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Bioelectrical Impedance Analysis at Popliteal Regions of Human Body using BIMS

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Abstract

Bioelectrical impedance (BI) at popliteal regions was measured using a bioelectrical impedance measurement system (BIMS), which employs the multi-frequency and the two-electrode method. Experiments were performed as follows. First, a constant AC current of 800 μA was applied to the popliteal regions (left and right) and the BI was measured at eight different frequencies from 10 to 500 kHz. When the applied frequency greater than 50 kHz was applied to human's popliteal regions, the BI was decreased significantly. Logarithmic plot of impedance vs. frequency indicated two different mechanisms in the impedance phenomena before and after 50 kHz. Second, the relationship between resistance and reactance was obtained with respect to the applied frequency using BI (resistance and reactance) acquired from the popliteal regions. The phase angle (PA) was found to be strongly dependent on frequency. At 50 kHz, the PA at the right popliteal region was 7.8° slightly larger than 7.6° at the left popliteal region. Third, BI values of extracellular fluid (ECF) and intracellular fluid (ICF) were calculated using BIMS. At 10 kHz, the BI values of ECF at the left and right popliteal regions were 1664.14 Ω and 1614.08 Ω, respectively. The BI values of ECF and ICF decreased sharply in the frequency range of 10 to 50 kHz, and gradually decreased up to 500 kHz. Logarithmic plot of BI vs. frequency shows that the BI of ICF decreased noticeably at high frequency above 300 kHz because of a large decrease in the capacitance of the cell membrane.

Keywords: Bioelectrical impedance (BI), Resistance (R), Reactance (X_C), Phase angle (θ), Extracellular fluid (ECF), Intracellular fluid (ICF), Capacitance of cell membrane (C_m), bioelectrical impedance measurement system (BIMS).

1. INTRODUCTION

Bioelectrical impedance analysis (BIA) is a safe, practical, and non-invasive method for measuring components of biological tissues and biological materials [1-3]. BIA relies on the conduction of a radio-frequency electric current by fluids (water, interstitial fluid, and plasma) and electrolytes in the body, and the permeability or conductivity of cell membrane [4].

Depending on the applied frequency, BIA can be classified into single-frequency analysis and multi-frequency analysis. [6]. Single-frequency analysis is used to measure bioelectrical impedance (BI) while applying a constant alternating current (AC) to living tissues or biological samples. On the other hand, multi-frequency analysis is used to measure BI at each frequency by applying a chirp waveform in combination with multiple frequencies ranging from low-frequency (LF) to high-frequency (HF). The advantage of single-frequency analysis is that BI can be measured in over a short period of time, but the disadvantage is that BI cannot be analyzed in different frequency bandwidths [7]. In contrast, the advantage of multi-frequency analysis is that characteristics of the living tissue or biological samples can be analyzed in different frequency bandwidths. However, the measuring circuit for this multi-frequency analysis can be rather complicated [5].

Many studies on BIA have been carried out to analyze the composition of living tissues or biological samples [6-11]. Deurenberg et al. [6] examined the application of the BI method to measure the composition changes in human body. Kushner et al. [7] utilized BIA to determine the quantities of extracellular water (ECW) and total body water (TBW) in human body. Kanai et al. [8] discussed the problems of measuring the distribution of intracellular fluid (ICF) and extracellular fluid (ECF) in living tissues using BI. They reported that the ICF and ECF distributions were related to some physiological parameter, such as blood circulation, metabolism of tissues, and electrolytic concentration

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of ICF and ECF. Dumler et al. [9] presented a low bio-impedance phase angle (PA) predicting a higher mortality and lower nutritional status in chronic dialysis patients. Kumar et al. [10] measured the PA in healthy human subjects through BIS. They reported that PA values were 7.322º in healthy subjects and that PA values had a significant positive correlation with body mass index (BMI) ($r=0.011$, $P<0.001$). Kim et al. [11] developed a BIMS, with a multi-frequency analysis method and two-electrode method, for measuring the BI in the body segments.

In this study, the BI at the popliteal regions was measured using BIMS with the multi-frequency analysis and two-electrode method. The experimental procedure was as follows. First, the BI was measured at eight different frequencies ranging from 10 to 500 kHz after attaching an in-planar (IP) electrode to the popliteal regions. When a current of 800 μA having a frequency higher than 50 kHz was applied to the left and right popliteal regions, the BI decreased significantly. The logarithmic plot of impedance vs. frequency indicated that there were different mechanisms in the impedance phenomena, according to the applied frequency (also energy). Second, the relationship between resistance (R) and reactance (X_C) was acquired from experimental data, which indicated a strong dependence of PA (θ) on the applied frequency. Third, the BI values of ECF and ICF were computed using multi-frequency BIMS. The BI values of ECF and ICF were rapidly decreased rapidly in the range of 10 to 50 KHz, and then, decreased gradually in the range of 50 to 500 kHz. From a logarithmic plot of BI vs. frequency, the ECF and ICF, BI of ICF was remarkably reduced at 300 kHz or higher. This is consistent with the report [12] that the capacitance of the cell membrane is considerably reduced at high frequencies.

2. RESEARCH METHOD

2.1 Theory of Impedance

Impedance (Z) is an obstruction to the flow of alternating current, depending on the frequency of the applied current. Impedance is often represented as a complex quantity (Z) [13]; the polar form conveniently captures both magnitude $|Z|$ and PA (θ) characteristics. The magnitude $|Z|$ represents the difference ratio of the voltage-amplitude to the current-amplitude, while the symbol θ indicates the phase angle difference between voltage and current. Resistance (R) is the real part of impedance; a device with purely resistive impedance exhibits no phase shift between voltage and current. The capacitor affects the current, and has the ability to stop the current flow when fully charged. Since an AC voltage is applied, there is a root mean square (*RMS*) current; but this current is limited by the capacitor. The capacitive reactance (Xc) of a capacitor (C) is inversely proportional to both the C and the angular velocity (ω). Hence, greater capacitance and higher frequency results in smaller X_C [15]. The capacitor reacts differently at different frequencies. Capacitors impede low frequencies the most, because low frequencies allows time to get charged, and hence, stop the current flow. If the frequency goes to zero (DC), X_C approaches infinity, and the current becomes zero once the capacitor is charged. At very high frequencies, the capacitor's reactance approaches zero, and it no longer impedes the current flow. The physical significance of the complex impedance is that the steady-state current is not in phase with the applied voltage. Resistance and reactance together determine the magnitude and PA of the impedance.

The primary conductor in the human body is water. In the human body, a low resistance is associated with large amounts of lean body mass. And a high resistance is associated with small amounts of lean body mass. Low resistance, indicating high conductivity, is due to large amounts of water in the body. Resistance in the body is thus proportional to the amount of lean body mass, since water is contained solely within the lean body mass. Reactance (X_C) is the imaginary part of impedance; a component with a finite reactance induces a PA shift between voltage and current. Reactance is a measure of the cells' ability to store energy. A body that stores energy easily has high reactance and a body that stores energy poorly has low reactance. Energy is stored in the cell membrane; therefore, this reading gives an indication of the amount of intact cell membranes in the body. Since intact cellular membranes are contained primarily within the body cell mass, the reactance of the body is proportional to the amount of body cell mass. Thus, the reactance helps to calculate the proportion of the body that is metabolically active.

The energy of all living things comes from cells that consume oxygen and nutrients, and expel carbon dioxide and waste. The quantity and efficiency of cells directly affect the PA. The outer boundary of a cell is a plasma membrane of phospholipid molecules that act as a dielectric to form an electrical capacitor, when a radio-frequency signal is introduced to the cell's environment. Capacitance is fundamental to any human PA measurement. The higher the capacitance, the greater is the PA. Lower PA suggests cell death or decreased cell integrity, while higher PA suggests large quantities of intact cell membranes. Body capacitance is the absolute amount of the energy storage of the body due to intact cellular membranes. A high capacitance indicates that the body stores energy effectively. A low capacitance would suggest that cells have trouble storing energy. Normal values of body capacitance are between 500 to 1000 pF. An elite athlete has a higher PA than a sedentary person. It has been well documented that the PA decreases with disease, age, and reduced activity levels. The PA has been found to be a prognostic indicator in several clinical conditions. For adults, it ranges from 3^o to 10^o, with normal values being between of 6^o to 8^o. A PA of 5^o or lower can indicate a serious energy deficiency. However, a PA higher than 8^o indicates a healthy state.

2.2 Equivalent Circuit of ECF, Cell Membrane, and ICF

TBW occupies about 60% of the weight depending on the age, sex, and obesity. The ICF accounts for about 40% of the TBW and the ECF for about 20% of TBW. Further, the interstitial fluid (ISF) occupies about 15% of ECF and the plasma about 5% of ECF. Despite having lower protein content, the composition of ISF is similar to that of plasma. Cells constituting the human organs consist of ICF and ECF, which behave as an electrical conductor, while the cell membrane acts as an electrical capacitor [16].

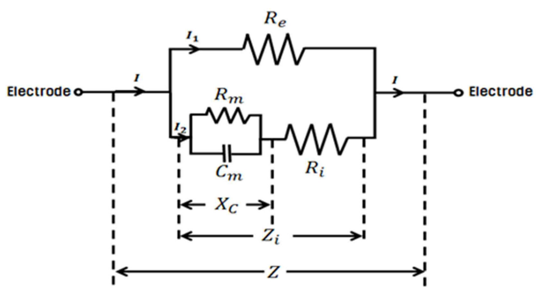

Fig. 1. Equivalent circuit of the cell membrane (C_m), ECF (R_e), and ICF (R_i).

Table 1. Descriptions of symbols in Fig. 1

Symbol	Description
C_m	Capacitance of cell membrane
R_m	Resistance of cell membrane
R_e	Resistance of ECF
R_i	Resistance of ICF
X_c	Reactance of cell membrane
Z_i	Impedance of X_c and R_i (ICF)
Z	Impedance of Z_i and R_e (ECF)
I	Current through both ECF & ICF
I_1	Current through only ECF
I_2	Current through both cell membrane and ECF

Fig. 1 indicates an equivalent circuit of the cell membrane, ECF, and ICF. Table 1 gives a description of the indicated symbols.

Since the resistance (R_m) and the capacitance (C_m) of the cell membrane are connected in parallel, the reactance (X_C) of the cell membrane in Fig. 1 can be represented by Eq. (1).

$$X_C = \frac{1}{\dfrac{1}{R_m + j\omega C_m}} = \frac{R_m}{1 + j\omega R_m C_m} = \frac{R_m}{1 + j2\pi f R_m C_m} \tag{1}$$

Impedance (Z_i) of the cell membrane and the ICF can be represented by Eq. (2):

$$Z_i = X_C + R_i = \frac{R_m}{1 + j2\pi f R_m C_m} + R_i \tag{2}$$

The total impedance (Z), which consists of the reactance (Xc) of the cell membrane, the resistance (Ri) of the ICF, and the resistance (R_e) of the ECF can be represented by Eq. (3).

$$Z = \frac{1}{\dfrac{1}{Re} + \dfrac{1}{Z_i}} = \frac{ReZ_i}{Re + Z_i} \tag{3}$$

The reactance (Xc) of the cell membrane depends on the applied frequency. When the frequency is high, Z is decrease since X_C in Eq. (1) and Z_i in Eq. (2) decreases Vice versa, when the applied frequency is low, Z increases as the opposite phenomenon occurs.

2.3 Bioelectrical Impedance Measurement System (BIMS)

BIMS was described in detail in our previous paper [11]. BIMS consisted of a main control unit (MCU, ATmega128, NewTeC Co., Korea), a multi-frequency generation (MFG) unit, an automatic gain control (AGC) unit, a constant current source (CCS), an in-planar (IP) electrode, a preprocessing part, and PC. MCU outputs the control command with respect to the frequency generated by the MFG, and controls the overall function of the BIMS. Frequencies of 10, 50, 100, 150, 200, 300, 400, and 500 kHz are generated in the MFG unit. The output voltage of the frequency generated by the MFG is automatically controlled in the AGC unit. Constant AC current of 800 μA is generated in the CCS. While the current_flows into the popliteal regions (left and right), the segmental BI is measured and transferred to the PC after preprocessing.

The PC program was developed using LabVIEW (LabVIEW 2010, National Instruments Co., USA) to control BIMS and analyze the measured BI values. The PC program was configured to set parameters such as starting frequency, incremental frequency range, frequency setting number, and output voltage. The measured BI was displayed in the form of bode plots and tables, and stored in the PC.

3. RESULTS

The experimental subjects were five male adults with a mean age of 27.5 years (±2.5 years), an average height of 173 cm (±3.2 cm), and an average weight of 75 kg (±4.1 kg). Each experiment was conducted five times for 10 subjects using BIMS. Each measurement was performed after a 10-min break. Prior to the experiment, the purpose and method of this study were explained to the subjects and their consents were obtained.

The BI of popliteal regions in the body was measured using multi-frequency BIMS with the two-electrode method. Ag/AgCl electrode (Monitoring electrode, 3M Co., USA) commercialized for ECG measurements was used as the IP electrode. Eight different frequencies generated from the MFG unit were sequentially applied to the IP electrodes attached to the popliteal regions (left and right) through AGC and CCS. A constant current of 800 μA at each frequency ranging from 10 to 500 kHz was then supplied. Fig. 2 (a) shows the BI values as a function of frequency (*f*) and energy (*eV*) when the IP electrodes with separation of 7 cm, were positioned on the popliteal regions. At a frequency of 10 kHz, the BI values at the left and right popliteal regions were 1560 Ω and 1540 Ω, respectively. BI values decreased, abruptly for the frequencies 10 to 50 kHz, and then gradually for frequencies from 50 to 500 KHz. These results are consistent with the result reported by Lukaski et al. [17]. When a low frequency (~ 10 kHz) is applied to the popliteal regions, the BI is observed to be high since the current flows through the ISF. When a medium frequency (10 ~ 50 kHz) is applied, the BI is slightly lower since the current flows mainly through the ECF (ISF and plasma). When a high frequency (50 ~ 500 kHz) is applied to the popliteal regions, the BI is very low because the current flows through both ECF and ICF. It is interesting to note that the BI of the right popliteal region was slightly lower than that of the left popliteal region. This is because the experimental subjects had a slightly greater muscle mass on the right side of the popliteal region.

Fig. 2 (b) illustrates a logarithmic plot of impedance (Z) as a function of the applied frequency (Hz) and energy (*eV*). In particular, Fig. 2 (b) has different slopes of log Z vs. log f at 4.70 Hz in the log scale, indicating that there are different mechanisms for the current flow in the cell before and after 4.70 Hz. Furthermore, Fig. 2 (b) has another different slope for log Z vs. log f at 5.48 Hz, suggesting yet another mechanism in the current flow at the popliteal regions (left and right).

As the frequency is increased from 50 to 300 kHz, the conductivity of the cell membrane is gradually increased, which allows the current to flow into ICF through the cell membrane. When a frequency greater than 300 kHz are applied to the cell, more current flows into ICF through the cell membrane, leadings to a significant decrease in the BI. Overall, the measured BI of the right popliteal region was slightly lower than that of the left popliteal region as shown in Fig. 2(a).

Fig. 3 shows the relationship between the resistance and reactance at the popliteal regions. The values of the resistance and the reactance were reduced as the frequency was increased from 10 to 500 kHz. The curve of resistance vs. reactance measured at the right popliteal region is slight higher than that measured at the left popliteal region. This is because the muscle mass and cell

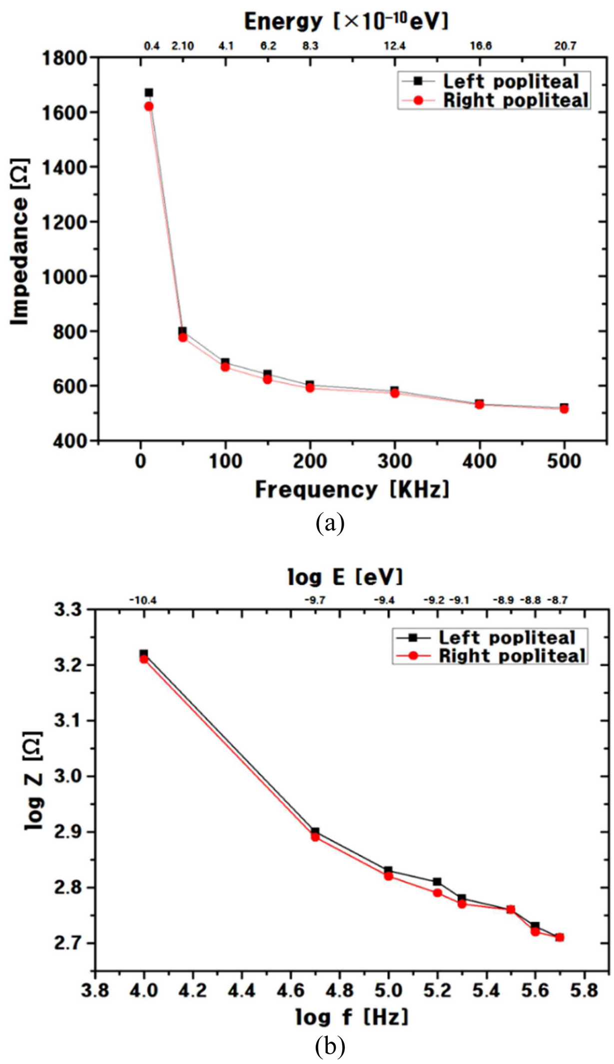

Fig. 2. (a) Plot of BI vs. frequency and (b) Plot of log BI vs. log frequency at the left and right popliteal regions.

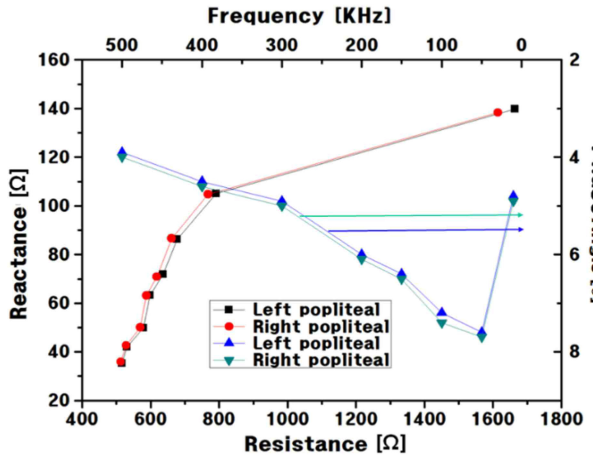

Fig. 3. Resistance, reactance, and PA obtained using BIMS.

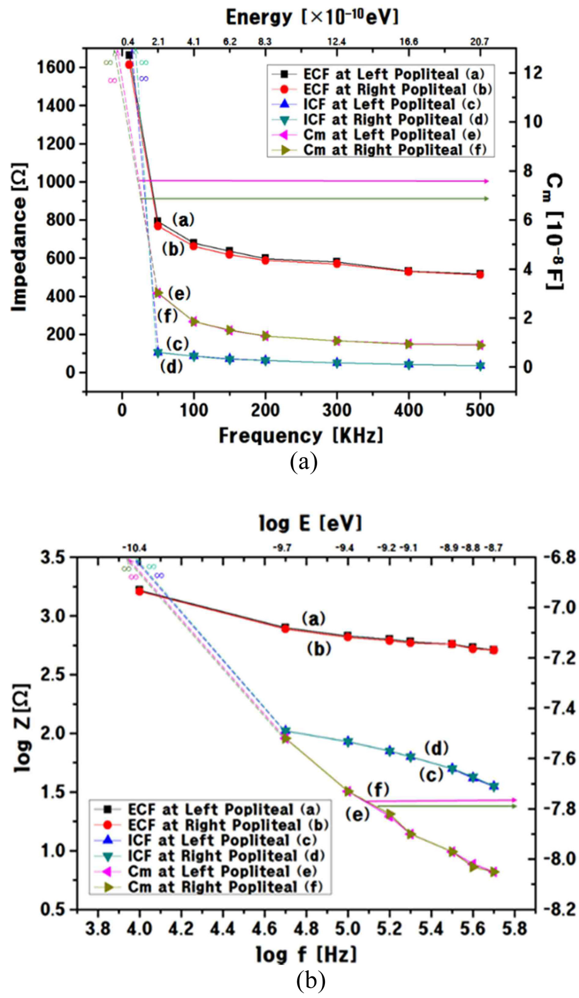

Fig. 4. (a) Plot of BI (ECF, ICF, and C_m) vs. frequency (energy) and (b) Plot of log BI (ECF, ICF, and C_m) vs. log frequency (energy).

membrane integrity at the right popliteal region are higher compared to those at the left popliteal region. Reactance is proportional to the muscle mass and the structural integrity of the cell membrane [18].

The right side of Fig. 3 (plot of frequency vs. PA) illustrates the PA (θ) between resistance and reactance at the popliteal regions, in the frequency range of 10 to 500 KHz. The PA (θ) was increased significantly between 10 and 50 kHz, and then reduced gradually from 50 to 500 kHz. The PA (θ) at the right popliteal region was also slightly larger than that at the left popliteal region. The PA (θ) measured at the left and right popliteal regions at 50 kHz were 7.6^{o} and 7.8^{o}, respectively. The PA (θ) has been suggested to be an indicator of cellular health, where higher values reflect higher cellularity, greater cell membrane integrity, and better cell function [19]. S. Kumar et al. [14] reported that the PA (θ) values for males and females were 7.43±0.98º and 7.05 ±1.1.58º, respectively.

Fig. 5 (a) shows the BI values of popliteal regions as a function of ~~the~~ frequency and energy (eV). These values were reduced as the frequency increased from 10 to 500 kHz. In Fig. 5(a), BI values of ECF and ICF were calculated using multi-frequency BIMS. At 10 kHz, the BI values of ECF in the left and right popliteal regions were 1664.14 Ω and 1614.08 Ω, respectively. BI of ECF decreased rapidly from 10 to 50 kHz, and then gradually decreased from 50 to 500 KHz. It was estimated that BI of ICF at 10 kHz approached a high value because the current could not flow through the cell membrane. An 800 uA current at a frequency of 10 kHz has an energy of $4.14\times10^{-11}\,eV$, which is lower than the threshold potential ($2.05\times10^{-10}\,eV$) of the ion (Na^{+}) channel in the cell membrane. However, these values decreased rapidly in the BI of ICF when the frequency increased from 10 to 50 kHz. A current of 50 KHz has an energy ($2.10\times10^{-10}\,eV$), which is greater than the threshold potential of the ionic channels in the cell membrane. The right side of Fig. 4 (a) represents the capacitance (C_m) as a function of frequency. Capacitance (C_m) values of the cell membrane were calculated by substituting the BI values of ICF and the applied frequency in Eq. (1) [21], and these values (represented by curves (e) and (f)) are indicated by violet and green arrows.

Fig. 4 (b) illustrates a logarithmic plot of log Z (log BI values of ECF and ICF) as a function of the log of applied frequency (Hz) and log energy (eV). Curves (a) and (b) represent the BI values of ECF. These BI values decreased with increase in frequency, but the slopes were a bit different at 50 kHz. Curves (c) and (d) represent the BI values of ICF. These values decreased rapidly from 10 to 50 kHz, gradually decreased from 50 to 200 kHz, and then significantly decreased from 300 to 500 kHz.

Therefore, BI values of ECF and ICF exhibit different behaviors in the impedance phenomena before and after 50 kHz. Curves (e) and (f) in the right side of Fig. 4 (b) represent the log capacitance (C_m) as a function of log frequency.

4. CONCLUSION

While applying a constant alternating current of 800 μA to the popliteal region, BI was measured at eight different frequencies ranging from 10 to 500 kHz using multi-frequency BIMS and IP electrode. The distance between the IP electrodes attached to the popliteal region in the body was 7 cm.

The experimental results are as follows. First, BI was measured at eight frequencies ranging from 10 to 500 kHz using multi-frequency BIMS. The results indicated that BI rapidly decreased rapidly from 10 to 50 kHz, and then decreased gradually from 50 to 500 kHz. A logarithmic plot of impedance (Z) as a function of applied frequency (Hz) and energy (eV) illustrated that BI had different slopes of log Z vs. log f at 4.7 Hz in the log scale, indicating that there were different mechanisms for the current flowing in the cell membrane before and after 4.7 Hz in the log scale. Second, the relationship between resistance and reactance was obtained with respect to frequency. The curve of resistance vs. reactance measured at the right popliteal region was slight higher than that measured at the left popliteal region. The PA (θ) was found to be strongly dependent on the applied frequency. At 50 kHz, the PA (θ) at the right popliteal region was 7.8°, which is slightly larger than the 7.6° at the left popliteal region. This is due to the fact that majority of the subjects had slightly greater muscle mass and better cell membrane integrity in the right popliteal region. Third, BI values of both ECF and ICF decreased rapidly from 10 to 50 kHz, and then decreased gradually from 50 to 500 kHz. Logarithmic plot of BI vs. frequency revealed that the BI of ICF was greatly reduced. This is because, as the applied frequency surpasses 300 kHz, the capacitance of the cell membrane decreased considerably.

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