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Abstract. Offer a blood-mimicking fluids (BMF) for test object of medical ultrasound diagnostic instruments in this article is done. Both the acoustical (speed of sound and attenuation) and physical (density and viscosity) properties were defined and specified in the International Electrotechnical Commission standard and are given as a constant value. However, the viscosity of human blood depends greatly on the shear rate of the blood. In accurate studies that use a specified flow phantom, the BMFs with the appropriate viscosity are required. Furthermore, since the BMF considered as Newtonian, the viscosity it can has an arbitrary viscosity (more than 4.1 mPa.s). This article displays a general overview about the effect of density and molecular weight of dextran which is a component of mixture fluid that was used in BMF preparation. For this reason, we replaced the dextran 185000D (0.90 g/ml) with dextran 150000D powder material (0.25 g/ml). To produce liquids with proper viscosity in unit of mPa.s, we have prepared a new ternary fluid made of water, glycerol and dextran 150000D powder material.

1. Introduction
Using medical Doppler ultrasound technique for measuring the blood flow depends on the changing or variation of ultrasound frequency that reflected from movable particles of blood during the flow. The variation of frequency known as the Doppler shift or frequency shift and increases with increasing the speed of blood flow [1]. The achievement of Doppler medical ultrasound instrument can be estimated with several test objects [2-5]. The test object is also known as a flow phantom which has been used in clinical research field. Both the acoustical and physical properties blood mimicking fluid (BMF) must closely match to the human blood to make the examination meaningful.

The International Electrotechnical Commission (IEC) was used as a standard and constant values of the acoustical and physical properties of Blood Mimicking Fluid for flow phantoms Table 1. [2, 6]. Several various Blood Mimicking Fluids have been reported [2, 6-10]. Some scattered particles were added to the Blood Mimicking Fluid to produce scattering influence of blood cells, also the variating of the characteristics based on if the scattered particles deposit down or float.
Traditional BMFs have contained scattered particles material such as nylon powder (Orgasol), polystyrene microspheres, starch, Sephadex, and silicon powder in the fluid to form BMF which was used for different purposes. However, the densities of these scattered particles should closely match the densities of mixture fluids to prevent float or deposit down of those particles and thus to allow keep neutrally buoyant of scattered particle materials inside this liquid.

Table 1. Specifications of the BMF defined as the IEC standard. \( f \) is the acoustic frequency (Hz).

| Attenuation coefficient \([\text{dB/cm/MHz}]\) | \(<0.1 \times 10^{-4} \times f\) |
| Speed of sound \((\text{m/s})\) | 1570 ± 30 |
| Density \((\times 10^3 \text{ kg/m}^3)\) | 1.050 ± 0.040 |
| Viscosity \((\times 10^{-3} \text{ Pas})\) | 4.0 ± 0.4 |

2. Materials and Methods

In this research study, our target was to prepare a new BMF with proper speed of sound, attenuation, and density that defined and known in the IEC standard [2], in addition to the arbitrary viscosity since the BMF does not depend on shear rate. Water, glycerol, and dextran 150000D dispersed with nylon 12 particles were used to obtain the BMF.

However, in this experiment research, the concentration of glycerol and dextran is changed several times to get good acoustical and physical properties which known in the IEC standard. In this research study, three different experiments were made. In the first experiment, both the dextran 150000D concentration and the glycerol concentration were increased in the mixture with small ratios. The second experiment was done in the same way of the first experiment but with changing the items’ concentration to obtain the suitable speed of sound and density which should be equivalent to IEC standard, the mixture made of the dextran 150000D concentration with the same amounts of the glycerol concentration. Finally, the glycerol was increased and fixed dextran 150000D concentration in some mixture samples and then fixed the glycerol and increase the dextran concentration in the other mixture. However, to make sure that the values of the sample number 3 in Table 2 are the optimum values for preparing the BMF. The changes of the items’ concentrations around this sample were done in Table 3.

Applying of Scheffé equation for several reasons. Firstly, because the mixture made of three different components (water, glycerol, and dextran) for the new BMF. Secondly, the density of the liquids have no linear relationship between each other in several experimental results [25, 26] for designing tests with a mixture fluid. The function of the three different components \( M \) represented by three different factors are \( S_1, S_2, \) and \( S_3 \). Equations 1 and 2.

\[
M = H_1S_1 + H_2S_2 + H_3S_3 + H_{12}S_1S_2 + H_{13}S_1S_3 + H_{23}S_2S_3 + H_{123}S_1S_2S_3
\]

\[
S_1 + S_2 + S_3 = 1
\]

Where \( S_1, S_2, \) and \( S_3 \) are weight fractions. And, \( H_1, H_2, H_3, H_{12}, H_{13}, H_{23}, \) and \( H_{123} \) are constants. There are seven coefficients \((H_1-H_{123})\) are unknown. In this case, the naming suggested by Scheffé equations (seven equations) will help us to find the values of the seven unknown coefficients since the Scheffé principle depends on seven equations with seven coefficients. The research study data \( M \) for seven group of values of \( S_1, S_2, \) and \( S_3 \) can be utilized to design the BMF. The answer of \( M \) to ingredients of \( S_1, S_2, \) and \( S_3 \) are given by the research study data displayed in Table 4.

Through the exchange \( M = V_o \) or \( M = P_o \), With the substitution \( M = V_o \) or \( M = P \), the general equation given by Equation 2 can be rearranged. Where the \( V_o \) and \( P_o \) are the speed of sound and the density of the mixture fluid samples, respectively. The typical formula determined by Equation 1, can be rearranged. The amounts of \( V_o \) and \( P_o \) for various group of values of \( S_1, S_2 \) and \( S_3 \) are given by the
research study data displayed in Table 3. Where S1, S2 and S3 are the components of water, glycerol, and dextran 150000D as wt% in the sample, respectively.

The BMF items of glycerol, dextran 150000D, water, and nylon were used in the research study experiments are 99% pure, supplied by Sigma Aldrich Company. The water was distilled utilizing a quartz distiller. The dynamic viscosities of the items were measured applying an electronic rotational viscometer in mPa.s, displayed in Tables 3 and 4. The temperature readout during measurement was 37 °C like human body temperature. The densities of the materials were measured applying density meter DMA35 with an accuracy of 0.001. The speed of sound in the materials was specified by the pulse-echo PE method by applying A-scan GAMPT ultrasonic device with uncertainty of ±3 m/s, and this uncertainty was estimated from repeated measurements of the reference distilled water scan in the Plexiglas and it was ±0.01 m/s as a random uncertainty and ±3.0 m/s as a systemic uncertainty. The temperature of the samples item when measuring the densities, speed of sound and viscosity was preserved within ±1 °C.

3. Results and Discussion
In the first experiment, when both the dextran 150000D concentration and the glycerol concentration were increased with slightly ratios, the speed of sound of mixture solution decreased and the density decreased too (proportional relationship) but there are no suitable result values between the density and speed of sound of the mixture fluid like values defined in the IEC standard. The reason of decreasing the speed of sound that it is directly proportional to the density, and the density of mixture decreases due to the glycerol concentration was more than the dextran concentration and the dextran density much lower (five times) than glycerol density. At the second experiment, the speed of sound of mixture solution decreased and the density decreased too, but also there was a mismatch between the speed of sound and the density according to IEC standard. The main cause of mismatch is the densities of items, such as glycerol has density more than blood density about 0.2 g/ml and the dextran 150000D has density less than the blood density about -0.8 g/ml. Thus, the minus value more than the positive value. At the final test, we noted that both the speed of sound and the density of the mixture solution increased (proportional relationship) when the glycerol increased and fixed dextran 150000D concentration, whereas the speed of sound and density decreased when we fixed the glycerol concentration and increased the dextran concentration Table 2, because the dextran density is much lower (five times) than glycerol density and vice versa. However, because the first two experiments were not a suitable to be mixture fluid for preparing BMF. Thus, we chosen the results of the final one experimental research study since it was a suitable result of the mixture solution for preparing BMF. Sample number 3 at Table 2 shows the BMF that has a proper speed of sound density and viscosity. However, the density of a suitable mixture fluid at sample 3 was 1.020 g/ml, speed of sound was 1581 m/s and the dynamic viscosity was 4.4 mPa.s. The viscosity of ternary mixture was increased gradually due to used dextran in all mixture with high molecular weight 150000D. And, when the molecular weight increases, the viscosity will increase directly.
Table 2. Constitutions and physical properties of water, glycerol and dextran 150000D solutions at change the mixture fluid ratios (temperature: 37.0 °C).

| Number of samples | Water Wt% | Glycerol Wt% | Dextran 150000D Wt% | density (g/ml) | speed of sound (m/s) | viscosity (mPas) |
|-------------------|-----------|--------------|---------------------|----------------|----------------------|------------------|
| 1                 | 100       | 0            | 0                   | 0.998          | 1508                 | 0.89             |
| 2                 | 95        | 4.5          | 0.5                 | 1.01           | 1522                 | 3.9              |
| 3                 | 90        | 9.5          | 0.5                 | 1.02           | 1581                 | 4.4              |
| 4                 | 88        | 11.5         | 0.5                 | 1.03           | 1610                 | 5.4              |
| 5                 | 86        | 12           | 2                   | 1.012          | 1578                 | 7.4              |
| 6                 | 85        | 12           | 3                   | 1.001          | 1530                 | 10.9             |
| 7                 | 83        | 12           | 5                   | 0.97           | 1485                 | 16               |

The research study results by using Scheffé equation displayed in Table 2 give the amounts of \( V_o, P_o, S_1, S_2, \) and \( S_3 \) in Equation 1, and we get the following:

\[
\begin{array}{cccccccc}
S_1 & S_2 & S_3 & S_{12} & S_{13} & S_{23} & S_{123} & \text{Density} \\
1.000 & 0.000 & 0.000 & 0.000 & 0.000 & 0.000 & 0.000 & \text{H1} & 0.988 \\
0.950 & 0.045 & 0.005 & 0.043 & 0.005 & 0.0002 & 0.0002 & \text{H2} & 1.01 \\
0.900 & 0.095 & 0.005 & 0.086 & 0.005 & 0.001 & 0.001 & \text{H3} & 1.02 \\
0.880 & 0.115 & 0.005 & 0.101 & 0.004 & 0.001 & 0.001 & \text{H4} & 1.03 \\
0.860 & 0.120 & 0.020 & 0.103 & 0.017 & 0.002 & 0.002 & \text{H5} & 1.012 \\
0.850 & 0.120 & 0.030 & 0.102 & 0.026 & 0.004 & 0.003 & \text{H6} & 1.001 \\
0.830 & 0.120 & 0.050 & 0.010 & 0.042 & 0.006 & 0.005 & \text{H7} & 0.970 \\
\end{array}
\]

Due to the number of formulas is like the number of unknown factors, thus the seven formulas are easy to solve together. Also, can specified \( H_1, H_2, H_3, H_{12}, H_{13}, H_{23}, \) and \( H_{123} \) relating to the density. So, the values of \( H_1, H_2, H_3, H_{12}, H_{13}, H_{23}, \) and \( H_{123} \) relating to the density are 0.998, 0.962, 0.933, 0.923, 0.900, 0.830, and 0.800, respectively. Thus, the standard deviation is ± 0.20. In other words, the different results between measurements by the equipment and Scheffé equation because the components of the mixture and density of the liquid don’t have linear relationship between each other g/ml.

In addition. The data that resulted from the research study can be specified that \( H_1, H_2, H_3, H_{12}, H_{13}, H_{23}, \) and \( H_{123} \) are relating to the speed of sound. As \( V_o \) and \( P_o \) are known in the IEC standard, we can utilize \( V_o = 1581 \text{ m/s} \) and \( P_o = 1.02 \text{ g/ml} \). The graph with the relation between the speed of sound and density in the dextran 150000D and glycerol aqueous solution are displayed in Figure. 1. The lines of the density \( P_o \) and speed of sound \( V_o \) intersect in the S1, S2, and S3 plane at values of S1, S2, and S3 for that the fluid meets both the speed of sound and density needs of the BMF. However, medical ultrasound technique produces Doppler spectral waveforms by the scattering of red blood cells inside a vessel. To mimic and simulate the scattering influence by red blood cells in the blood, scattered particles material of the BMF is dispersed with nylon 12 particles. Nylon 12 particles supplied by Sigma Aldrich were utilized in the experimental study. The density of the nylon 12 particles corresponds completely with the amount known in the IEC standard. Thus, the neutral buoyancy effect of the nylon 12 particles is guaranteed in the fluid with a density of 1.02 g/ml. Moreover, the particle size (diameter) of nylon 12 in this experiment is 5-10 µm.
Figure 1. Suitable speed of sound and density of fluid mixture of a BMF at different concentrations by wt% of (a) water (b) glycerol and (c) dextran 150000D, respectively.
Moreover, the values that resulted from mixture fluid in Table 3 to make sure that sample number 3 in Table 2 is the optimum specimen, were indicated that the sample number 3 is the optimum sample of mixture fluid to mixing with nylon particles for preparing BMF, see Table 3.

Table 3. Constitutions and physical properties of water, glycerol and dextran 150000D solutions at change the mixture fluid ratios (temperature: 37.0 °C), around sample number 3 in Table 2.

| Number of samples | Water Wt% | Glycerol Wt% | Dextran 150000D Wt% | density (g/ml) | speed of sound (m/s) | viscosity (mPas) |
|-------------------|-----------|--------------|---------------------|----------------|----------------------|-----------------|
| 1                 | 100       | 0            | 0                   | 0.998          | 1508                 | 0.89            |
| 2                 | 95        | 4.5          | 0.5                 | 1.01           | 1522                 | 3.9             |
| 3                 | 93        | 6.5          | 0.5                 | 1.014          | 1543                 | 4.1             |
| 4                 | 91        | 8.5          | 0.5                 | 1.018          | 1567                 | 4.3             |
| 5                 | 90        | 9.5          | 0.5                 | 1.02           | 1581                 | 4.4             |
| 6                 | 89        | 10.5         | 0.5                 | 1.025          | 1594                 | 4.9             |
| 7                 | 88        | 11.5         | 0.5                 | 1.03           | 1610                 | 5.4             |

Three different types of ternary aqueous solution were used for preparing the mixture fluid (water (90.0 wt %), glycerol (9.5 wt %), and dextran 150000D (0.5 wt %)) of BMF. The research study data, speed of sound and density of mixture fluids were 1581 m/s and 1.02 g/ml, respectively. The values were corresponded well with the theoretically calculated results. The BMF was prepared with both the density and speed of sound agreed in IEC standard. However, the items densities play an important role in the mixture fluid to get a suitable speed of sound and density, while the molecular weight plays an important role in mixture fluid viscosity.

4. Conclusion
New blood-mimicking fluid with the density (1.02 g/ml), viscosity (4.4 mPa.s), and speed of sound 1581 have been wanted for test object of medical ultrasound diagnostic instruments. The results from the initial search study pointed out that we can prepare the fluid mixture of BMF from water, glycerol, and dextran. As a conclusion, the acoustical features of this BMF was agreed the IEC standard. Moreover, changing the dextran sort affects the physical and acoustical features of the fluid mixture.

References
[1] Smith, H.-J., *Quantitative Doppler Flowmetry I. Construction and Testing of a Duplex Scanning System*. Acta Radiologica Diagnosis (Sweden), 1984. 25(4): p. 305-312.
[2] Yoshida, T., K. Sato, and T. Kondo, *Blood-mimicking fluid using glycols aqueous solution and their physical properties*. Japanese Journal of Applied Physics, 2014. 53(7S): p. 07KF01.
[3] Browne, J., et al., *Assessment of the acoustic properties of common tissue-mimicking test phantoms*. Ultrasound in medicine & biology, 2003. 29(7): p. 1053-1060.
[4] Law, Y., et al., *On the design and evaluation of a steady flow model for Doppler ultrasound studies*. Ultrasound in medicine & biology, 1989. 15(5): p. 505-516.
[5] Hoskins, P.R., *Simulation and validation of arterial ultrasound imaging and blood flow*. Ultrasound in medicine & biology, 2008. 34(5): p. 693-717.
[6] Ramnarine, K.V., et al., *Validation of a new blood-mimicking fluid for use in Doppler flow test objects*. Ultrasound in medicine & biology, 1998. 24(3): p. 451-459.
[7] Tanaka, K., et al., *Blood-Mimicking Fluid for Testing Ultrasonic Diagnostic Instrument*. Japanese Journal of Applied Physics, 2012. 51(7S): p. 07GF18.
[8] Oates, C., *Towards an ideal blood analogue for Doppler ultrasound phantoms*. Physics in medicine and biology, 1991. 36(11): p. 1433.
[9] Samavat, H. and J. Evans, *An ideal blood mimicking fluid for doppler ultrasound phantoms*. Journal of Medical Physics, 2006. 31(4): p. 275.

[10] Hoskins, P., T. Loupas, and W. McDicken, *A comparison of the Doppler spectra from human blood and artificial blood used in a flow phantom*. Ultrasound in medicine & biology, 1990. 16(2): p. 141-147.

[11] Scheffé, H., *Experiments with mixtures*. Journal of the Royal Statistical Society. Series B (Methodological), 1958: p. 344-360.

[12] Cornell, J.A., *Experiments with mixtures: designs, models, and the analysis of mixture data*. Vol. 895. 2011: John Wiley & Sons.