Early Detection and Monitoring of Dengue by Image Processing and Bioimpedance Analysis

***Abstract* – This paper covers the development of an early dengue detection and monitoring device. This device evaluates and classifies the dengue severity of a patient based on the hydration status and the total platelet count. The hydration status is determined using bio-impedance analysis, a non-invasive technique used to measure total body water. For the platelet analysis, a software was developed that would process microscopic blood images to automatically generate the total platelet count. The performance of these subsystems is expounded on this paper.**

Keywords: Dengue, Platelet, Hydration Level, Image Processing, Bioelectrical Impedance

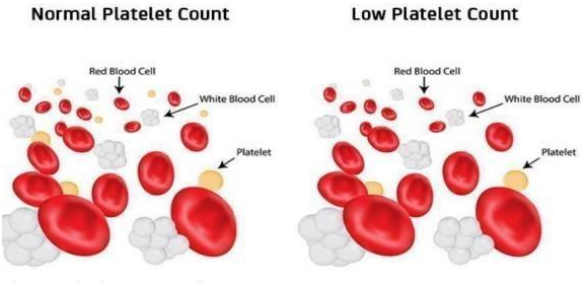
I. INTRODUCTION

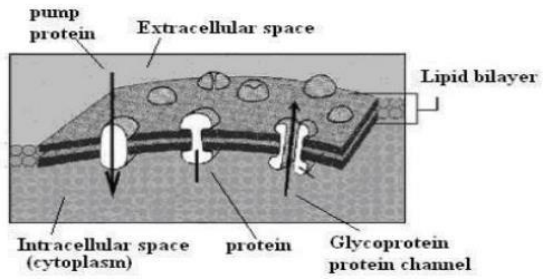
Dengue is a viral infection transmitted by the bite of an infected mosquito. After 3-14 days that the initial infection occurred, the Dengue fever will kick in the system of the victim affecting any age bracket. The symptoms can be quite unclear such as high fever, headaches, vomiting, skin rash, and muscle pain but you can never be sure without clinical tests [1]. The incidences of dengue disease in the Philippines were reported mostly from populated urbanized areas. The number of reported cases fluctuated throughout 2000s, with an overall increase in cases over time. According to World Health Organization (WHO), in the recent years, dengue is a mosquito- borne viral disease that has speedy growth in all regions. Only female mosquitoes contract the dengue virus. Methods had been launched by the WHO in order to cure Dengue infection. Most of these are based in laboratory tests to identify the infection'sstatus [2] The disease can be dangerous if the stage of the virus is on a high level. Determining platelet count is a way to know the stage level of the dengue virus. Blood test procedure commonly known as Complete Blood Count (CBC), used to detect different disorders and evaluate the health of a person [3]. In medical field, Complete Blood Count is essential in determining the cell count of a person. Cells count is important in knowing the capability of a body system, abnormal count of cells can be diagnosed into a specific kind of disease like dengue infection. Having a high fever and vomiting can dehydrate the body. One treatment to consider is preventing the patient from dehydration. Dehydration is a state of a person wherein the body does not have enough water to function normally. It occurs when the body lost more fluid than taking in, fail to replace lost fluids leads to dehydration [4]. One way to monitor a dengue infection is the change in platelet count of a person. Thrombocytes is a term that is also used for platelets. To prevent the person to bleed or to prevent clotting, there is a condition called thrombocytopenia which is the other term for low platelet count. There are methods to monitor the hydration like using instruments that measure the physiological properties of the skin, which almost all of the measurements are done in a hospital setting. To prevent the expensive, bulky instruments and achieve a low-cost hydration monitoring, the Bioimpedance Analysis (BIA) is one way to use for monitoring the hydration level. The measurement of the body impedance is a method that has a relationship to Total Body Water (TBW), which can be expressed into a person's hydration state.

II. RELATED STUDIES

This section aims to display a number of gathered resources by the proponents particularly journals and articles, which serve as the reference of the study. The possible materials, algorithms, and methods that the proponents used in the study will also be discussed.

A top ranked dangerous disease in the Philippines is Leukemia. Detecting the disease earlier is the most initial step to prevent it from getting worse. The detection process in the medical laboratory is time consuming and expensive. The aim of this study entitled *Leukemia Detection with Image Processing Using Matlab with GUI* is to use a Matlab software for detection, which calculates the percentage of White Blood Cells area in the image and classify the image as a Leukemia or not. [5] Another study entitled *Hybrid Approach to Detect Dengue Virus Present in Blood Cell Images* used image processing method for dengue and WBC classification. For this purpose, canny edge detector is used to identify strong edges. A decision tree classifier and Dilation is applied in the method. After dilation, WBC and Dengue classification is done by using trained data. The analysis results are filtered based on the set criteria to obtain the effected WBC [6].





The body needs small blood cells that aids that body to produce clot that helps stop bleeding, these small blood cells are called platelets. A signal is sent out to the platelets once a blood vessel is damaged. The cells then go to the damage site and forms clog to fix the damage. [7] Platelet count is important in determining some diseases, with that specialist can detect medical issues.

There is also a study based on white blood cells and it avoids the complexity of lab technicians and reduces errors. The process includes image processing methods for detecting WBC abnormalities. The system also uses CNN based recognition for analyzing visual images. CNNs uses little pre-processing compared to other image classification algorithms [8].

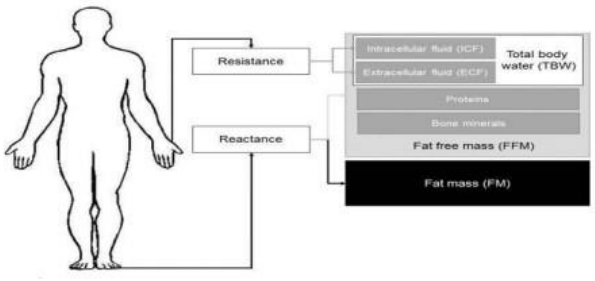
At the present, there are similar studies that are related to the proposed topic. One of which is entitled *Microscopic Image Segmentation Using Hybrid Technique for Dengue Prediction*. The study aims to create a system that focuses on platelet counting using segmentation from other blood with a microscopic image. A hybrid soft computing technique efficiently segments platelets [9].

For related studies regarding the hydration state of a person, a study named *Wearable Hydration Sensor with Conformal Nanowire Electrode.* This study is about a wearable skin hydration sensor using electrodes and capacitors. Silver nanowires in a polydimethylsiloxane matrix are used for the capacitors and parallel electrodes that are integrated. Moreover, multimodal sensing is developed through various components like electrodes and strain sensors. This device can be used for monitoring the hydration of the skin and other health related data knowing that this is low-cost [10]. Another study aims to develop a wearable device using a Bio Impedance Analysis method. BIA is used to identify sedentary dehydration. Body impedance measurement is a

method that has the benefit of being directly linked to the total body water volume (Total Body Water) equal to the body's hydration state [11].

Human skin has high impedance to alternating current of low frequency. The Bioelectrical Impedance Analysis (BIA) technique is the worthiest out of the possible sensing techniques in determining the hydration. Total Body Water of a person can be measured using BIA where pair of electrode pads is placed on the skin to inject a small current.

According to Dunkelmann, Bioimpedance devices are now used to evaluate body composition and water content of children and adults. These devices are safe and easy to use, and correctly estimate the water content of children and adults. Also, the measurement of hydration can be achieved accurately by using the resistance of the body at 50 kHz [12].



There is a study entitled *Wearable Health Monitoring Systems* where sensor systems are becoming less obtrusive and more powerful following advances in wire-less technology, low power electronics and the Internet ofThings (IoT). Sensors are used to monitor various activities such as brain activity, heart activity, and muscle activity [13].

According to the National Institutes of Health, BIA measures in terms of electrical and biological parameters vary from person to person. Instruments for BIA introduced into the body known amount of current (I), about 800μA at a frequency of 50 kHz. The current is transmitted between two electrodes and generates voltages between different points in the body volume according to Ohm's law[14].

III. METHODOLOGY

The main objective of this study is to create a device that can count platelets per microscopic blood image and identify the hydration state of a person for rapid monitoring of dengue patients. To do so, each of the established specific objectives as stated in Chapter 1 must be addressed. This section highlights the underlying engineering principles and theoretical analysis of the methods behind the solutions used to realize the objectives of this research endeavor.

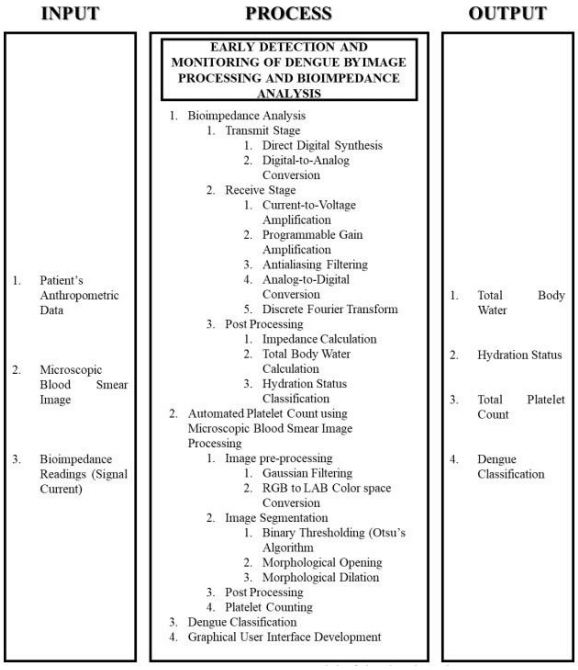


Figure 1: Input-process-output model of the developed system

The developed system takes the following as input: patient's anthropometric data, microscopic blood smear image, and bio impedance readings. The input anthropometric data includes age, weight, and height. These data are inputted via the graphical user interface and are used in the calculation of the total body water. These variables are needed in the total body water empirical formula. As for the microscopic blood smear image, this is inputted via USB communication protocol.

The device supports USB 2.0 storages devices such as thumb drives and hard drives. The graphical user interface allows users to browse the image file of the microscopic blood images on the containing directory. The developed software supports portable network graphic (PNG) and joint photographic experts group (JPEG) image file formats. The automated platelet counting using image processing involves various processes such as image pre-processing, image segmentation, post processing and platelet counting. The image processing prepares the input image for further processing. Noise are removed from the image using Gaussian filters. The denoised image is then converted to LAB color space from RGB. The b-channel containing blue to yellow information is used as the input for the image segmentation. The image segmentation process eliminates the RBCs and WBCs on the blood smear image leaving only the platelets. The post processing enhances the segmented image prior to the counting of platelets.

Once the total platelet count is obtained and the hydration status is known, dengue classification is now possible For the bio impedance analysis, readings in the form of signal current is inputted to the bio impedance analyzer circuit. The transmit stage is made up of a phase accumulator direct 36 digital synthesizer and a digital-to-analog converter followed by an amplifier circuit. This stage provides the output excitation signal at a particular frequency for the excitation of external impedances, in this case the human body.

The excitation signal is coupled to the human body using wet electrodes. The signal current that is developed across the unknown impedance flows into the voltage input of the analyzer circuit. This signal will then pass through the receive stage and will undergo series of amplification including a current-tovoltage amplification. The received amplified signal is passed directly to the digital signal processing (DSP) core of the AD5933. This block performs discrete Fourier transform (DFT) on the sampled data. The DFT algorithm returns both a real (R) and imaginary (I) data-word at each frequency point along the frequency sweep. These data are then used to calculate the magnitude, phase and consequently the resistance at 50 kHz (R50). The total body water can now be calculated, and the hydration status may now be determined.

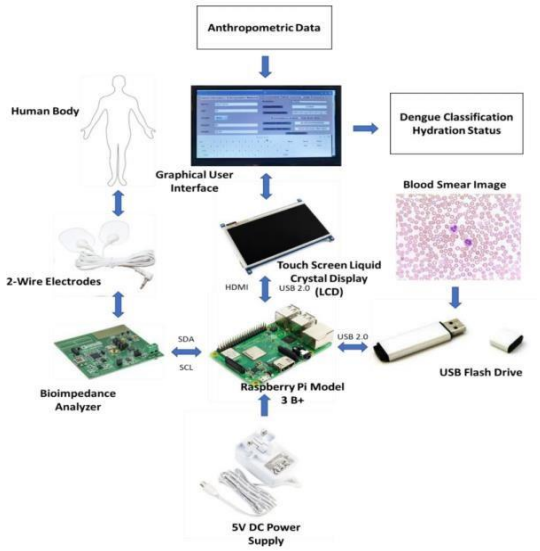


Figure 2 Block Diagram of the Early Detection and

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Figure 2 shows the block diagram of the developed system. It illustrates the interconnections of the system components as well as the system variables, inputs, and outputs. The Raspberry Pi 4 Model B is the main processing and computing unit of the system. Wet electrodes are used to couple the excitation signal generated by the bio impedance analyzer to the human body.

The response of the body to this excitation signal is also returned to the bio impedance analyzer trough the same electrodes. After DFT calculations, impedance real and imaginary data are sent to the main computing unit I2C communications protocol. The bio impedance analyzer circuit is interfaced to the Raspberry Pi using the SDA, SCL pins. The analyzer circuit is also powered by the Raspberry Pi using the 3.3V and GND pins. Microscopic blood smear images are inputted to the Raspberry Pi using a USB flash drive.

The images can also 39 be imported to the Raspbery Pi via cloud. Since Raspberry Pi supports WiFi, images can be download straight from the cloud. For the user's convenience, the device is equipped with a 7" touch screen liquid crystal display for the graphical user interface. Anthropometric data is inputted to the system through this. This also shows the hydration status and the dengue classification of the patient. The display is interfaced with the single board computer using HDMI cable and USB 2.0 cable. The whole system is powered by a 5V DC adapter.

IV. RESULTS AND DISCUSSION

A total of 36 out of 360 digital blood smear images only were used and selected by the proponents to evaluate the platelet count analysis of the device due to the limitation of the online blood image dataset that had been gathered. In the 360 images, not all has an annotation for platelet count.

There are only few images that shows platelet count with annotation and that results the proponents to only use images that had platelet count annotation for the device's evaluation. Using 36 microscopic blood images with annotations indicating the locations of the platelets from an online database, the performance of the developed program for the automated platelet count was evaluated. Each blood images underwent image processing to automatically generate a total platelet count.

The results of each image processing technique employed on the developed system. First the raw and unprocessed blood image is retrieved. The first technique that was applied to the blood image is Gaussian filtering. This technique removes random noise on the image and is known to give the image a blur. This is followed by the conversion of the blood image form RGB color space to LAB color space. After the conversion to the LAB color space, the composite image was split into its corresponding luminance and color layers. The b-channel or layer is the input for the image segmentation.

Image No.	True Positive (TP)	False Positive (FP)	False Negative (FN)	Total Count (Ground Truth)	Total Count (Proposed Algorithm)
12	2	0	0	2	2
37	2	0	0	2	2
39	2	0	0	2	2
45	1	0	0	1	1
46	1	0	0	1	1
55	3	0	0	3	3
103	1	0	0	1	1
117	2	1	0	2	2
135	3	0	1	4	3
145	3	0	0	3	3
148	1	0	0	1	1
156	6	1	0	6	6
168	2	0	0	2	2
174	1	0	0	1	1
197	2	0	0	2	2
212	1	0	0	1	1
214	2	0	0	2	2
215	2	0	0	2	2
222	4	1	0	4	4
223	3	0	0	3	3
233	2	0	0	2	2
254	2	0	0	2	2
277	2	0	0	2	2
296	3	0	0	3	3
297	3	0	0	3	3
310	3	0	0	3	3
314	5	0	0	5	5
322	3	0	0	3	3
325	3	0	0	3	3
327	2	0	0	2	2
365	3	0	0	3	3
369	3	0	0	3	3
375	2	0	0	2	2
403	2	0	0	2	2
407	3	0	0	3	3
410	4	0	0	4	4
TOTAL	89	3	1	90	89

Table 1: Platelet Count Analysis

After segmentation, it is evident that there were small debris or noise that were not removed. Also, the platelets are somehow distorted. To address this issue morphological transformations were employed. The opening operation is a combination of erosion and dilation. The small pixels which correspond to noise were removed afterwards. To compensate for the noise and losses during binarization another morphological operation known as dilation was utilized to help round up again the platelets.

After the contours were determined, contours that have pixels below 15 or those that have pixels greater than 110 were disregarded. These values were determined by experimentation. Now to evaluate the performance of this algorithm, for each processed image, the true positive, false positive and false negative detections were counted. True positives are correct platelet 140 detections that are listed in the given blood image annotation. False positives are platelet detections that are found to be true but are not listed in the annotation. Lastly, false negatives are platelets that were listed in the annotation that are not detected by the system. The results were tabulated above.

To calculate the performance metrics accuracy, sensitivity, and we use equations below.

$$Accuracy = \frac{TP + TN}{TP + FP + FN + TN} \, x \, 100$$

$$Accuracy = \frac{89 + 0}{89 + 3 + 1 + 0} x \, 100 = 95.70\%$$

$$Precision = \frac{TP}{TP + FP} \, x \, 100$$

$$Precision = \frac{89}{89 + 3} \, x \, 100 = 96.74\%$$

$$Sensitivity = \frac{TP}{TP + FN} \, x \, 100$$

$$Sensitivity = \frac{89}{89 + 1} \, x \, 100 = 98.89\%$$

	TP	FP	FN	Accuracy (%)	Sensitivity (%)	Precision (%)
Platelet	89	3	1	95.7900	98.8900	96.7400

Table 2: Performance Metrics

Table 2 shows the performance matrix of the device. The accuracy value of 95.79% means that there is only 4.21% probability that the device can incorrectly label an image component of the entered image as platelet. The sensitivity of 98.89% means that there is only 1.11% probability that our developed system will falsely detect a platelet on the blood image. Finally, the precision value of 96.74% implies that on the average, only 3.26% is the probability of our developed system to identify that a platelet detection is an outlier or other blood component.

The results of the platelet analysis are subjected to statistical analysis. For the case of the comparison when the results of the proposed algorithm were the same with that of the ground truth data, a two-tailed Mann Whitney U test calculator was used. The Mann-Whitney U test, also called the Wilcoxon rank-sum test, is a non-parametric test. It checks continuous or ordinal data for a significant difference between two independent groups. The test merges the data from the two groups. Then, it sorts the data by the value. Unlike the t-test that compares the groups' averages, the rank test compares the entire distributions. When the two groups' distributions have a similar shape, the test will also compare the median of each group. For symmetrical distribution, the median is the average. For this test, a significance level of 0.05 was chosen, outliers were also considered in the analysis and Z approximation was used. Since Z approximation was used, continuity correction is observed. The hypotheses for this test are as follows:

$$H_0 = Group_1 = Group_2$$

$$H_1: Group_1 \neq Group_2$$

where group 1 is the total count based on the ground truth while group 2 is the total count using the proposed algorithm. The U calculation is given by:

$$U_1 = n_1 n_2 + \left(\frac{n_1(n_1 + 1)}{2}\right) - R1$$

$$U_2 = n_1 n_2 + \left(\frac{n_1(n_1 + 1)}{2}\right) - R1$$

$$(U_1 + U_2) = n_1 n_2$$

Since the distribution is symmetrical, usually U is the minimum between U1 and U2

$$U = (U_1 U_2)$$

In order to get more accurate results, ties and continuity correct is used. The test statistic Z is then calculated as

$$Z = \frac{U_2 - \mu_u + C_{continuity}}{\sigma_u}$$

Where

$$\mu = \frac{n_1 n_2}{2}$$

$$\sigma^2 = \frac{n_1 n_2 (n_1 + n_2 + 1)}{12}(1 - C_{ties})$$

For ties correction we have,

$$n = n_1 + n_2$$

$$C_{ties} = \sum_{i=1}^{t} \frac{f_t^3 - f_t}{n^3 - n}$$

Where t is the group number of ties and ft is number of values in group t. 144 For continuity correction for right tail, or two tails with positive Z, (U2 > μ), Ccontinuity = -0.5. For continuity correction for leftt tail, or two tails with negative Z, (U2 < μ), Ccontinuity = 0.5. Using the data from the platelet count analysis tables using Tukey Fence (k=1.5) as the outliers' detection method, the total count (ground truth) contains 2 potential outliers, which is 5.56% of the observations. Similarly, the total count (proposed algorithm) contains 2 potential outliers which is also equal to .56% of the observations.

The table below shows the calculated test parameters.

	Ground Truth	Proposed Algorithm
Sample average (x bar)	2.5	2.47222
Sample size (n)	36	36
Sample SD (S)	1.13389	1.10805
Median	2	2
Skewness	0.99605	1.07475
Skewness Shape	Asymmetrical, right/positive (pval=0.011)	Asymmetrical, right/positive (pval=0.006)
Normality	0.00079	0.00038
Outliers	5, 6	5, 6
Outliers Count	2	2
U	641	655
Ties Correction	0.09624	

Table 3: Test Paramters

Before using the device for the measurement of total body water, the bio impedance analyzer must be calibrated first to avoid erroneous results. The ideal value for the calibration resistor RCAL is 500 Ω. However, there is no resistor available with that exact value which is why a resistor with a value of 510 Ω was used instead. Following the step-by-step calibration procedure, the following calibration data was obtained.

To obtain correct impedance and measurement at every frequency sweep point, the corresponding gain factor and system phase must be calculated.

$$Gain\ Factor = \left(\frac{Admittance}{Code}\right) = \left(\frac{\frac{1}{Impedance}}{Magnitude}\right)$$

Sweep Index	Frequency (Hz)	Real Data	Imaginary Data	Magnitude (Ω)	Phase (°)
0	49,990.00	5308	-8136	9714.389	303.1208
1	49,992.00	5309	-8137	9715.773	303.1225
2	49,994.00	5310	-8138	9717.157	303.1242
3	49,996.00	5311	-8134	9714.354	303.1421
4	49,998.00	5308	-8137	9715.227	303.1176
5	50,000.00	5311	-8136	9716.029	303.1356
6	50,002.00	5310	-8135	9714.645	303.1339
7	50,004.00	5309	-8133	9712.423	303.1354
8	50,006.00	5308	-8134	9712.714	303.1272
9	50,008.00	5312	-8135	9715.738	303.1438
10	50,010.00	5308	-8137	9715.227	303.1176

Table 4: Calibration Data (RCAL= 510 ohms

To demonstrate the gain factor calculation, we take the measurement at 50 kHz as an example. The impedance connected across the VIN and VOUT during this frequency sweep is equal to RCAL=510 Ω. Substituting the value of RCAL and the magnitude at 50 kHz to Equation 5, we have:

$$Gain\ Factor = \left(\frac{\frac{1}{510\ \Omega}}{9716.029}\right) = 2.01809228e-7$$

The developed bio impedance analyzer was first tested using known resistors prior to testing with human body. There are different resistors used ranging from 46 Ω up to 4640 Ω to test and evaluate the correctness of the readings within the expected impedance range as well as below and beyond the range. The resistance of the known resistors is measured using a FLUKE 18B+ Digital Multimeter. The result of this measurement was set as our ground truth data. The values that will be measured using the analyzer will be compared to the ground truth data. See Figure 51 for the resistance measurement for the known resistors that will be used as the unknown impedance, ZUNKNOWN.

After measuring all the resistance, each resistor will be connected across the VIN and VOUT

pin of the AD5933. The test set up is similar to that of the calibration setup. See Figure 50. The only difference is that instead of connecting the RCAL, the known resistors will be connected. For each resistor, a frequency sweep is performed, and the results were tabulated together with the results of the digital multimeter measurements. In this section, only the tables and graphs of the resistors that is within the range of 300 Ω to 700 Ω will be shown. The rest of the gathered data is presented on the Appendix. The table shows the values of the resistors for each test.

All the resistors have a standard ±5% tolerance.

Test No.	Resistor Value (Ω)
1	46
2	100
3	220
4	300
5	330
6	510
7	680
8	1000
9	1200
10	1800
11	3300
12	4640

Table 5: Calibration Data (RCAL= 510)

Test 4		FLUKE 18B+ Digital Multimeter		Bioimpedance Analyzer Circuit			Errors	
Theoretical Value	Frequency (Hz)	Resistance (Ω) DMM	Phase (°) DMM	Magnitude (Ω) BIA	Phase (°) BIA	Resistance (Ω) BIA	Percentage Error (%) Resistance	Absolute Phase Error (°)
300 Ω ± 5% (285 Ω - 315 Ω)	49990	301.7000	0.0000	302.7032	0.2343	302.7006	0.3317	0.2343
	49992	301.7000	0.0000	302.5527	0.2755	302.5492	0.2815	0.2755
	49994	301.7000	0.0000	302.4160	0.2555	302.4130	0.2363	0.2555
	49996	301.7000	0.0000	302.5387	0.2346	302.5361	0.2771	0.2346
	49998	301.7000	0.0000	302.3563	0.2748	302.3528	0.2164	0.2748
	50000	**301.7000**	**0.0000**	**302.6164**	**0.2400**	**302.6138**	**0.3029**	**0.2400**
	50002	301.7000	0.0000	302.5620	0.2068	302.5600	0.2851	0.2068
	50004	301.7000	0.0000	302.4168	0.2335	302.4143	0.2368	0.2335
	50006	301.7000	0.0000	302.1864	0.2788	302.1829	0.1600	0.2788
	50008	301.7000	0.0000	302.2907	0.2593	302.2876	0.1948	0.2593
	50010	301.7000	0.0000	302.5907	0.2328	302.5882	0.2944	0.2328

Table 6: Comparison of DMM Resistance Reading vs BIA Resistance Reading ($Z_{UNKNOWN}$ = 300 Ω)

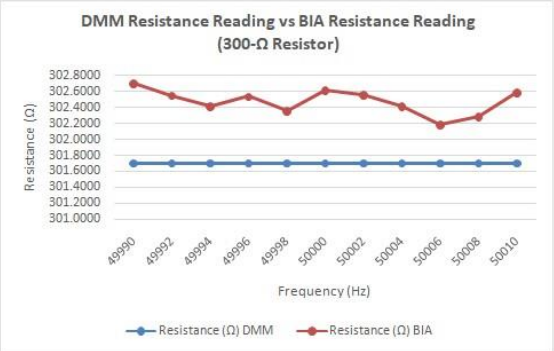

Table 7: Comparison Plot of DMM Resistance Reading vs BIA Resistance Reading ($Z_{UNKNOWN}$ =300 Ω)

V. CONCLUSION

A prototype of a new system for total body water estimation and hydration status determination was developed. This device has an integrated automated platelet counting software which takes in microscopic blood smear images and generates total platelet count. In addition to this, based on the inputted multiplier and the generated total platelet count, dengue classification is also provided.

Test and evaluations were performed for the bio impedance analyzer using resistors with known values. This demonstrated that the calibration process is very important to obtain correct and accurate impedance readings for the total body water estimation. Test and evaluations were also performed on human body where repeatability of measurements was also proved as several frequency sweeps on a same human subject generated measurement which are within acceptable limits.

Using an established empirical formula, the total body water was estimated, and hydration status were determined For the automated platelet analysis, the developed software was verified using microscopic blood smear images from publicly available datasets. The developed software showed promising results as platelet detections indicated high accuracy.

The proponents concludes that the general objective of our study is possible and attainable due to the step by step process of specific objectives. The development of software algorithm in terms of counting platelet, integrated by the impedance analyzer helps the system to achieve the desired output of the study. 149 With this, all the objectives of the study were met. With some hardware improvements and additional data for software optimization, this prototype would be very useful to our local healthcare facilities in monitoring and detecting dengue fever for dengue probable patients.

Chapter 6

RECOMMENDATIONS

The proponents of this study would recommend the following to further the work done in this study. First, a stable external oscillator should be used to improve stability and reduce the variations observed in the measured results particularly on the phase measurements. The selection of BIA formula is very critical, most analysis and modellings for the TBW measurement were done abroad and the participants usually resides there also.

In fact study shows that race or ethnicity somehow affects the TBW so instead of adopting the empirical formulas for TBW on previous journals, write a formula instead or make some analysis using data from Philippine populations to be able to come up with a more accurate prediction or estimation of TBW for Filipinos.

It is also recommended to compare the results of various BIA methods such as single frequency BIA (SF-BIA), multi-frequency (BIA) or bio impedance spectroscopy (BIS), to determine which type if bio impedance measurement is best suited for the determination of a person's hydration status. For the platelet analysis image processing algorithm, use more blood images to further optimize the algorithm

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