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Deriving a Blood-Mimicking Fluid for Particle Image Velocimetry in Sylgard-184 Vascular Models

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***Abstract*— A new blood-mimicking fluid (BMF) has been developed for particle image velocimetry (PIV), which enables flow studies in vascular models (phantoms). A major difficulty in PIV that affects measurement accuracy is the refraction and distortion of light passing through the interface between the model and the fluid, due to the difference in refractive index (n) between the two materials. The problem can be eliminated by using a fluid with a refractive index matching that of the model. Such fluids are not commonly available, especially for vascular research where the fluid should also have a viscosity similar to human blood. In this work, a blood-mimicking fluid, composed of water (47.38% by weight), glycerol (36.94% by weight) and sodium iodide salt (15.68% by weight), was developed for compatibility with our silicone (Sylgard 184; n = 1.414) phantoms. The fluid exhibits a dynamic viscosity of 4.31 ± 0.03 cP which lies within the range of human blood viscosity (4.4 ± 0.6 cP). Both refractive index and viscosity were attained at $22.2 \pm 0.2\,^{\circ}$C, which is a feasible room temperature, thus eliminating the need for a temperature-control system. The fluid will be used to study hemodynamics in vascular flow models fabricated from Sylgard 184.**

I. INTRODUCTION

Particle image velocimetry (PIV) is an engineering technique used as a gold standard to obtain quantitative flow information. The fluid is seeded with tiny particles (light scatterers), and a pulsed laser sheet of light is used to illuminate these particles. Images are taken, using a high resolution camera, in consecutive time intervals synchronized with the laser pulses. The images are analyzed with correlation algorithms in order to calculate the distance traveled by the particles between the tow intervals, from which velocity vectors are obtained.

To use PIV for vascular research, the blood vessels are modeled in transparent phantoms fabricated from different materials, such as glass (refractive index, n = 1.47), acrylic (n = 1.49), and silicone (n = $1.40 - 1.44$) [1]–[5]. The latter is of particular interest due to its versatility: silicone is readily available in a two-part liquid form that makes it easy to cast into the required geometry and dimensions, with high accuracy. Accordingly silicone (Sylgard 184, Dow Corning) was used in this work to cast anthropomorphic phantoms of vascular models using a lost-core casting technique [6]. The phantoms will be used for studying hemodynamics at the bifurcation of the common carotid artery (Fig. 1).

Imaging with PIV requires a match of the refractive index between the transparent model and the flowing fluid in order to eliminate optical problems such as image distortion due to refraction. Moreover, in vascular research, the PIV fluid should mimic human blood with respect to dynamic viscosity (μ) so as to obtain realistic blood-flow modeling measurements. Refractive index and viscosity are known to be temperature dependent therefore it is important that both are attained at the same working temperature. A blood-mimicking fluid (BMF) that has a refractive index matching to silicone and a dynamic viscosity matching to blood, both at the same temperature and preferably room temperature, is not readily available in the literature.

A blood analogue commonly used in the literature is a composition of glycerol and water [1], [7], [8], which demonstrates good optical properties as well as safety, availability and compatibility with silicone models. However, this fluid fails to satisfy the refractive index and viscosity properties, simultaneously. Although a composition of 36.6% (by vol.) glycerol in water would match the viscosity of blood [1], [7], [8], the resulting refractive index of 1.38 is lower than the refractive index of commonly used silicone

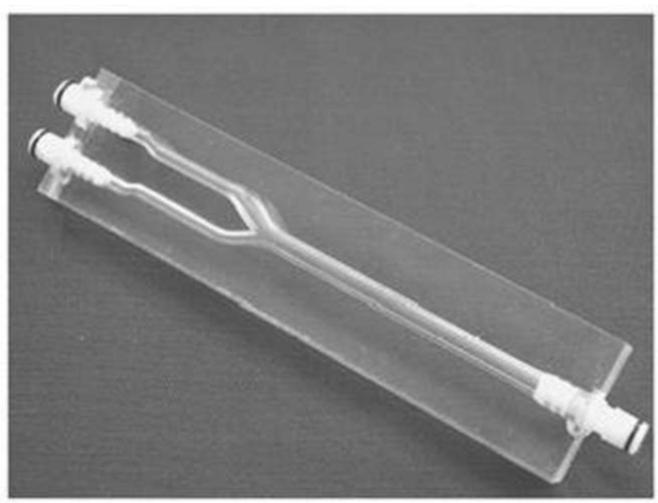

Fig. 1. Example of a silicone phantom incorporating a model of the carotid artery bifurcation with disease-free geometry.

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elastomers. Another composition of 59% (by vol.) of glycerol in water has been shown to have a refractive index of 1.43, providing a match with a specific silicone elastomer, with a kinematic viscosity of 6.55 cSt [2] (corresponding to a dynamic viscosity of approximately 7.0 cP). Such viscosity is higher than the range of dynamic viscosities for human blood (4.4 ± 0.6 cP) found in the literature (Table 1). When different compositions of glycerol and water, with refractive indices matching to silicone (Sylgard 184), were tested in this work at different temperatures, they also produced viscosity values higher than human blood.

Another fluid, accompanying an empirical model, introduced by Nguyen *et al.* [1], suggests combinations of diethyl phthalate (DEPH) and ethanol as a BMF with a wide range of refractive index and viscosity values at different temperatures. Applying the model on silicone elastomers shows good results for the refractive index, where various silicone elastomers can be matched by mixing different percentages of the DEPH and ethanol at different temperatures, including room temperature ($20-25\,^{\circ}$C). However, the resulting viscosity values were lower than typical dynamic viscosity of human blood. As shown experimentally by Nguyen *et al.* [1], the model produces a kinematic viscosity of 3.327 cSt (corresponding to a dynamic viscosity of approximately 3.3 cP) by mixing 55.6% (by vol.) DEPH and 44.4% (by vol.) ethanol to obtain a refractive index of 1.44 at $16.9\,^{\circ}$C. Such viscosity lies well outside the lower bound of human blood viscosity (3.8 cP) based on Table 1. In addition, the model suggests lowering the amount of DEPH to obtain fluids with refractive indices below 1.44 for matching other elastomers (i.e. silicones with n between 1.40 and 1.44). This will further decrease the resulting viscosity of the BMF, since the DEPH represents the viscous part in the combination, with a viscosity about ten times that of the ethanol [1]. Accordingly the expected viscosity values for fluids with refractive indices matching to silicone elastomers, will be well below an acceptable level for human blood. Note that the empirical model also suggests a relatively low temperature ($16.9\,^{\circ}$C) to achieve the proposed viscosity, which requires a temperature–control system.

Finally, a mixture of water, glycerol, and sodium iodide has been also used in the literature as a BMF for vascular studies with optical techniques [9], [10], [11], [12], [13].

TABLE I

DYNAMIC VISCOSITY FOR HUMAN BLOOD AS REPORTED IN THE LITERATURE FOR HIGH SHEAR RATES ($94.5-400$ s^{-1})

Reference	Blood Dynamic Viscosity (cP)
Vaya *et al.* (2008) [14]	3.93
Galduroz *et al.* (2007) [15]	3.37 - 4.36
Carrera *et al.* (2008) [16]	4.06
Rajzer *et al.* (2007) [17]	4.3 - 5.1
Kucukatay *et al.* (2007) [18]	4.84
Fehr *et al.* (2008) [19]	4.90 - 4.97
Antonova *et al.* (2008) [20]	5.83
Mean $\pm$ Standard Deviation	**4.4 $\pm$ 0.6**

However, the formulation was mostly prepared for phantoms fabricated from glass or acrylic which both have refractive index values (n_{glass} = 1.47, $n_{acrylic}$ = $1.485-1.492$) higher than common silicone elastomers. Moreover, the corresponding dynamic viscosity values shown by some references, were also higher than human blood viscosity found in the literature (4.4 ± 0.6 cP).

In summary, none of the PIV fluids of previous work mentioned in the literature demonstrates a simultaneous match of the refractive index and viscosity to enable vascular research with silicone models. This paper introduces a novel method for developing a blood-mimicking fluid with a refractive index matching to that of Sylgard 184 and a dynamic viscosity within an acceptable range of human blood, simultaneously matched at room temperature.

II. Materials and Methods

A. Flow Loop

The method that we propose is based on detecting the visual match of refractive index between the fluid and the phantom. A solution with constant amounts of water and glycerol is prepared, and sodium iodide salt is added in regular increments. The fluid's refractive index increases with the addition of sodium iodide until a formulation with a matching refractive index to our phantom, is achieved. The mixture is allowed to mix for around 30 minutes between increments to ensure that the salt (sodium iodide) is fully dissolved and the fluid is homogeneously mixed. The experimental apparatus is illustrated schematically in Fig. 2, where the fluid is mixed in a reservoir equipped with a magnetic stirrer. Flow is generated by a peristaltic pump to perfuse the fluid through the phantom placed over a grid paper. Images are taken of the phantom using a digital camera to detect the progression of the refractive-index matching process. The fluid proceeds through an Abbe refractometer (ATAGO NAR-3T) back to the mixing reservoir.

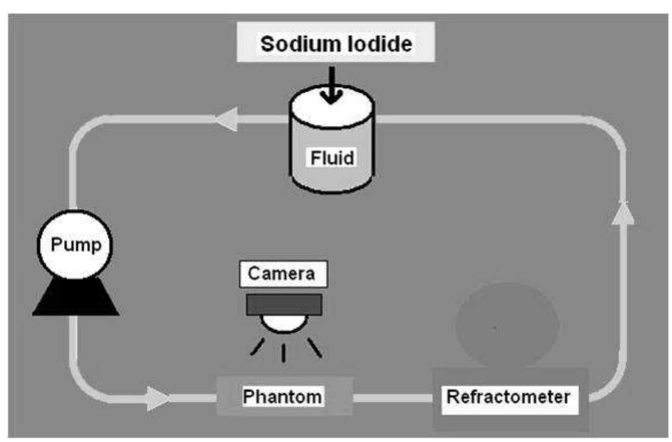

Fig. 2. A schematic of the flow loop showing the in-line set up for simultaneous perfusion of the phantom and refractometer while monitoring the distortion of grid lines beneath the phantom.

B. Refractive Index Measurement

A solution was prepared with a refractive index around 1.405, which is close to the lower bound of silicone elastomers (n = 1.40−1.44). The fluid was composed of water (50.21% by weight), glycerol (39.14% by weight) and sodium iodide salt (10.65% by weight). This solution has a glycerol-to-water ratio of (44:56 by weight) which was held constant during the experiment. Similar increments of approximately 0.5% (by weight) of sodium iodide were added while precisely measuring the refractive index for each concentration using the Abbe refractometer. The phantom was imaged at each concentration to monitor the distortion of grid lines, as shown in Fig. 3. The match in refractive index between the phantom and fluid was visually detected by the elimination of the distortion of grid lines (Fig. 3C).

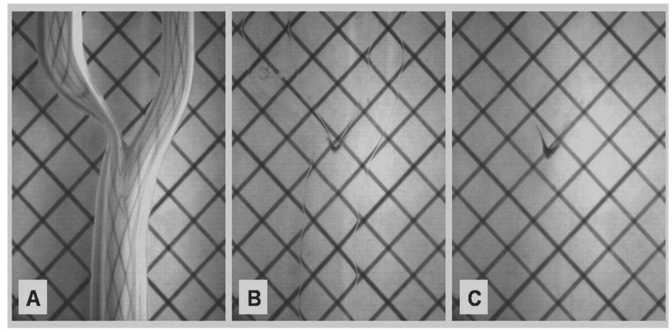

Fig. 3. Visual check of the refractive-index match based on monitoring the distortion of grid lines beneath a phantom filled with (A) air, showing high distortion, (B) nearly matched fluid (n = 1.4110) still showing some distortion, and (C) optimally matched fluid with no distortion (n = 1.4140). Note the unintentional stain at the bifurcation apex which provides a convenient landmark.

C. Viscosity Measurement

A sample of the fluid was prepared with relative concentration ratios similar to the formulation that matched the refractive index of our phantom. The fluid's kinematic viscosity was measured using a Cannon-Fenske (size 100 ml) viscometer, and density was measured using a volumetric flask and a digital scale. Dynamic viscosity was calculated from the product of kinematic viscosity and density.

Measurements were repeated five times for each of the kinematic viscosity and density. Mean and standard deviation were taken to obtain the final value of dynamic viscosity. All measurements were taken at 22.2±0.2 °C.

III. Results

We were able to visually detect the match of refractive index between the newly developed fluid and our phantom when the distortion of grid lines beneath the phantom completely disappeared (Fig. 3C). The matching fluid was composed of water (47.38% by weight), glycerol (36.94% by weight) and sodium iodide salt (15.68% by weight). The refractive index of the resulting fluid was 1.4140±0.0002 based on our Abbe refractometer. The dynamic viscosity of the fluid was 4.31±0.03 cP which lies within the range of human blood viscosity (4.4±0.6 cP) found in recent literature (Table. 1). An ideal match was found for this composition, which corresponds to n = 1.4140, but an acceptable match appeared between 1.4132 and 1.4148 thus indicating a range of uncertainty for the phantom's refractive index (n = 1.4140±0.0008). Note that both properties of refractive index matching and viscosity were achieved simultaneously at 22.2±0.2 °C.

IV. Discussion

The method used in this work allowed us, indirectly, to measure the effective refractive index (1.4140±0.0008) of our solid Sylgard-184 phantom, whereas literature sources specify a range of 1.41−1.43 [2], [21] for the same material. This demonstrates a variability in refractive index, and thus the possible need to adjust the formulation to match each individual phantom, while maintaining consistent viscosity. Characterization of refractive index as functions of sodium iodide concentration (Fig. 4) enables the adjustment of refractive index to match for different phantoms (i.e. batch-to-batch variance in refractive index) or different materials, whereas the ratio of glycerol to water enables good control of viscosity.

Human blood is a non-Newtonian fluid which means that the viscosity is dependent on shear rate [22], [23]. However the dependence on shear rate is thought to occur only in small vessels (diameter (d) $<$ 0.5−0.6 mm) [22], [23], therefore in large vessels it is acceptable to consider blood as a Newtonian fluid [22]. In addition, the non-Newtonian behavior of blood is considered to occur at shear rates below 50 s^{-1} [23]. In this work we are modeling the carotid artery bifurcation with a diameter between 4 and 8 mm, therefore we may consider blood as a Newtonian fluid and thus the high-shear viscosity in table 1 can be a reasonable target.

The final colorless fluid is composed of well-known, inexpensive components that are generally deemed to be relatively safe based on MSDS (Material Safety Data Sheet). However, the fluid exhibits a discoloration (yellowing) caused by ionization, which fortunately can be treated by adding 0.1% (by weight) sodium thiosulphate [24] to retrieve the colorless condition. Finally, the fluid works at an ideal room temperature (22.2±0.2 °C) therefore it does not require

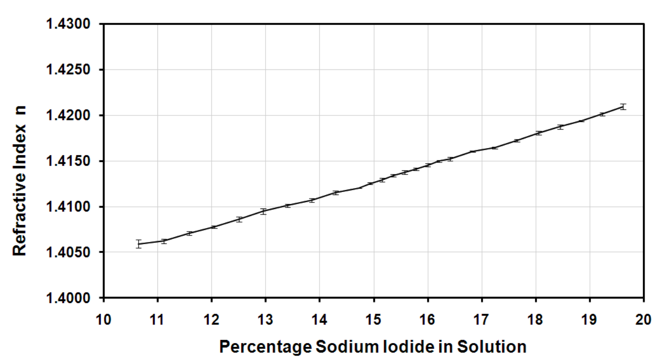

Fig. 4. Refractive index as function of sodium iodide in a solution of glycerol-to-water (44:56 by weight).

a temperature-control system.

V. CONCLUSIONS

A new BMF for PIV was developed and characterized in this work, which enables vascular flow studies in Sylgard–184 models with improved accuracy due to the elimination of image distortion, as well as matching of the appropriate dynamic viscosity for blood flow modeling.

The fluid successfully demonstrates all desired properties of an optimal PIV fluid, including refractive index match, blood-like viscosity, optical clarity, safety, and availability.

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