of the voltage reading. The NIR LED and InGaAs photodiode with a large active area are relatively expensive compared to the normal infrared LED and photodiode. In addition, the combination of several techniques of non-invasive blood glucose can be applied for measurement comparison,accuracy improvement and environmental control such as humidity and atmospheric pressure which can be considered. For better data analysis, more test concentrations could be used as well as infrared light at many more different wavelengths. Lastly, the use of a better processor can improve the device in terms of processing speed, and data processing. Therefore, the calibration for different users can be done easily. This can simplify and save users' time in calibration processes.

REFERENCES

American Diabetes Association, 2014. Diagnosis and classification of diabetes mellitus. Diabetes care, 37(Supplement 1), pp.81-90.

Amir, O., Weinstein, D., Zilberman, S., Less, M., Perl-Treves, D., Primack, H., Weinstein, A., Gabis, E., Fikhte, B. and Karasik, A., 2007. Continuous Noninvasive Glucose Monitoring Technology Based on "Occlusion Spectroscopy." *Journal of Diabetes Science and Technology*, 1(4), pp.463–469.

Arimoto, H., Tarumi, M. and Yamada, Y., 2003. Instrumental Requirements for Non-Invasive Blood Glucose Measurement Using NIR Spectroscopy. *Optical Review*, 10(3), pp.161–165.

Aziz, N.A.M., Arsad, N., Menon, P.S., Laili, A.R., Laili, M.H. and Halim, A.A.A., 2015. Analysis of difference light sources for non-invasive aqueous glucose detection. *5th IEEE International Conference on Photonics (ICP)*, pp.150–152.

Baba, J.S., Cameron, B.D., Theru, S. and Coté, G.L., 2002. Effect of temperature, pH, and corneal birefringence on polarimetric glucose monitoring in the eye. *Journal of Biomedical Optics*, 7(3), pp.321–328.

Badugu, R., Lakowicz, J.R. and Geddes, C.D., 2003. A Glucose Sensing Contact Lens: A Non-Invasive Technique for Continuous Physiological Glucose Monitoring. *Journal of Fluorescence*, 13.

Bequette, B.W., 2010. Continuous glucose monitoring: real-time algorithms for calibration, filtering, and alarms. *Journal of Diabetes Science and Technology*, 4(2), pp.404–418.

BERNAMA, 2018. Malaysia mempunyai kadar diabetes tertinggi di Asis. Berita Harian, 25 July.

Blevins, T., Bode, B., Garg, S., Grunberger, G., Hirsch, I., Jovanovič, L., Nardacci, E., Orzeck, E., Roberts, V. and Tamborlane, W., 2010. Statement by the American Association of Clinical Endocrinologists Consensus Panel on Continuous Glucose Monitoring. *Endocrine Practice*, 16(5), pp.730–745.

Buda, R.A. and Addi, M.M., 2014. A Portable Non-Invasive Blood Glucose Monitoring Device. *IEEE Conference on Biomedical Engineering and Sciencess* (*IECBES)*, pp.8–10.

C8 MediSensors, 2012. C8 MediSensors Gains CE Mark Approval for the C8 MediSensors Optical Glucose Monitor(TM) System for People with Diabetes [onlinet] Available at: https://www.prnewswire.com/news-releases/c8-medisensors-gains-ce-mark-approval-for-the-c8-medisensors-optical-glucose-monitortm-system-for-people-with-diabetes-175821951.html [Accessed 28 Febuary 2016].

Caldwell, J., 2014. *TI Designs – Precision : Verified Design 1 MHz , Single-Supply , Photodiode Amplifier Reference Design*.

Chee, F. and Fernando, T., 2007. Closed-Loop Control of Blood Glucose. 1st ed. Springer Science & Business Media.

Cho, O.K., Kim, Y.O., Mitsumaki, H. and Kuwa, K., 2004. Noninvasive measurement of glucose by metabolic heat conformation method. *Clinical Chemistry*, 50(10), pp.1894–1898.

Chua, C.D., Gonzales, I.M., Manzano, E. and Manzano, M.C., 2014. Design and Fabrication of a Non-Invasive Blood Glucometer Using Paired Photo-Emitter and Detector Near-Infrared LEDs. *DLSU Research Congress*, pp.1-7.

Clark, L.C. and Lyons, C., 1962. Electrode systems for continuous monitoring in cardiovascular surgery. *Annals of the New York Academy of Sciences*, 102(1), pp.29–45.

Clarke, S.F. and Foster, J.R., 2012. A history of blood glucose meters and their role in self-monitoring of diabetes mellitus. *British Journal of Biomedical Science*, 69(2), pp.83–93.

Clarke, W.L., Cox, D., Gonder-Frederick, L.A., Carter, W. and Pohl, S.L., 1987. Evaluating clnical accuracy of systems for self-monitoring of blood glucose. *Diabetes Care*, 10(5), pp.622–628.

Cnoga, 2015. Combo Glucometer [online] Available at.: http://cnogacare.co/portfolio-item/combo-glucometer/ [Accessed on 8.Mac.2016].

Cunningham, D.D. and Stenke, J.A., 2010. *A series of monographs on analytical chemistry and its applications: In Vivo Glucose sensing.* New Jersey: John Wiley &Sons. Inc.

David Mendosa, 2005. History of Blood Glucose Meters. *Mendosa.com*. [online] Available at: http://www.mendosa.com/history.htm [Accessed on 8 December 2016].

Delpy, D.T., Cope, M., van der Zee, P., Arridge, S.R., Wray, S. and Wyatt, J.S., 1988. Estimation of optical pathlength through tissue from direct time of flight measurement. *Physics in Medicine and Biology*, 33(12), pp.1433–1442.

Dimeski, G., Jones, B.W., Tilley, V., Greenslade, M.N. and Russell, A.W., 2010. Glucose meters: evaluation of the new formulation measuring strips from Roche (Accu-Chek) and Abbott (MediSense). *Annals of Clinical Biochemistry*, 47(Pt 4), pp.358–365.

Ferrante do Amaral, C.E. and Wolf, B., 2008. Current development in non-invasive glucose monitoring. *Medical Engineering and Physics*, 30(5), pp.541–549.

Flacke, F., 2004. Pendra®- Non-invasive continuous glucose monitoring based on impedance spectroscopy [online] Available at: http://www.diabetes-symposium.org/index.php?menu=view&id=136#chart=4 [Accessed on 2 Mac 2016].

Freedom Meditech, 2013. ClearPath DS-120 [online] Available at: http://freedom-meditech.com/ [Accessed on 24.January 2016].

GlucoTrack, 2016. GlucoTrack gives you freedom from finger-pricking to help you manage your diabetes. [online] Available at:: http://www.glucotrack.com.au/glucotrack-product-features.html [Accessed 21 October.2016].

Guenter Gauglitz and Vo-Dinh, T., 2006. *Handbook of spectroscopy* 1st ed. Germany: John Wiley & Sons.

Guevara, E. and González, F.J., 2010. Joint optical-electrical technique for noninvasive glucose monitoring. *Revista Mexicana de Fisica*, 56(5), pp.430–434.

Guo, D.X., Shang, Y.Z., Peng, R., Yong, S.S. and Wang, X.A., 2015. Noninvasive Blood Glucose Measurement Based on NIR Spectrums and Double ANN Analysis. *Journal of Biosciences and Medicines*, pp.42–48.

Hamamatsu Photonics, 2015. *InGaAs PIN photodiodes*. Hamamatsu, Japan.

Harper, A. and Anderson, M.R., 2010. Electrochemical glucose sensors-developments using electrostatic assembly and carbon nanotubes for biosensor construction. *Sensors*, 10(9), pp.8248–8274.

Holt, R.I.G., Cockram, C., Flyvbjerg, A. and Goldstein, B.J., 2011. *Textbook of Diabetes* 4th ed. John Wiley & Sons.

Hotmartua, R., Pangestu, P.W., Zakaria, H. and Irawan, Y.S., 2015. Noninvasive Blood Glucose Detection Using Near Infrared Sensor. *5th International Conference on Electrical Engineering and Informatics 2015*, pp.687–692.

Jeon, K.J., Hwang, I.D., Hahn, S. and Yoon, G., 2006. Comparison between transmittance and reflectance measurements in glucose determination using near infrared spectroscopy. *Journal of Biomedical Optics*, 11(1), pp.1-7.

Kamboh, M.A. and Khan, A., 2013. *EDN Networks*: Non-invasive blood glucose monitoring using near-infrared spectroscopy. [online] Available at: http://www.edn.com/design/systems-design/4422840/Non-invasive-blood-glucose-monitoring-using-near-infrared-spectroscopy [Accessed on 22 Febuary 2016].

Keenan, D.B., Mastrototaro, J.J., Voskanyan, G. and Steil, G.M., 2009. Delays in minimally invasive continuous glucose monitoring devices: a review of current technology. *Journal of Diabetes Science and Technology (Online)*, 3(5), pp.1207–1214.

Khalil, O.S., 1999. Spectroscopic and clinical aspects of noninvasive glucose measurements. *Clinical Chemistry*, 45(2), pp.165–177.

Klonoff, D.C., 2005. Continuous Glucose Monitoring: Roadmap for 21st Century Diabetes Therapy. *Diabetes Care*, 28(5), pp.1231–1239.

Ko, J.B., Cho, O.K., Kim, Y.O. and Yasuda, K., 2004. Body Metabolism Provides a Foundation for Noninvasive Blood Glucose Monitoring. *Diabetes Care*, 27(5), pp.1211–1212.

Krushinitskaya, O., 2012. *Osmotic sensor for blood glucose monitoring applications*. Vestfold University College.

Lai, J.L., Huang, S.Y., Lin, R.S. and Tsai, S.C., 2016. Design a non-invasive near-infrared LED blood glucose sensor. *International Conference on Applied System Innovation (ICAS)*. pp.5–8.

Larin, K. V., Eledrisi, M.S., Motamedi, M. and Esenaliev, R.O., 2002. Noninvasive blood glucose monitoring with optical coherence tomography: A pilot study in human subjects. *Diabetes Care*, 25(12), pp.2263–2267.

Liu, R., Deng, B., Chen, W. and Xu, K., 2005. Next step of non-invasive glucose monitor by NIR technique from the well controlled measuring condition and results. *Optical and Quantum Electronics*, 37(13–15), pp.1305–1317.

MacKenzie, H.A., Ashton, H.S., Spiers, S., Shen, Y., Freeborn, S.S., Hannigan, J., Lindberg, J. and Rae, P., 1999. Advances in photoacoustic noninvasive glucose testing. *Clinical Chemistry*, 45(9), pp.1587–1595.

Maruo, K., Tsurugi, M., Chin, J., Ota, T., Arimoto, H., Yamada, Y., Tamura, M., Ishii, M. and Ozaki, Y., 2003. Noninvasive blood glucose assay using a newly developed near-infrared system. *Journal of Selected Topics in Quantum Electronics*, 9(2), pp.322–330.

Mclachlan, K. and Neal, D.O., 2007. The Use of the Continuous Glucose Monitors in Pregnancy. *Infusystems USA*, 4(4), pp.25–32.

Medgadget, 2007. Non-invasive Glucometer from OrSense Approved in Europe [online] Available at: https://www.medgadget.com/2007/06/noninvasive_glucometer_from_orsense_approved_in_europe.html [Accessed on 1 January 2016].

Menon, K.U., Hemachandran, D. and Abhishek, T.K., 2013. A survey on non-invasive blood glucose monitoring using NIR. *International Conference on Communication and Signal Processing (ICCSP)*, pp.1069–1072.

NADI, 2011. Burden of Diabetes in Malaysia. *Ministry of Health Malaysia.*

Narkhede, P., Dhalwar, S. and Karthikeyan, B., 2016. NIR based non-invasive blood glucose measurement. *Indian Journal of Science and Technology*, 9(41), pp.0–7.

Newman, J.D. and Turner, A.P.F., 2005. Home blood glucose biosensors: A commercial perspective. *Biosensors and Bioelectronics*, 20(12), pp.2435–2453.

Norman Colthup, 2012. *Introduction to Infrared and Raman Spectroscopy* 2nd ed., New York: Academic Press Inc.

Oliver, N.S., Toumazou, C., Cass, A.E.G. and Johnston, D.G., 2009. Glucose sensors: A review of current and emerging technology. *Diabetic Medicine*, 26(3), pp.197–210.

Osi Optoelectronics, 2009. *Photodiode Characteristics*.

Parab, J.S., Gad, R.S. and Naik, G.M., 2010. Noninvasive glucometer model using partial least square regression technique for human blood matrix. *Journal of Applied Physics*, 107(10).

Paul, B., Manuel, M.P. and Alex, Z.C., 2012. Design and Development of Non invasive Glucose Measurement System. *1st IEEE International Symposium Physics and Technology of Sensors, ISPTS*, pp.2–5.

Pickup, J.C., Hussain, F., Evans, N.D., Rolinski, O.J. and Birch, D.J.S., 2005. Fluorescence-based glucose sensors. *Biosensors and Bioelectronics*, 20(12), pp.2555–2565.

Poddar, R., Andrews, J.T., Shukla, P. and Sen, P., 2008. *Non-Invasive Glucose Monitoring Techniques: A review and current trends*, arXiv preprint arXiv:0810.5755.

Sato, J., Hirose, T. and Watada, H., 2012. Continuous glucose monitoring system: Is it really accurate , safe and clinically useful?. *Journal of Diabetes Investigation*, 3(3), pp.225–230.

Siesler, H.W., Ozaki, Y., Kawata, S. and Heise, H.M. 2008. Near-Infrared Spectroscopy: Principles, Instruments. John Wiley & Sons.

Sison. S., 2017. These Are the Best Colors to Wear According to Your Skin Tone. [online] Available at: https://www.preview.ph/fashion/best-colors-to-wear-according-to-your-skin-tone-a00191-20170524 [Accessed on 7 November 2018].

So, C.F., Choi, K.S., Wong, T.K.S. and Chung, J.W.Y., 2012. Recent advances in noninvasive glucose monitoring. *Medical Devices: Evidence and Research*, 5(1), pp.45–52.

Song, K., Ha, U., Park, S., Bae, J. and Yoo, H.J., 2015. An Impedance and Multi-Wavelength Blood Glucose Estimation. *IEEE Journal of Solid-State Circuits*, 50(4), pp.1025–1037.

Steinhart, J. S., Hart , S. R., 1968. Calibration curves for thermistors, Deep-Sea Research and Oceanographic, Volume 15, Issue 4, pp.497-503.

Tenhunen, J., Kopola, H. and Myllylä, R., 1998. Non-Invasive Glucose Measurement based on selective Near-Infrared Absorption: Requirements on Instrumentation and Spectral Range. *Measurement*, 24(3), pp.173–177.

Thorlabs, 2007. *Ultra Bright NIR LED Specifications and Documentation*. Thorlabs, USA.

Troy, T.L. and Thennadil, S.N., 2001. Optical properties of human skin in the near infrared wavelength range of 1000 to 2200 nm. *Journal of Biomedical Optics*, 6(2), pp.167-176.

Valery V. Tuchin, 2008. *Handbook of Optical Sensing of Glucose in Biological Fluids and Tissues*, New York: CRC Press.

Vashist, S.K., 2013. Continuous Glucose Monitoring Systems: A Review. *Diagnostics*, 3(4), pp.385–412.

Vashist, S.K., 2012. Non-invasive glucose monitoring technology in diabetes management: A review. *Analytica Chimica Acta*, 750, pp.16–27.

Wang, J., 2008. Electrochemical glucose biosensors. *Chemical Reviews*, 108(2), pp.814–825.

Wang, J., 2001. Glucose Biosensors: 40 Years of Advances and Challenges. *Electroanalysis*, 13(12), pp.983–988.

WHO, 1999. Definition, Diagnosis and Classification of Diabetes Mellitus and its Complication. Report of WHO Consultation. *World Health Organization*, pp.1-59.

WHO, 2016. Diabetes country profiles :Malaysia. *World Health Organization*.

WHO, 2016. Global Report on Diabetes. *World Health Organization*, 978, pp.4–65.

Yadav, J., Rani, A., Singh, V. and Murari, B.M., 2015. Comparative Study of Different Measurement Sites Using NIR Based Non-invasive Glucose Measurement System. *Proceedings Computer Science*, 70, pp.469-475. Jan 2015. Elsevier.

Yadav, J., Rani, A., Singh, V. and Murari, B.M., 2014. Near-infrared LED based non-invasive blood glucose sensor. *2014. International Conference on Signal Processing and Integrated Networks (SPIN)*, pp.591–594. February 2014. *IEEE.*

Yadav, J., Rani, A., Singh, V. and Murari, B.M., 2015. Prospects and limitations of non-invasive blood glucose monitoring using near-infrared spectroscopy. *Biomedical Signal Processing and Control*, 18, pp.214–227.

Yatim, N.N.M., Zain, Z.M., Jaafar, M.Z., Yusof, Z.M., Laili, A.R., Laili, M.H. and Hisham, M.H, 2014. Noninvasive glucose level determination using diffuse reflectance near infrared spectroscopy and chemometrics analysis based on in vitro sample and human skin. *IEEE Conference in Systems, Process and Control (ICSPC)*, pp. 30-35. *IEEE.*

Yoo, E.H. and Lee, S.Y., 2010. Glucose biosensors: an overview of use in clinical practice. *Sensors (Basel, Switzerland)*, 10(5), pp.4558–4576.

Zeng, B., Wang, W., Wang, N., Li, F., Zhai, F. and Hu, L., 2013. Noninvasive Blood Glucose Monitoring System Based on Distributed Multi-Sensors Information Fusion of Multi-Wavelength NIR. Engineering, 5(10), pp. 553-560.

Zhang, J., Hodge, W., Hutnick, C. and Wang, X., 2011. Noninvasive diagnostic devices for diabetes through measuring tear glucose. *Journal of Diabetes Science and Technology*, 5(1), pp.166–172.

APPENDIX A

The experiments data based on a single subject algorithm

Table A.1: The single subject algorithm tested with sample from same single subject used for mathematical model development

Readings	Times	Prototype Device (mmol/L)	Glucose Meter (mmol/L)	Error Percentage (%)
1	6:00 am	4.7	4.6	2.12
2	8:00 am	5.3	5.2	1.88
3	10:00 am	5.8	5.6	3.44
4	12:00 pm	6.5	6.6	1.53
5	2:00 pm	12.2	11.5	5.73
6	4:00 pm	6.2	6.4	3.22
7	6:00 pm	6.6	6.5	1.51
8	8:00 pm	6.4	6.3	1.56
9	10:00 pm	8.1	7.8	3.79
10	12:00 am	5.6	5.5	1.78
11	6:00 am	4.7	5.0	6.38
12	8:00 am	5.6	5.6	0.0
13	10:00 am	6.1	5.8	4.91
14	12:00 pm	10.7	10.5	1.86
15	2:00 pm	7.0	7.2	2.85
16	4:00 pm	6.1	6.3	3.27
17	6:00 pm	6.5	6.4	1.53
18	8:00 pm	9.1	9.3	2.19
19	10:00 pm	6.6	6.4	3.03
20	12:00 am	6.8	6.7	1.47
21	6:00 am	4.9	5.1	4.08
22	8:00 am	5.4	5.3	1.85
23	10:00 am	5.6	5.2	5.35
24	12:00 pm	5.4	5.2	3.07
25	2:00 pm	10.3	9.9	3.88
26	4:00 pm	7.8	7.6	2.56

27	6:00 pm	6.2	5.9	4.83
28	8:00 pm	5.1	4.9	3.92
29	10:00 pm	8.9	9.1	2.24
30	12:00 am	6.7	6.5	2.98

Table A.2: The single subject algorithm tested with sample from different single subject used for mathematical model development

Readings	**Times**	**Prototype Device (mmol/L)**	**Glucose Meter (mmol/L)**	**Percentage Error (%)**
1	6:00 am	4.7	4.9	4.25
2	8:00 am	5.8	5.6	3.44
3	10:00 am	5.6	5.4	3.57
4	12:00 pm	6.8	6.9	1.47
5	2:00 pm	7.0	7.1	1.42
6	4:00 pm	6.2	6.3	1.61
7	6:00 pm	6.5	6.7	3.07
8	8:00 pm	6.7	6.9	2.98
9	10:00 pm	6.2	6.5	4.83
10	12:00 am	5.5	5.3	4.83
11	6:00 am	4.9	5.0	2.04
12	8:00 am	6.1	6.1	0.0
13	10:00 am	6.5	6.7	3.07
14	12:00 pm	6.4	6.5	1.56
15	2:00 pm	5.9	5.7	3.38
16	4:00 pm	6.2	6.3	1.61
17	6:00 pm	5.7	5.4	5.26
18	8:00 pm	5.9	5.7	3.38
19	10:00 pm	6.3	6.5	3.17
20	12:00 am	6.5	6.6	1.53
21	6:00 am	4.7	4.8	2.12
22	8:00 am	5.6	5.5	1.78
23	10:00 am	6.1	6.3	3.27
24	12:00 pm	6.5	6.8	4.61
25	2:00 pm	6.8	7.1	4.41
26	4:00 pm	6.4	6.5	1.56

27	6:00 pm	6.6	6.8	3.03
28	8:00 pm	6.5	6.7	3.07
29	10:00 pm	6.3	6.4	1.58
30	12:00 am	6.6	6.8	3.03

Table A.3: The single subject algorithm tested with sample from multiple subjects

Samples	Prototype Device (mmol/L)	Glucose Meter (mmol/L)	Percentage Error (%)
1	4.7	4.9	4.25
2	5.8	5.6	3.45
3	4.9	5.1	4.08
4	5.6	5.3	5.35
5	5.2	5.3	1.92
6	6.8	7.1	4.41
7	6.9	7.1	2.89
8	6.5	6.8	4.61
9	7.0	6.8	2.85
10	6.2	6.2	0.0
11	6.8	6.5	4.41
12	6.6	6.5	1.51
13	7.0	7.1	1.42
14	6.5	6.7	3.07
15	4.9	5.0	2.04
16	5.4	5.2	3.7
17	6.2	6.3	1.61
18	6.1	6.1	0.0
19	6.5	6.8	4.61
20	6.4	6.2	3.12
21	5.9	5.7	3.38
22	6.2	6.4	3.22
23	5.7	5.4	5.26
24	6.9	6.1	11.59
25	6.3	6.5	3.17
26	6.5	6.6	1.53
27	4.9	5.0	2.04

28	5.6	5.5	1.78
29	6.1	6.4	4.91
30	6.3	6.4	1.58
31	6.5	6.4	1.56
32	6.6	6.6	0.0
33	6.2	6.8	9.67
34	6.5	7.2	10.76
35	5.8	5.5	5.17
36	5.7	5.7	0
37	5.2	5.5	5.76
38	5.7	5.7	0.0
39	6.3	6.3	0.0
40	5.8	5.1	12.06
41	6.1	5.8	4.91
42	6.6	6.4	3.03
43	5.6	5.3	5.35
44	5.2	5.0	3.84
45	4.9	4.9	0.0
46	8.2	8.0	2.43
47	7.7	7.3	3.59
48	4.2	4.6	9.52
49	3.7	4.2	13.51
50	4.8	4.5	6.25
51	8.4	8.1	3.57
52	9.2	8.1	10.98
53	11.2	11.4	1.78
54	3.9	4.3	10.25
55	6.1	6.4	4.91
56	9.1	8.2	9.89
57	5.4	5.1	5.55
58	6.1	6.5	6.55
59	8.2	8.1	1.21
60	5.6	5.4	3.57
61	4.9	5.0	2.04
62	6.8	6.8	0.0
63	6.4	6.3	5.16

64	7.1	7.1	0.0
65	13.4	11.7	12.68
66	6.4	6.3	1.56
67	4.1	4.6	12.19
68	5.3	5.0	5.66
69	8.7	8.1	6.89
70	5.1	5.3	3.92
71	6.9	7.1	2.89
72	6.8	6.7	1.47
73	5.8	5.5	5.17
74	6.2	6.1	1.61
75	6.5	6.8	4.61
76	6.0	6.3	5.0
77	12.3	11.9	3.25
78	9.0	8.1	10.0
79	4.7	4.6	2.12
80	5.2	5.5	5.76

APPENDIX B

The experiments data based on a single subject algorithm

Table B.1: The multiple subjects algorithm tested with sample from single subject

Readings	Times	Prototype Device (mmol/L)	Glucose Meter (mmol/L)	Percentage Error (%)
1	6:00 am	4.7	4.6	2.12
2	8:00 am	5.8	5.7	1.72
3	10:00 am	5.6	5.2	7.14
4	12:00 pm	6.8	6.5	4.41
5	2:00 pm	7.0	6.8	2.85
6	4:00 pm	6.2	5.9	4.83
7	6:00 pm	6.8	6.6	2.94
8	8:00 pm	6.5	6.2	4.61
9	10:00 pm	6.2	6.1	1.61
10	12:00 am	5.5	5.1	7.27
11	6:00 am	4.9	4.7	4.08
12	8:00 am	5.8	5.5	5.17
13	10:00 am	6.1	6.3	3.27
14	12:00 pm	6.4	6.2	3.12
15	2:00 pm	6.2	5.9	4.83
16	4:00 pm	5.9	5.3	10.16
17	6:00 pm	5.7	5.4	5.26
18	8:00 pm	5.5	5.2	5.45
19	10:00 pm	6.3	5.9	6.34
20	12:00 am	6.5	6.1	6.15
21	6:00 am	5.1	4.7	7.84
22	8:00 am	5.6	5.2	7.14
23	10:00 am	6.2	5.8	6.45
24	12:00 pm	9.7	9.1	6.18
25	2:00 pm	7.2	6.7	6.94
26	4:00 pm	6.3	5.9	6.34
27	6:00 pm	5.4	5.5	1.86
28	8:00 pm	6.5	7.1	9.23
29	10:00 pm	9.5	9.8	3.15

30	12:00 am	7.3	6.8	6.84

Table B.2: The multiple subjects algorithm tested with sample from multiple subjects

Samples	Prototype Device (mmol/L)	Glucose Meter (mmol/L)	Percentage Error (%)
1	4.7	4.9	4.25
2	5.8	5.5	5.17
3	4.9	4.5	8.16
4	5.6	5.3	5.35
5	5.2	5.3	1.92
6	6.8	7.0	2.94
7	6.9	6.6	4.34
8	6.5	6.8	4.61
9	7.0	6.8	2.85
10	6.2	6.2	0.0
11	6.8	6.1	10.29
12	13.9	11.6	16.54
13	7.0	7.1	1.42
14	6.5	6.9	6.15
15	4.9	4.5	8.16
16	5.4	5.2	3.7
17	6.2	6.3	1.61
18	9.2	8.4	8.69
19	6.5	6.8	4.61
20	6.4	6.2	4.12
21	5.9	6.3	6.77
22	6.2	6.4	3.22
23	5.7	5.4	5.26
24	6.9	5.9	14.49
25	6.3	6.5	3.17
26	12.5	10.3	17.6
27	4.9	5.0	2.04
28	5.6	5.5	1.78
29	12.3	10.8	12.19
30	6.3	6.4	1.58

31	6.5	6.4	1.53
32	6.6	5.2	2.12
33	6.2	6.8	9.67
34	9.8	9.4	4.08
35	5.8	5.5	5.17
36	5.7	5.7	0.0
37	5.2	4.8	7.69
38	5.7	5.2	8.77
39	6.3	6.3	0.0
40	5.8	5.1	12.06
41	6.1	5.8	4.91
42	6.6	6.4	3.03
43	5.6	5.3	5.35
44	5.2	5.0	3.84
45	4.9	4.9	0.0
46	8.2	8.0	2.43
47	7.7	7.3	5.19
48	4.2	4.6	9.52
49	3.7	4.2	13.51
50	4.8	4.5	6.25
51	8.4	7.9	5.95
52	9.2	8.7	5.43
53	13.2	11.4	13.63
54	3.9	4.4	12.82
55	6.1	6.4	4.91
56	9.1	8.2	9.89
57	5.4	4.9	9.25
58	6.1	6.5	6.55
59	8.2	8.1	1.21
60	5.6	5.4	3.57
61	4.9	5	2.04
62	6.8	6.8	0.0
63	6.4	6.3	1.56
64	7.1	7.1	0.0
65	13.4	11.7	12.68
66	6.4	6.3	1.56

67	4.1	4.6	12.19
68	5.3	5.0	5.66
69	8.7	8.1	6.89
70	5.1	5.3	3.92
71	6.9	7.1	2.89
72	6.8	6.7	1.47
73	5.8	5.5	5.17
74	6.2	6.1	1.61
75	6.5	6.8	4.61
76	6.0	6.3	5.0
77	12.3	11.9	3.25
78	9.0	8.1	10.0
79	4.7	4.6	2.12
80	5.2	5.5	5.76

APPENDIX C

Near-infrared LED Data Sheet

Part 1. Introduction: LED1450E Ultra Bright NIR LED

The LED1450E emits light with a spectral output centered at 1450 nm. This LED is composed of heterostructures (HS) grown on an InGaAsP substrate. The diode is encapsulated in a round clear epoxy casing with a 5 mm diameter.

Part 2. Specifications for an LED1450E

2.1. Electrical Specifications

	Typical	Maximum Ratings
Power Dissipation		120 mW
Reverse Voltage		5.0 V
DC Forward Current		100 mA
Forward Voltage @ 20 mA	1.2 V	1.5 V
Reverse Current V_r = -5 V		10 µA
Pulsed Current (1 ms pulse with 10% duty cycle)		1000 mA
Operating Temperature		-30 °C to 85 °C
Storage temperature Range		-30 °C to 100 °C

Note: All maximum measurements specified are at 25 °C.

2.2. Optical Specifications

	Typical
Center Wavelength	1450 nm (±50 nm)
FWHM	100 nm (±10 nm)
Half Viewing Angle	15° (±3°)
Forward Optical Power	1.9 mW @ 20 mA
Total Optical Power	2.0 mW @ 20 mA (±0.3 mW)
Rise (Fall) Time	10 (10) ns

2.3. Soldering Specifications

	Conditions
Manual Soldering	295 °C ± 5 °C , for less than 3 seconds
Wave Soldering	260 °C ± 5 °C , for less than 5 seconds
Reflow Soldering	**Preheating:** 70 °C to 80 °C , for 30 seconds **Soldering:** 245 °C ± 5 °C , for less than 5 seconds

2.4. Cleaning Solvents

Solvent	Ethyl Alcohol	Isopropyl Alcohol	Propanol	Acetone	Chloroseen	Tricloroethylene	MKS
Approved	Yes	Yes	Yes	No	No	No	No

2.5. Physical Specifications

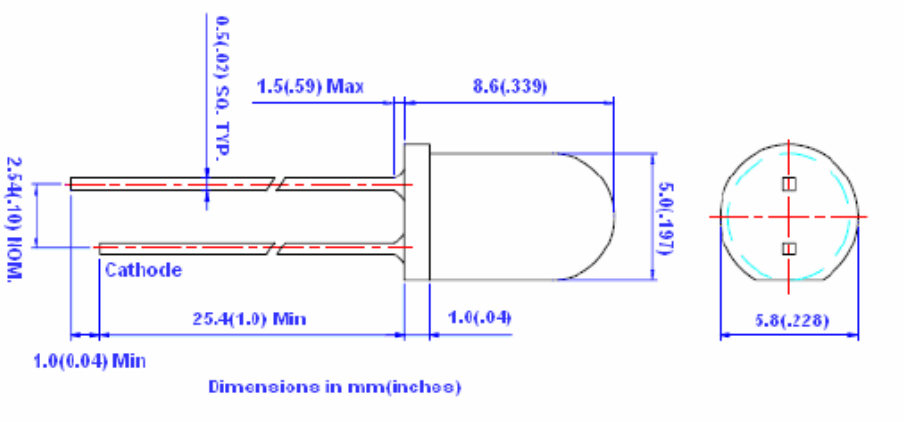
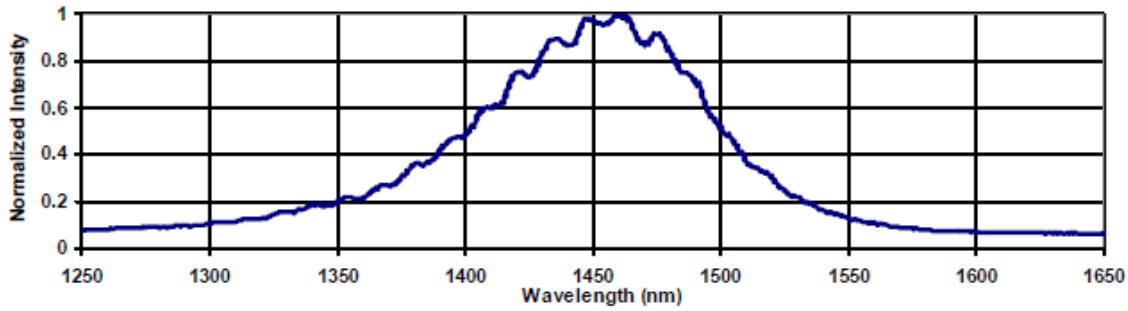
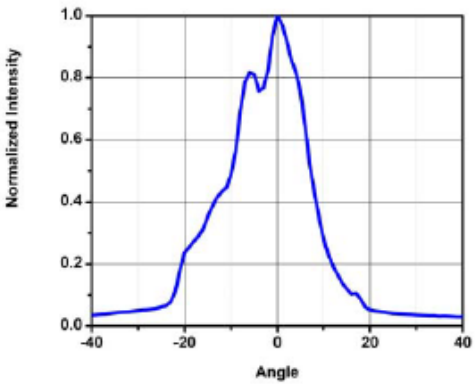


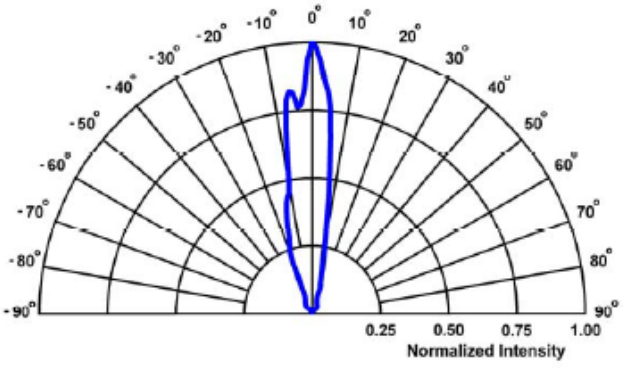
Figure 1: LED1450E. The cathode is the short lead and the anode is the long lead.

2.6. Typical Spectral Intensity Distribution



2.7. Typical Radial Intensity Distribution



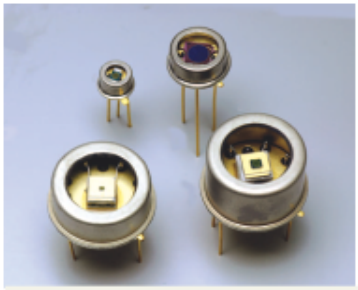


APPENDIX D

Indium Gallium Arsenide Photodiode Data Sheet

HAMAMATSU
PHOTON IS OUR BUSINESS

InGaAs PIN photodiodes



G12180 series

Photosensitive area from ϕ0.3 mm to ϕ5 mm

InGaAs PIN photodiodes have large shunt resistance and feature very low noise. Hamamatsu provides various types of InGaAs PIN photodiodes with photosensitive area from ϕ0.3 mm to ϕ5 mm.

Features

- Low noise, low dark current
- Low terminal capacitance
- Large photosensitive area
- Various photosensitive area sizes available

Applications

- Laser monitors
- Optical power meters
- Laser diode life test
- NIR (near infrared) photometry
- Optical communications

Options

Amplifier for InGaAs PIN photodiode	C4159-03
Heatsink for one-stage TE-cooled type	A3179
Heatsink for two-stage TE-cooled type	A3179-01
Temperature controller for TE-cooler type	C1103-04

Specifications/Absolute maximum ratings

Type no.	Dimensional outline/ Window material*1	Package	Cooling	Photosensitive area (mm)	Absolute maximum ratings: Thermistor power dissipation (mW)	TE-cooler allowable current (A)	TE-cooler allowable voltage (V)	Reverse voltage (V)	Operating temperature*2 (°C)	Storage temperature*2 (°C)	Soldering conditions
G12180-003A	(1)/K	TO-18	Non-cooled	ϕ0.3	-	-	-	20	-40 to +100	-55 to +125	260 °C or less, within 10 s
G12180-005A				ϕ0.5							
G12180-010A				ϕ1				10			
G12180-020A	(2)/K	TO-5		ϕ2				5			
G12180-030A				ϕ3							
G12180-050A	(3)/K	TO-8		ϕ5				2			
G12180-110A	(4)/K		One-stage TE-cooled	ϕ1	0.2	1.5	1	5	-40 to +70	-55 to +85	
G12180-120A				ϕ2							
G12180-130A				ϕ3							
G12180-150A				ϕ5				2			
G12180-210A	(5)/K		Two-stage TE-cooled	ϕ1		1	1.2	5			
G12180-220A				ϕ2							
G12180-230A				ϕ3							
G12180-250A				ϕ5				2			

*1: K: borosilicate glass with anti-reflective coating (optimized for 1.55 µm peak)
*2: No dew condensation
When there is a temperature difference between a product and the surrounding area in high humidity environment, dew condensation may occur on the product surface. Dew condensation on the product may cause deterioration in characteristics and reliability.
Note: Exceeding the absolute maximum ratings even momentarily may cause a drop in product quality. Always be sure to use the product within the absolute maximum ratings.

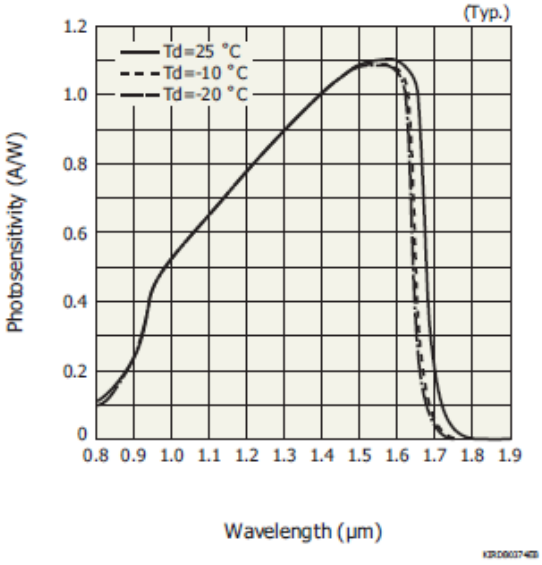
The G12180 series may be damaged by electrostatic discharge, etc. Be careful when using the G12180 series.

Electrical and optical characteristics (Typ. unless otherwise noted)

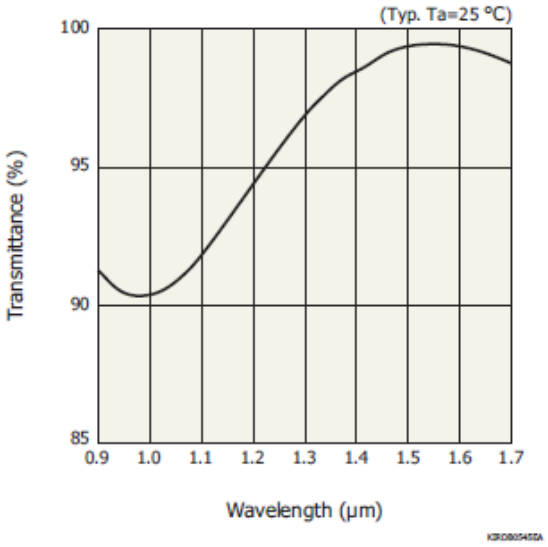
Type no.	Measurement condition: Element temperature (°C)	Spectral response range λ (μm)	Peak sensitivity wavelength λp (μm)	Photosensitivity S: 1.3 μm Min. (A/W)	Photosensitivity S: 1.3 μm Typ. (A/W)	Photosensitivity S: λ=λp Min. (A/W)	Photosensitivity S: λ=λp Typ. (A/W)	Dark current ID VR=1 V Typ. (nA)	Dark current ID VR=1 V Max. (nA)	Temperature coefficient of dark current ΔTID VR=1 V	Cutoff frequency fc VR=1 V RL=50 Ω Min. (MHz)	Cutoff frequency fc VR=1 V RL=50 Ω Typ. (MHz)	Terminal capacitance Ct VR=1 V f=1 MHz Typ. (pF)	Terminal capacitance Ct VR=1 V f=1 MHz Max. (pF)	Shunt resistance Rsh VR=10 mV Min. (MΩ)	Shunt resistance Rsh VR=10 mV Typ. (MΩ)	Detectivity D* λ=λp Min. (cm·Hz^1/2/W)	Detectivity D* λ=λp Typ. (cm·Hz^1/2/W)	Noise equivalent power NEP λ=λp Typ. (W/Hz^1/2)	Noise equivalent power NEP λ=λp Max. (W/Hz^1/2)
G12180-003A	25	0.9 to 1.7	1.55	0.8	0.9	0.9	1.1	0.1*3	0.5*3	1.09	450*4	600*4	5*5	7.5*5	200	1000	2.4×10^{12}	6.3×10^{12}	4.2×10^{-15}	1.2×10^{-14}
G12180-005A								0.15*3	0.75*3		160*4	200*4	15*5	20*5	80	400			7.0×10^{-15}	1.9×10^{-14}
G12180-010A								0.8*3	4*3		25*4	60*4	55*5	120*5	25	125			1.4×10^{-14}	3.8×10^{-14}
G12180-020A								1.5	7.5		4	13	250	800	6.5	30			2.8×10^{-14}	7.5×10^{-14}
G12180-030A								2.5	12.5		2.5	7	450	1500	4	20			4.4×10^{-14}	1.1×10^{-13}
G12180-050A								5	25		0.5	3	1000	7000	1.3	6.5			7.0×10^{-14}	1.9×10^{-13}
G12180-110A	-10	0.9 to 1.67						0.02	0.1		20	40	75	140	750	3750	1.6×10^{13}	4.4×10^{13}	2.0×10^{-15}	5.4×10^{-15}
G12180-120A								0.1	0.5		4	13	250	800	200	900			4.0×10^{-15}	1.1×10^{-14}
G12180-130A								0.15	0.8		2.5	7	450	1500	120	600			4.9×10^{-15}	1.4×10^{-14}
G12180-150A								0.33	1.67		0.5	3	1000	7000	40	200			8.6×10^{-15}	2.3×10^{-14}
G12180-210A	-20	0.9 to 1.65						0.01	0.06		20	40	75	140	1750	8750	2.6×10^{13}	6.7×10^{13}	1.3×10^{-15}	3.5×10^{-15}
G12180-220A								0.04	0.2		4	13	250	800	500	2000			2.7×10^{-15}	6.5×10^{-15}
G12180-230A								0.07	0.35		2.5	7	450	1500	280	1400			3.2×10^{-15}	8.7×10^{-15}
G12180-250A								0.15	0.75		0.5	3	1000	7000	90	500			5.3×10^{-15}	1.5×10^{-14}

*3: VR=5 V
*4: VR=5 V, RL=50 Ω, -3 dB
*5: VR=5 V, f=1 MHz

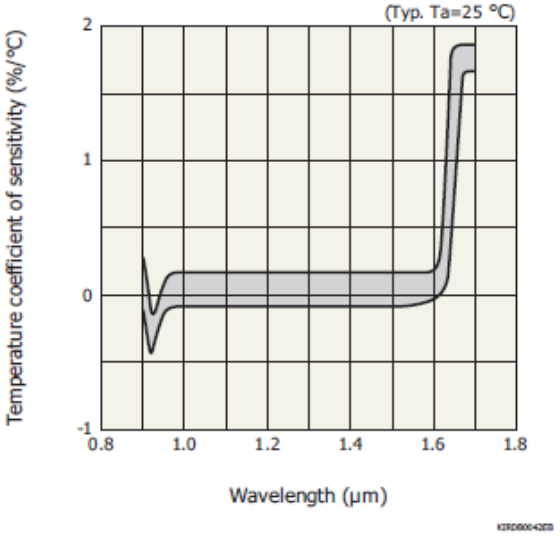
Spectral response



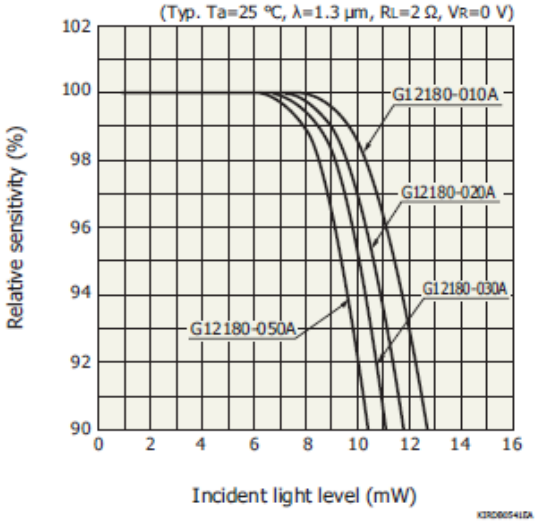
Spectral transmittance characteristics of window material



Photosensitivity temperature characteristics

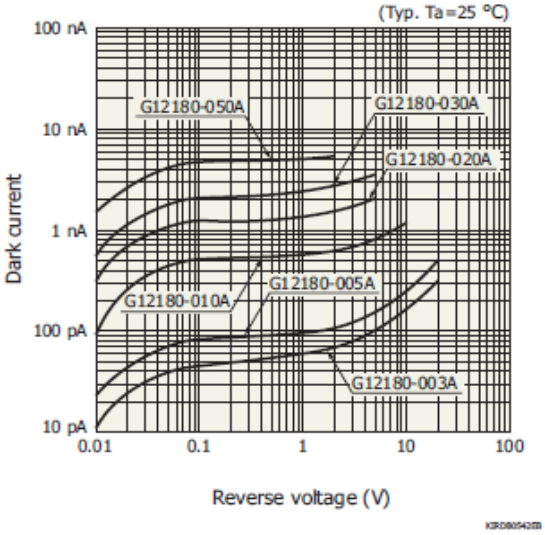


Linearity

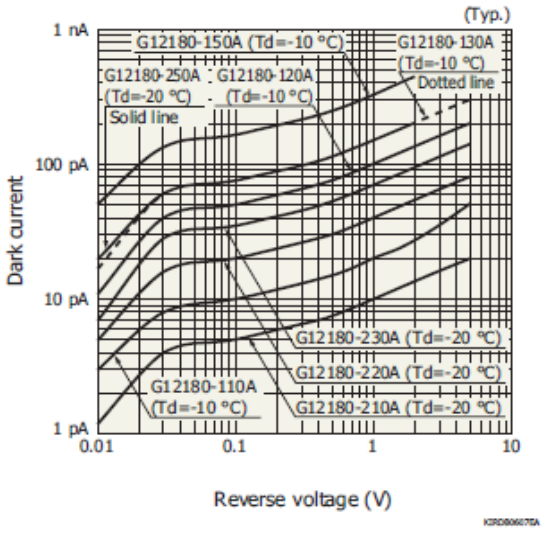


Dark current vs. reverse voltage

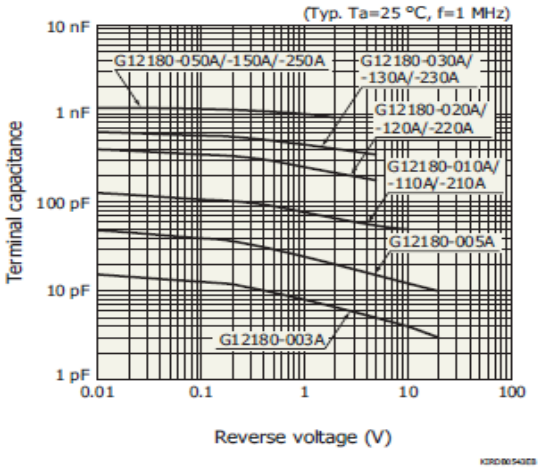
Non-cooled type



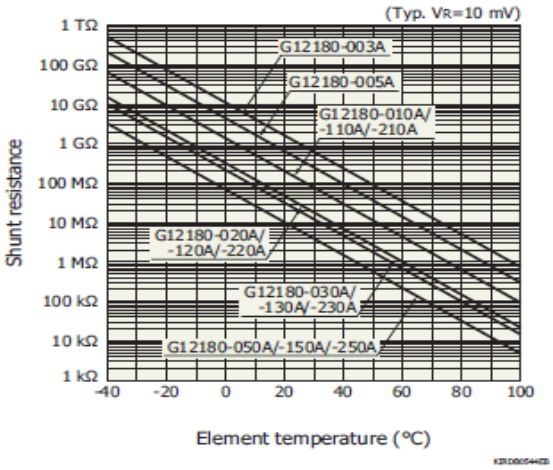
TE-cooled type



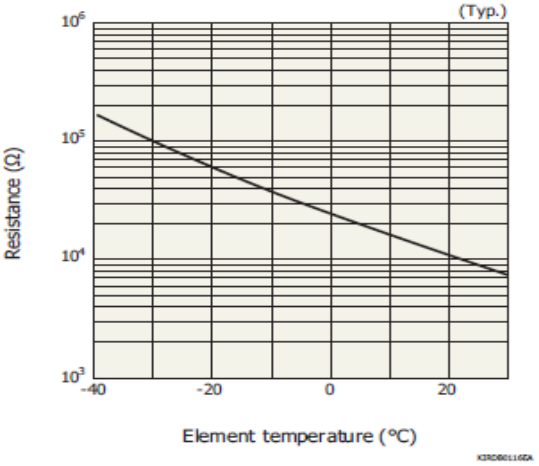
Terminal capacitance vs. reverse voltage



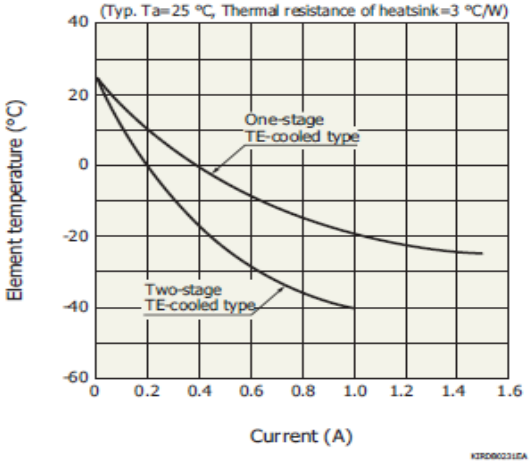
Shunt resistance vs. element temperature



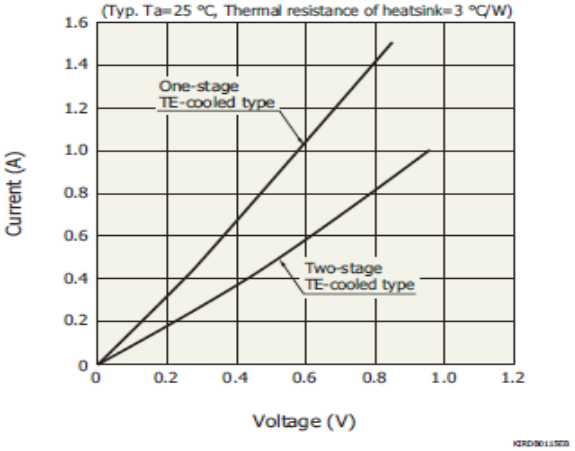
Thermistor temperature characteristics



Cooling characteristics of TE-cooler

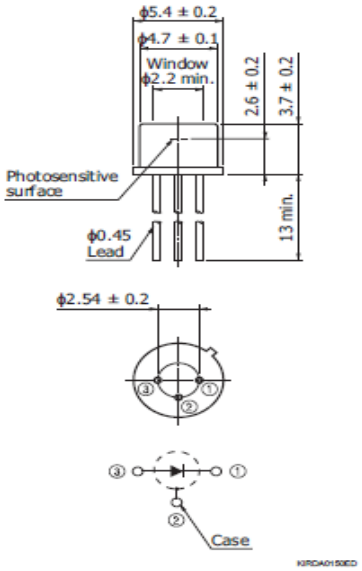


Current vs. voltage (TE-cooler)



Dimensional outlines (unit: mm)

(1) G12180-003A/-005A/-010A



APPENDIX E

Bluno Microcontroller Board

Bluno - BLE with Arduino Uno

Introduction

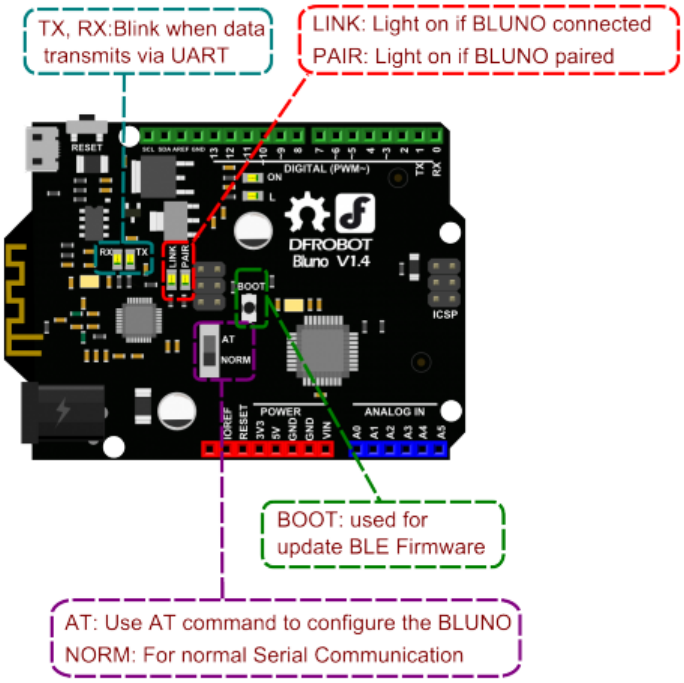
It's time to get Bluetooth 4.0 into your project, together with your phone! For aficionados of smart devices and wearables, now you can go further than hacking things bought in the market to building your own prototype out of garage. The Bluno board is first of its kind in intergrating BT 4.0(BLE) module into Arduino Uno, making it an ideal prototyping platform for both software and hardware developers to go wireless. You will be able to develope your own smart bracelet , smart pedometer and more. Through the low- power Bluetooth 4.0 technology, real-time low energy communication can be made really easy.

Bluno integrates with a TI CC2540 BT 4.0 chip with the Arduino UNO development board. It allows wireless programming via BLE, supports Bluetooth HID, supports AT command to config the BLE, and you can upgrade BLE firmware easily. Bluno is also compatible with all Arduino Uno pins which means any project made with Uno can directly go wireless!

Specification

- On-board BLE chip: TI CC2540
- Wireless Programming Via BLE
- Support Bluetooth HID
- Support AT command to config the BLE
- Transparent communication through Serial
- Upgrade BLE firmware easily
- DC Supply:USB Powered or External 7V~12V DC
- Microcontroller: Atmega328
- Bootloader: Arduino Uno (disconnect any BLE device before uploading a new sketch)
- Compatible with the Arduino Uno pin mapping
- Size: 60mm*53mm
- Weight: 30g

Board Overview



APPENDIX F

Ethic of Conduct Form for Human Health Related Research

Ver 3.0 September 2014 *NMRR/FORM/IAHODIA*

INVESTIGATOR'S AGREEMENT, HEAD OF DEPARTMENT AND ORGANISATIONAL / INSTITUTIONAL APPROVAL
PERSETUJUAN PENYELIDIK DAN KEBENARAN KETUA JABATAN DAN PENGARAH ORGANISASI/INSTITUSI

This document is intended for online submission for formal research registration. It is issued as the Investigator's Agreement to participate in the research as well as the investigator's **Head of Department and Director's Approval**. Please upload this document in the required section in NMRR upon completion.
**Note: This form is NOT to be used for obtaining permission to conduct the research at the named / selected study site(s).

*Dokumen ini adalah untuk penghantaran 'online' mengikut prosedur rasmi pendaftaran penyelidikan. Borang ini dikeluarkan sebagai pengakuan penyelidik untuk menjalankan penyelidikan dan persetujuan serta kebenaran daripada **Ketua Jabatan dan Pengarah masing-masing**. Sila lengkapkan borang ini dan memuat naik ke dalam sistem NMRR di seksyen yang telah ditetapkan.*
***Nota : Borang ini BUKAN digunakan untuk tujuan mendapatkan kelzinan untuk menjalankan penyelidikan di lokasi kajian yang dipilih.*

Research Title *[Tajuk Penyelidikan]*	Development of Portable Bluetooth Device for Noninvasive Glucose and Heart Rate Monitoring Systems for Diabetic targeted patient		
Research ID *[Nombor Pendaftaran]*	27085	**Protocol Number (if available)** *[Nombor Protokol (jika ada)]*	

INVESTIGATOR'S AGREEMENT *[PERSETUJUAN PENYELIDIK]*

I have understood the above mentioned proposed research and I agree to participate as an investigator and being responsible to conduct the research.

Saya faham atas cadangan penyelidikan di atas dan bersetuju untuk mengambil bahagian serta bertanggungjawab untuk melaksanakan penyelidikan tersebut.

Name *[Nama]*	WIRA HIDAYAT BIN MOHD SAAD
IC number *[Nombor K/P]*	841026015961
Institute *[Institusi]*	UNIVERSITI TEKNIKAL MALAYSIA MELAKA (UTEM)
Signature and Official stamp *[Tandatangan dan Cop Rasmi]*	DR. WIRA HIDAYAT BIN MOHD SAAD Pensyarah Kanan Fakulti Kejuruteraan Elektronik Dan Kejuruteraan Komputer Universiti Teknikal Malaysia Melaka (UTeM) Hang Tuah Jaya 76100 Durian Tunggal, Melaka
Date *[Tarikh]* 25/3/17	

HEAD OF DEPARTMENT AGREEMENT *[PERSETUJUAN KETUA JABATAN]*

I agree to allow the above named investigator to conduct the above titled research.

Saya bersetuju dan membenarkan pegawai seperti bernama di atas untuk menjadi penyelidik di dalam projek penyelidikan tersebut di atas.

Name of Head : *[Nama Ketua Jabatan]*	
Signature and Official stamp *[Tandatangan dan Cop Rasmi]*	PROFESOR MADYA DR. NURULFAIAR BIN ASD. MANAP Dekan Fakulti Kejuruteraan Elektronik dan Kejuruteraan Komputer Universiti Teknikal Malaysia Melaka (UTeM) Hang Tuah Jaya 76100 Durian Tunggal, Melaka
Date *[Tarikh]*	28/3/17

ORGANISATIONAL / INSTITUTIONAL APPROVAL *[KEBENARAN ORGANISASI / INSTITUSI]*

I acknowledge and approve the named officer to conduct the above titled research.

Saya mengesahkan dan mengambil maklum penglibatan pegawai ini di dalam penyelidikan tersebut.

Name of Director *[Nama Pengarah]*	
Signature and Official stamp *[Tandatangan dan Cop Rasmi]*	PROF. DATUK Ts. DR. SHAHRIN BIN SAHIB Naib Canselor Universiti Teknikal Malaysia Melaka
Date *[Tarikh]*	28/3/17

This is computer generated. Borang ini adalah cetakan komputer. [27085/36550/100110]

APPENDIX G

Research Subject Information and Consent Form

Borang Maklumat dan Keizinan Subjek

Tajuk Kajian: Development and Analysis of Near-infrared (NIR) Spectroscopy Technique for Non-invasive Blood Glucose Monitoring System.

Nama Penuh: .....

Tarikh Lahir: ..... Umur: .....

Pekerjaan: .....

Berat: ..... Tinggi: .....

BMI: .....

Pengidap Kecing Manis: Ya ( ) Tidak ( )

Nyatakan sekiranya mempunyai masalah kesihatan lain: .....

.....

Kajian	Bacaan meter tidak invasif	Bacaan meter invasif
1		
2		
3		

Saya, secara sukarela, bersetuju menyertai kajian penyelidikan ini, mematuhi segala prosedur kajian dan memberi maklumat yang diperlukan.

....

Tanda Tangan Subjek Tarikh