Effect of the Insonification Angle on the Doppler Backscattered Power Under Red Blood Cell Aggregation Conditions

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***Abstract*— It has been reported that power color Doppler ultrasound has important advantages over conventional color Doppler flow imaging. Some of these advantages are the aliasing free capability, the increased sensitivity to flow, and the angular independence. This last characteristic of power Doppler ultrasound was evaluated to verify if it was still valid in some well-defined flow conditions where porcine whole blood, calf red cells suspended in saline solution, and carbon fibers suspended in a water-glycerol mixture were used as scattering particles. Experiments were conducted under steady flow conditions (mean shear rates across the tube between 8.5 and 102 s^{-1}) for insonification angles between 40 and 80°. Different hematocrit values (5, 10, 20, and 40%) were specifically tested for porcine whole blood. Results indicated no angular dependence for the saline suspension of calf red cells while a strong anisotropy was observed for the carbon fibers. In this last situation, the Doppler power in decibels increased linearly with the insonification angle. The maximum found at 80° suggests that the fibers were aligned with the direction of the flow. For porcine whole blood, an angular dependence was observed at some specific shear rate conditions. At 40% hematocrit, the anisotropy was about 5 dB for shear rates between 17 and 51 s^{-1}, while for a lower (8.5 s^{-1}) or higher shear rate (102 s^{-1}), the anisotropy was reduced to approximately 2 dB. In all of these situations, the maximum Doppler power was observed for an insonification angle between 45 and 60°. For hematocrit values of 5, 10, and 20%, the anisotropy was respectively on the order of 2, 3, and 4 dB or less, depending on the shear rate conditions. Among the possible mechanisms that may explain the anisotropic effect observed in the present study, the structure of the red cell aggregates is believed to be the determinant factor. A hypothesis concerning the structure of the aggregates under flowing conditions in large diameter tubes is proposed.**

I. Introduction

BLOOD IS a biological tissue composed of numerous components (red and white cells, platelets, and protein macromolecules) suspended in a fluid plasma. Red blood cells (RBC) represent the dominant ultrasonic scattering factor since they constitute the majority of the cellular components of blood. Different theoretical models, classified into the particle or the continuum approach, were proposed to explain the echogenicity of blood [1]. These models are generally based on the Born approximation (no multiple scattering effect), which is valid when the dimensions of the scatterer are much smaller than the ultrasonic wavelength (Rayleigh scattering) and the acoustic mismatch between the scattering particles and the suspending fluid is small. It is also assumed that the ultrasound attenuation is negligible. From the particle model, it was found that the ultrasonic backscattering coefficient, BSC, from red blood cells suspended in saline is given by [2]

$$\mathrm{BSC} = \sigma_{bs}(H/V_c)W \tag{1}$$

where

$$\sigma_{bs} = \frac{\pi^2 V_c^2}{\lambda_0^4}\left[\frac{k_e - k_o}{k_o} - \frac{\rho_e - \rho_o}{\rho_e}\right]^2 \tag{2}$$

is the backscattering cross section of a single RBC, H is the hematocrit, V_c is the RBC volume, W is the packing factor depending on the interactions among scatterers, λ_0 is the ultrasonic wavelength, and k_e, ρ_e and k_o, ρ_o are the compressibility and mass density of the RBC's and surrounding fluid, respectively. From (1), no angular dependence of the ultrasonic backscattering coefficient is predicted since for Rayleigh scattering, the backscattering cross section (2) is independent of the shape of the RBC. In the continuum model, backscattering arising from spatial fluctuations in density and compressibility is also considered as isotropic.

In low shear flow conditions and some pathological situations, the echogenicity of whole blood was shown to significantly increase because of the presence of red cell aggregates, which represent larger scatterers [3]–[5]. Since the red blood cell aggregation involves the formation of long cylindrical straight chains of red cells which can branch, under very weak external forces, to form complex 3-D networks [6]–[8], it is possible that the acoustic Rayleigh scattering theory may no longer be applicable to model the echogenicity of RBC aggregates. For instance, experimental results using porcine whole blood at low shear rates and high ultrasonic frequency indicated that the frequency dependence was significantly lower than the fourth power relationship predicted by the theory [9], [10]. This result suggested that the shape of the scatterer may have to be taken into account in the theoretical evaluation of the backscattering coefficient with red blood cell aggregation. In these conditions, an angular dependence of the Doppler backscattered power due to the orientation of the rouleaux with the flow may be anticipated. The objective of the present study was to evaluate *in vitro* the angular dependence of the Doppler power in low velocity steady flow conditions

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where red blood cell aggregation is promoted. These results were compared with those obtained by circulating in the flow model washed red cells suspended in an isotonic saline solution and cylindrical carbon fibers suspended in a blood-mimicking fluid.

II. Materials and Methods

A. Flow Model and Experimental Design

Fresh calf and porcine blood collected from local slaughterhouses was used in this research. As recently quantified [11] with an erythro-aggregameter (Regulest, Nancy, France), calf whole blood has a minimal aggregation tendency while porcine whole blood has aggregation levels similar to normal human blood.[1] A solution of 1 g of EDTA (ethylenediamine tetra-acetic acid) per 10 mL of saline solution was added to the whole blood at a concentration of 30 mL per liter of blood to prevent coagulation. Concentrated erythrocytes were obtained by centrifuging the whole blood at 2318 g (3000 rpm) for 30 min with an IEC PR-6000 centrifuge, and by removing the plasma and the top white cell layer. For porcine whole blood experiments, the concentrated erythrocytes were recombined with the autologous plasma at a hematocrit of approximately 40%. This hematocrit was selected because it corresponds to the value where the aggregation tendency is maximum [12]. For calf red cells suspended in saline solution, blood was washed once with 0.9% normal saline solution buffed to pH 7.4. The washed erythrocytes were reconstituted with the buffered saline solution to a hematocrit of 40%. Using a suspension of red cells ensured that no aggregation was present. A microcentrifuge (Haemofuge, Heraeus Instruments) was used to measure the hematocrit by centrifuging the blood samples over 10 min at 14 980 g (12 000 rpm). Since the pH and temperature may affect the red cell properties, these parameters were monitored (Sentron pH meter, 1001) during each experiment. To investigate the non-Newtonian behavior of blood, the apparent viscosity as a function of the shear rate was measured using a cone-plate viscometer (Brookfield, RVDV111-CP) coupled to a heated water bath to maintain a constant temperature of 37°C.

All experiments were performed in a closed steady flow loop model. The flow conduit was a long cylindrical Plexiglas tube with an outer diameter of 19.05 mm and an inner diameter of 15.87 mm. The distance between the entrance of the conduit and the location of ultrasound insonification was 50 cm, allowing a fully developed velocity profile at the site of Doppler recordings. A vertical tube was selected to avoid sedimentation effects. Instead of using gravity driven flow between two reservoirs, a damping filter was used in the flow model to eliminate the oscillations produced by the peristaltic pump (Lung–Heart Peristaltic Pump, Sarn, Inc.). The flow rate was then controlled directly by the speed of the pump. A cannulating type flow probe (model SF616) coupled to an electromagnetic flowmeter (Cliniflow II, FM701D, Carolina

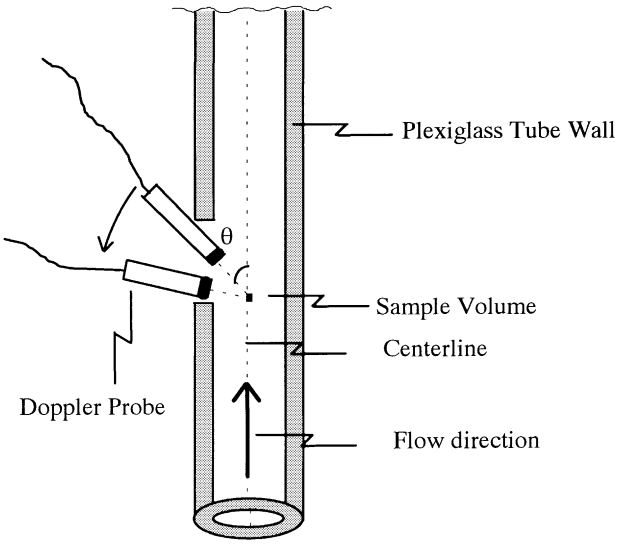


Fig. 1. Schematic diagram of the test section. The Doppler probe, which is free to rotate around the centerline of the tube, is illustrated.

Medical Electronics) served to monitor, on a Hewlett-Packard digitizing oscilloscope (model 54503A), the flow rate at the entrance of the tube. Measurements were performed for flow rates ranging between 0.3 L/min to 3.6 L/min (mean shear rate across the tube ranging from 8.5 s^{-1} to 102 s^{-1}, assuming a laminar parabolic velocity profile[2]). Blood was circulated in the model for at least one hour to eliminate air bubbles and raise blood temperature to 37°C before making Doppler measurements. The temperature was maintained by circulating hot water in the double-walled filling reservoir containing blood.

Blood echogenicity as a function of the Doppler angle and flow rate was evaluated with a single-gate 10-MHz Doppler system developed at Baylor College of Medicine, Houston, TX. The duration of the transmitted ultrasonic bursts was 0.8 μs and that of the gated returned echoes was 0.3 μs, which provided a sample volume length of approximately 0.86 mm [13], assuming a speed of ultrasound in blood of 1570 m/s. The pulse repetition frequency (PRF) was set to 19.5 kHz while the wall filter cut-off frequency was sufficiently low (3 Hz) to allow low Doppler frequency shifts to be measured. The frequency response of the Doppler flowmeter was characterized with a Brüel and Kjaer audio analyzer (model 2012) and compensated numerically to get a flat response between 3 Hz and PRF/2.

An angular positioner was used to hold and rotate the 1.5-mm diameter nonfocused Doppler probe which was free to rotate at angles varying from 40 to 80° between the tube axis and the ultrasonic beam (Fig. 1). To eliminate the strong attenuation effect from the Plexiglas tube, the transducer was positioned directly in the flow stream via a small rectangular window cut in the tube. The sample volume was localized upstream of the transducer tip to minimize flow disturbances caused by the intrusion of the transducer. To allow no blood

[1] The partial (γ_D) and total (γ_S) disaggregation thresholds (the shear rate needed to disrupt rouleaux) for human whole blood ($N = 17$) were 46 ± 4 s^{-1} and 114 ± 20 s^{-1}, respectively. For porcine whole blood ($N = 22$), $\gamma_D = 48 \pm 11$ s^{-1} and $\gamma_S = 135 \pm 30$ s^{-1}, while for calf whole blood ($N = 10$), $\gamma_D = 22 \pm 10$ s^{-1} and $\gamma_S = 29 \pm 18$ s^{-1}.

[2] The shear rate within the Doppler sample volume was not determined because it would require the knowledge of the true velocity profile across the tube. For a laminar parabolic velocity profile, the shear rate within a small sample volume located at the center of a tube is significantly less than the shear rate averaged over the cross section of the tube.

leakage through the window, a flexible neoprene membrane was fixed to the tube and tied around the Doppler transducer. Because the rotation center of the angular positioner was adjusted to coincide with both the position of the sample volume and the centerline of the tube, the ultrasonic path length was kept constant as the Doppler angle was changed. By doing so, the backscattered power measurements at different angles were always measured at the center of the tube with a constant sample volume dimension. The effect of blood attenuation due to the path length was also kept constant by this method. The depth of penetration of the ultrasonic pulses was approximately 11.3 mm (time delay of 14.4 μs) for all measurements, which was beyond the near field zone of the transducer. The beam profile of the transducer was axially symmetric, as experimentally determined using a hydrophone. At 11.3 mm from the face of the transducer, the diameter of the beam profile at -3 dB was 1.35 mm.

B. Effect of the Hematocrit of Porcine Whole Blood

Since the hematocrit has an important effect on the RBC aggregability [12], the effect of this parameter on the Doppler backscattered power from porcine whole blood was specifically studied. All Doppler measurements were performed at flow rates ranging from 0.3 to 1.8 L/min (mean shear rate across the tube ranging from 8.5 to 51 s^{-1}) according to the protocol described in Section II-A. The hematocrit values tested were 5, 10, 20, and 40%. Due to the large plasma quantity required at low hematocrit, plasma volumes from two different pigs were mixed together for each experiment. The experiments performed at 40% hematocrit were repeated here to assess the effect of mixing blood from two animals.

C. Measurements Using Carbon Fibers Suspended in a Water-Glycerol Mixture

A last series of measurements was performed using cylindrical carbon fibers (Goodfellow, PA, USA) to simulate the presence of rouleaux of red cells. The objective of these measurements was to verify that the carbon fibers aligned themselves in the direction of the flow, and that the echogenicity was maximum when the direction of the sound wave was perpendicular to the alignment of the fibers. These fibers, which had a mean radius of 7 μm and a mean length of 250 μm, were similar to those used by Mottley *et al.* [14] in a tissue-mimicking phantom used to study the anisotropy of the ultrasonic properties of myocardial tissue. In the present study, the carbon fibers were suspended in a test fluid made of a volume of 40% glycerol and 60% saline mixture. The water-glycerol mixture (without the fibers) had Newtonian properties since the apparent viscosity was relatively constant for shear rates varying between 4 and 538 s^{-1} (3.14 $\pm$ 0.16 cP at room temperature). Relatively low concentrations (0.4, 0.8, and 1.6 g/L) of carbon fibers were tested in order to get a backscattered power comparable to that measured with blood. All measurements were carried out at 23°C and at flow rates of 0.3, 0.6, and 0.9 L/min (mean shear rate across the tube of 8.5, 17, and 25 s^{-1}). The remaining details of the protocol were the same as those described in Section II-A.

D. Spectral Analysis and Computation of the Doppler Backscattered Power

The in-phase and quadrature demodulated Doppler signals were digitized for five seconds on an IBM-PC compatible with a 12-b resolution digitizing board (Data Translation, DT2821G-SE) at a sampling rate of 19.5 kHz. The blood flow signal obtained from the electromagnetic flowmeter was also sampled. A 10-ms Hanning window was applied to the Doppler signals and a 512-sample fast Fourier transform (FFT) was computed. The Hanning window was then slid over the Doppler signals by increments of 10 ms, and 500 consecutive spectra were evaluated. From these spectra, a mean Doppler spectrum was calculated. The mean Doppler power, in decibels, of both the forward and reverse components of the mean spectrum was determined in the frequency domain according to the following:

$$\text{mean Doppler power} = 10 \log \frac{\sum_{f_k=-\text{PRF}/2}^{\text{PRF}/2} P(f_k)}{N} \quad (3)$$

where $P(f_k)$ represents the power at frequency f_k, and $N = 512$ is the number of samples between $\pm$PRF/2. The forward mean Doppler velocity was also computed using

$$\text{forward mean Doppler velocity} = \frac{c}{2F_0 \cos\theta} \frac{\sum_{f_k=-BW}^{BW} f_k P(f_k)}{\sum_{f_k=-BW}^{BW} P(f_k)} \quad (4)$$

where $c = 1570$ m/s is the speed of ultrasound in blood, $F_0 = 10$ MHz is the Doppler transmitted frequency, θ is the insonification angle, and BW and $-BW$ are the higher and lower frequencies of the -5 dB bandwidth of the dominant forward frequency peak, respectively.

III. Results

A. Measurements Using Calf and Porcine Blood

For the first series of experiments using calf and porcine blood, all measurements were repeated four times on different days and with different blood samples. On average, the hematocrit of porcine whole blood was 40.1 $\pm$ 0.4%, the blood temperature was 37 $\pm$ 1°C, and the pH was 6.7 $\pm$ 0.1. For calf RBC suspensions, the hematocrit was 38.1 $\pm$ 1.6%, the temperature was 37 $\pm$ 1°C, and the pH was 7.0 $\pm$ 0.1. The mean viscosity for the calf erythrocyte suspension and the porcine whole blood as a function of the shear rate is presented in Fig. 2. The standard deviations of each measurement are also shown. It can be observed that calf erythrocyte suspension behaved like a Newtonian fluid in the range of shear rates tested whereas the viscosity of porcine whole blood significantly increased at low shear rates due to the formation of RBC aggregates.

Fig. 3 shows examples of Doppler mean spectrum recorded at an angle of 45° and at three different mean shear rates for calf erythrocyte suspension and porcine whole blood. For

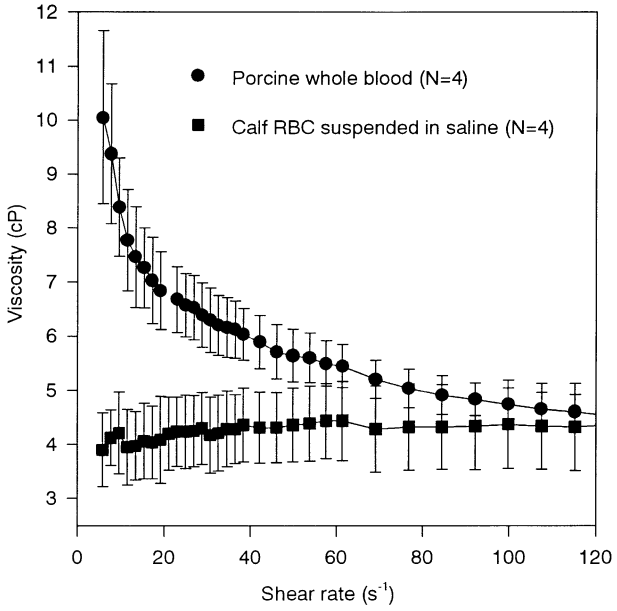
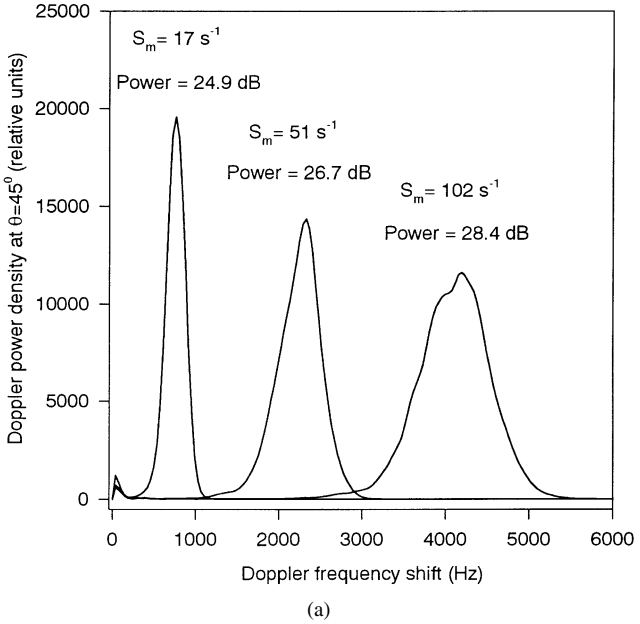


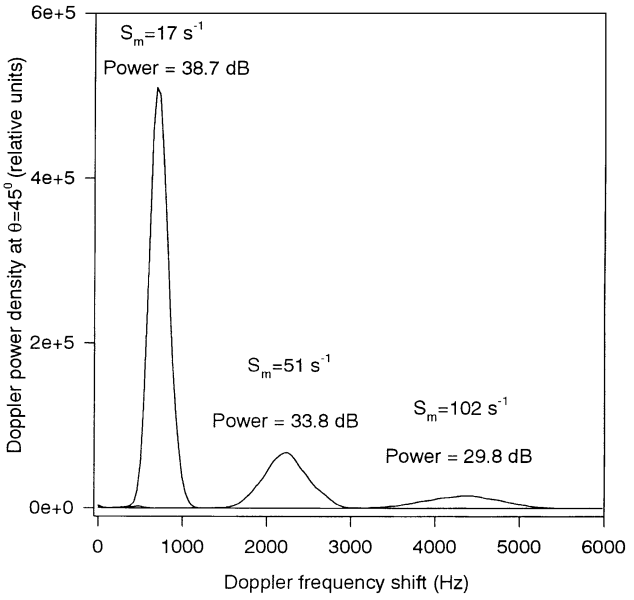
Fig. 2. Apparent viscosity as a function of the shear rate for porcine whole blood and saline suspensions of calf red blood cells at 40% hematocrit and 37°C.

porcine whole blood [Fig. 3(b)], an important increase of the Doppler power density was observed as the shear rate decreased, while for the calf erythrocyte suspension [Fig. 3(a)], less important changes were found. To verify that the position of the sample volume within the tube was independent of the insonification angle, the mean Doppler velocity, as given by (4), was evaluated for the different insonification angles and mean shear rates. Results are reported in Fig. 4. It can be seen that the Doppler mean velocity was relatively constant as a function of the angle, with the exception of the results obtained at high shear rates.

The Doppler power for the calf erythrocyte suspension and the porcine whole blood, as evaluated from (3), is depicted in Fig. 5 as functions of the insonification angle and mean shear rate. For calf erythrocyte suspension, an increase in the Doppler backscattered power of 5 dB was observed when the mean shear rate increased from 8.5 to 102 s^{-1}. No angular dependence of the backscattered power was observed. For porcine whole blood, an increase of the power by more than 16 dB was observed when the mean shear rate decreased from 102 to 8.5 s^{-1}. Angular dependencies of the backscattered power were found, especially for moderate mean shear rates (17 to 51 s^{-1}). For a mean shear rate of 102 s^{-1}, the backscattered power slightly decreased (≈2 dB) when the insonification angle increased from 40 to 80°. For mean shear rates of 51 and 25 s^{-1}, the backscattered power slightly increased between 40 and 45°, and then decreased (≈5 dB) for higher insonification angles. Similar angular dependence was also present for a mean shear rate of 17 s^{-1}, while for the lowest mean shear rate (8.5 s^{-1}), the backscattered power was relatively constant. It can be observed from Fig. 5 that at low shear rates, the standard deviations (variation from experiment to experiment) are more important for porcine whole blood than calf red cell suspension. A similar observation was also notified by Yuan and Shung [5] and the proposed explanation was the



(a)



(b)

Fig. 3. Typical mean spectra for mean shear rates across the tube of 17, 51, and 102 s^{-1} (mean flow rates of 0.6, 1.8, and 3.6 L/min, respectively). (a) Results for saline suspensions of calf red blood cells. (b) Results for porcine whole blood. The Doppler power of each spectrum in decibels is also shown.

variation of fibrinogen concentration among blood samples. In the present study, the concentration of fibrinogen and other plasma proteins affecting RBC aggregation [15] was not controlled.

B. Effect of the Hematocrit of Porcine Whole Blood

Results of the effect of the hematocrit on the angular dependence of the backscattered power for porcine whole blood are summarized in Fig. 6(a)–(d). For this series of experiments, all measurements were repeated three times and averaged. At 5% hematocrit, no angular dependence was observed for the mean shear rates tested (the range of power variations was always less than 2 dB). However, there was an

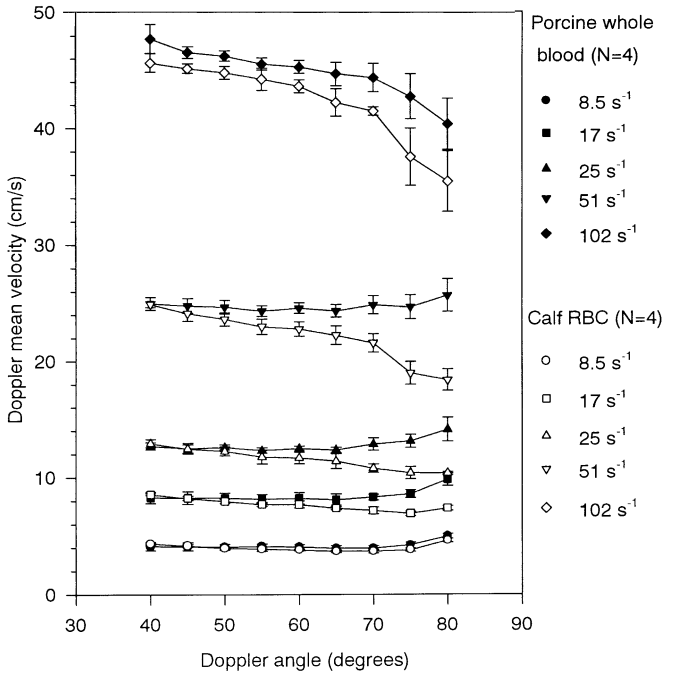


Fig. 4. Doppler mean velocity as a function of the insonification angle for porcine whole blood and saline suspensions of calf red blood cells. Results averaged over four experiments are presented for mean shear rates of 8.5, 17, 25, 51, and 102 s^{-1}.

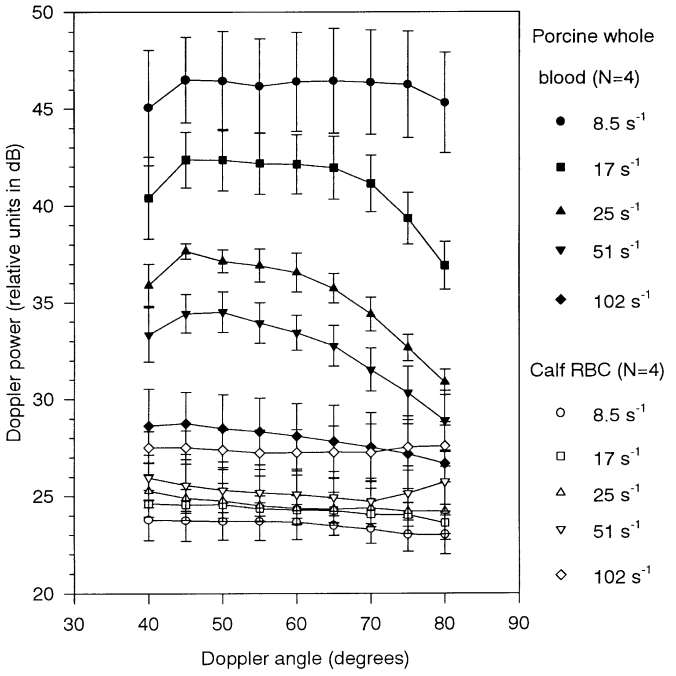


Fig. 5. Doppler power as a function of the insonification angle for porcine whole blood and saline suspensions of calf red blood cells. Results averaged over four experiments are presented for mean shear rates of 8.5, 17, 25, 51, and 102 s^{-1}.

increase in the backscattered power by approximately 7 dB when decreasing the mean shear rate from 51 to 8.5 s^{-1}. As the hematocrit increased, the angular dependence progressively appeared. At 10% hematocrit, the range of variations of the power as a function of the angle was around 2 dB. At 20% hematocrit, these variations increased to about 4 dB. At 40% hematocrit, the angular dependence was the most evident and was similar to those obtained in Fig. 5 when no mixing of the plasma from two different animals was done. This confirmed that the mixing of the plasma from two pigs did not alter the aggregation tendency of the red cells. The range of variations of the backscattered power as a function of the angle were around 4 dB at mean shear rates of 51 and 25 s^{-1}, 5 dB at a mean shear rate of 17 s^{-1}, and 3 dB at a mean shear rate of 8.5 s^{-1}.

C. Measurements Using Carbon Fibers Suspended in a Water–Glycerol Mixture

The results of the measurements using carbon fibers as the scattering particles are presented in Fig. 7. It can be seen that the angular dependence of the carbon fibers was completely different from that obtained with blood since, for the three mean shear rates tested, the backscattered power (in decibels) increased linearly with the insonification angle. The maximum (at 80°) to minimum (at 40°) changes in the Doppler power were approximately 8 dB. Similar Doppler power variations were obtained when increasing the carbon fiber concentration (results not presented).

IV. Discussion

Results of Fig. 4 indicated small variations in the Doppler velocities for high shear rates and large insonification angles, especially for calf red cell suspension. It is expected that these variations were due to the variations of the velocity profile as the Doppler angle was changed. For small Doppler angles, the intrusion of the Doppler transducer within the tube was minimal, while for larger Doppler angles, it was a bit deeper, which may have skewed the velocity profile and increased the centerline blood velocity. Clearly, this effect increased as the shear rates increased and was more important for the calf red cell suspension than for the porcine whole blood (the reason for this difference is unknown). Nevertheless, as shown in Fig. 5, these small velocity variations at high shear rates had no influence on the results of Doppler backscattered power. For instance, no angular dependence in the Doppler power was observed at high shear rates for both the calf red cell suspension and porcine whole blood.

The first experimental study showing an increase of the ultrasonic backscattering coefficient in the presence of flow disturbance was reported by Shung *et al.* [16]. Since then, studies by different groups [3], [17]–[20] investigated the effect of turbulence on the Doppler backscattered power. With erythrocyte suspensions at 40% hematocrit, power increases of 3 and 5.5 dB were found downstream of a grid under steady [3] and pulsatile flow [17], respectively, while an increase of approximately 3 dB was measured downstream of severe stenoses for both steady [18] and pulsatile flows [19], [20]. In the present study, a power increase of 5 dB was observed for calf RBC suspensions when increasing the mean shear rates from 8.5 to 102 s^{-1} (Reynolds number from 92 to 1100, approximately). Although the turbulence was not generated in the same way in all these studies, there is a strong consistency between the observed results.

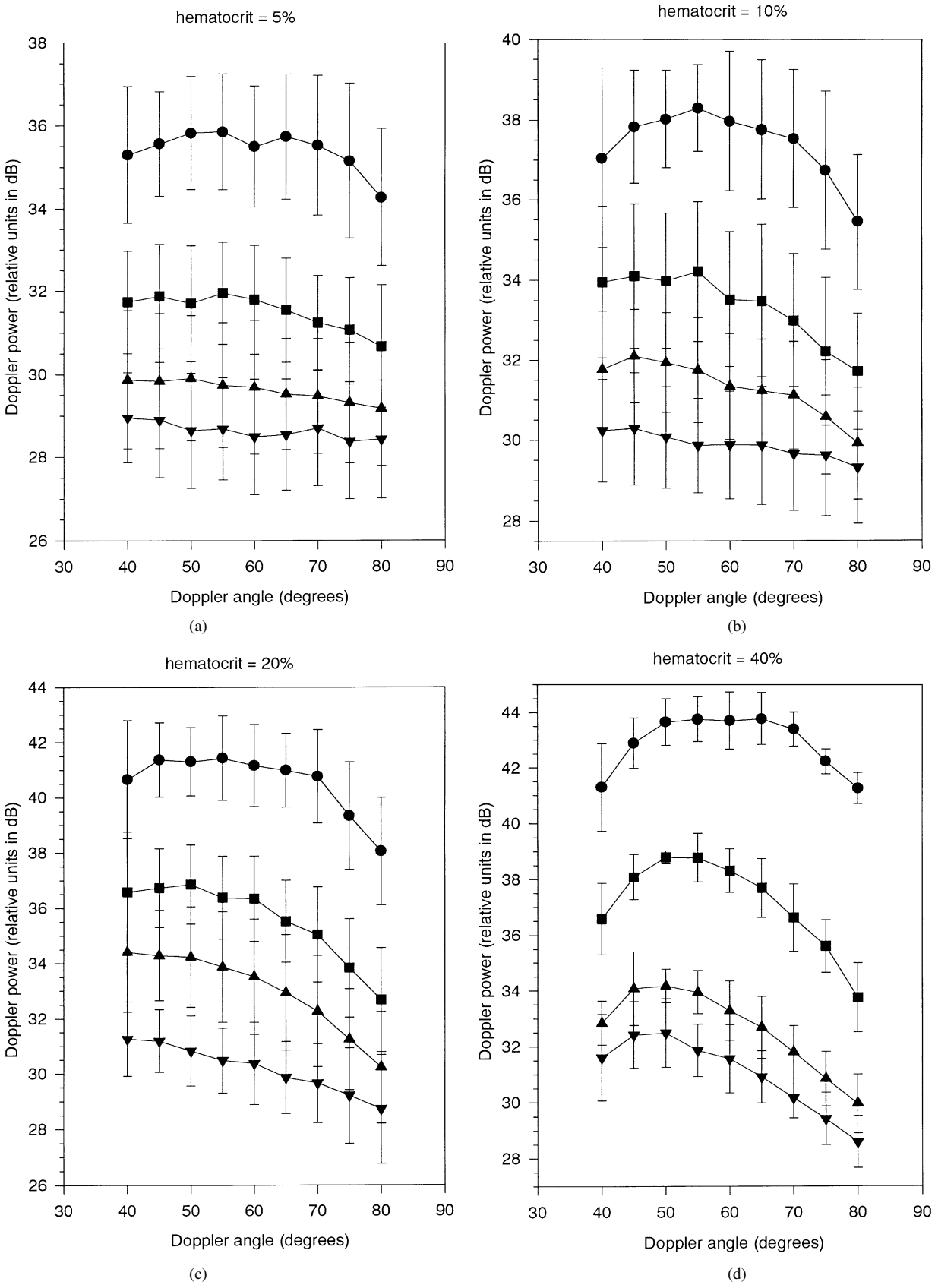


Fig. 6. Doppler power as a function of the insonification angle for porcine whole blood: (a) hematocrit of 5%, (b) hematocrit of 10%, (c) hematocrit of 20%, and (d) hematocrit of 40%. Results averaged over three experiments are presented for mean shear rates of 8.5 s^{-1}(•), 17 s^{-1}(■), 25 s^{-1}(▲), and 51 s^{-1}(▼).

At the high shear rates, both the calf red cell suspension and porcine whole blood have similar Doppler backscattered power levels (between 27 and 29 dB). However, as the blood flow rates decreased, the Doppler power for porcine whole

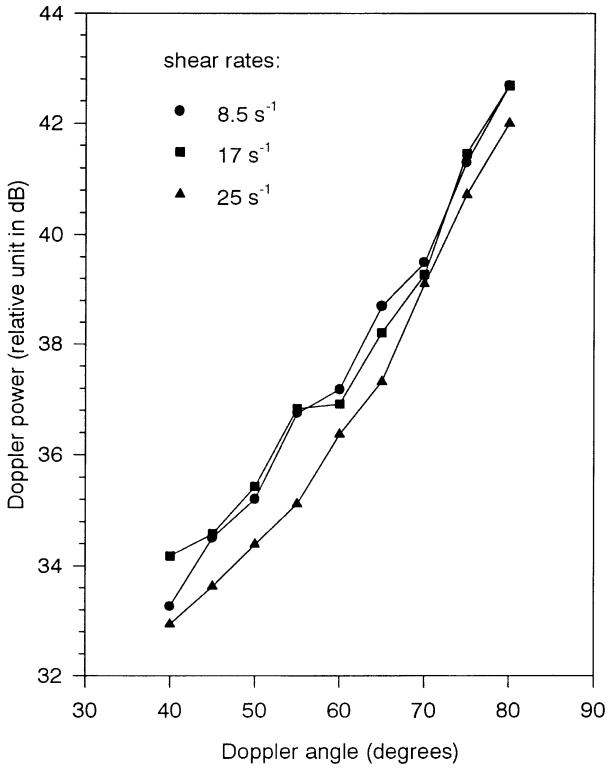


Fig. 7. Doppler power as a function of the insonification angle for cylindrical carbon fibers suspended in a water-glycerol mixture at a concentration of 0.4 g/L.

blood increased significantly due to the formation of red cell aggregates. An angular dependence was also observed. For calf red cell suspension, no aggregate was formed as the flow rate decreased and, consequently, the Doppler power decreased as the flow disturbance disappeared.

A. Anisotropic Effect

To explain their results on the cyclic variations of blood echogenicity, de Kroon *et al.* [21] as well as Cloutier and Shung [22], [23] summarized in their discussions the different factors that may influence the echogenicity of flowing blood. Among these factors, they stated that the shape and orientation of the aggregates may influence echogenicity. Because rouleaux align themselves in the direction of the flow [24], they postulated that higher echogenicity should be observed when the direction of the sound wave is perpendicular to the alignment of the rouleaux. This hypothesis was based on the fact that RBC circulating in a relatively large tube at low shear rates form long cylindrical straight, and sometimes branched, structures called networks.

It has been shown for many years that the ultrasonic properties from biological tissues containing directionally oriented structures are angular dependent. Angular dependence, or anisotropy, in the measurement of the attenuation and backscattering coefficients was demonstrated for the myocardium [14], [25]–[30], skeletal muscles [25], [31], kidney [32], [33], arterial wall [34], and human Achilles tendon [35]. Although the measurement techniques and sample preparation were generally different in each of these studies, it was generally observed that the backscattered power was maximum for a perpendicular ultrasonic incidence angle and minimum for a parallel incidence. On the other hand, it was demonstrated that these tissues had a larger attenuation coefficient when ultrasound traveled parallel to the fibers and a smaller one when ultrasound traveled perpendicular to the fibers. Although these studies did not provide a complete explanation of the structure and mechanism responsible for the ultrasonic scattering and attenuation in the above biological tissues, they underscored the importance of taking anisotropy into account in the interpretation of quantitative results in ultrasonic tissue characterization.

Our results using carbon fibers (Fig. 7) are consistent with the theoretical and experimental results for biological tissues containing directionally oriented structures. They confirmed the alignment of the fibers with the direction of the flow since the minimum and maximum scattering occurred at 40 and 80°, respectively. Based on the Rayleigh scattering theory, no angular dependence is anticipated for porcine whole blood at high shear rates and saline suspensions of calf red blood cells since no large rouleaux can develop. This was confirmed in the experimental results of Fig. 5. The angular dependence observed for porcine whole blood at low and moderate shear rates (Figs. 5 and 6) cannot, however, be explained by the scattering models available (either the RBC or tissue scattering models).

B. Explanations of the Anisotropy Based on the Structure of the Aggregates

The structure (shape and size) of the red cell aggregates is probably the most important factor that governed the observed anisotropy of the Doppler backscattered power. To validate this assumption, direct visualization of the structure of the aggregates within a large tube would have to be realized. Currently, it is recognized that no method is capable of completely describing the aggregation kinetics and aggregate morphology [36]. This observation is particularly true for flowing blood in large vessels. Some of the observations available so far on the aggregation process came from microcirculation studies. For instance, Goldsmith *et al.* [24], [37], [38] observed for a dilute suspension of human RBC flowing through narrow glass tubes, that single red cells and rouleaux undergo angular rotation. Depending on the length of the rouleaux and the value of the shear rate, the aggregates may also bend while rotating. For higher cell concentrations, red cells no longer undergo angular rotation, but are generally aligned in the direction of flow. They are also deformed into a variety of shapes as a result of particle crowding. Deformation of rouleaux is also observed and this phenomenon occurs even at very low shear rates.

Observations using a transparent cone-plate viscometer demonstrated that the size of the aggregates was dependent on the shear rate [39]. The hematocrit was also shown to influence the kinetics and the extent of RBC aggregation [12], [40]–[42]. Similarly, it was shown in many studies [3]–[5], [43] that ultrasound backscattering from porcine whole blood is also affected by the shear rate (size of the scatterers) and the

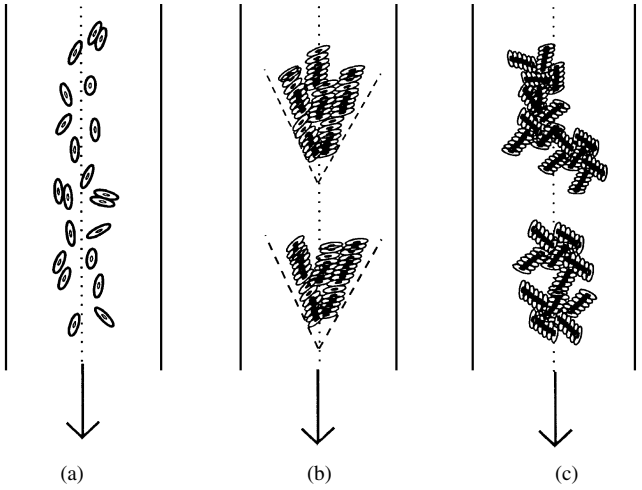


Fig. 8. Schematic representation (relative scale) of the structure of the aggregates for high, moderate, and low shear rates conditions across the tube. (a) High shear rate conditions prevented the formation of large aggregates, red cells were generally monodispersed and aligned in the direction of the flow, and the Doppler backscattered power was independent of the insonification angle. (b) At moderate shear rate conditions, cone-shaped aggregates are hypothesized since the Doppler backscattered power was maximum for angles between 45 and 60°. (c) Low shear rates conditions promoted the formation of large and complex aggregates and no important angular dependence was observed.

hematocrit. These observations were confirmed by the results presented in Figs. 5 and 6. These figures also indicated that the angular dependence of the Doppler power was a function of the hematocrit and was affected by the extent of RBC aggregation. This suggests that the structure of the red cell aggregates may be the dominant factor to explain the observed anisotropy of the Doppler power.

Based on observations above and our results, it seems that the modelization of the aggregates as long cylindrical chains of red cells is probably not exactly true. Consequently, the following hypothesis on the structure of the aggregates is proposed. It is reasonable to believe that low shear rate (8.5 s^{-1}) conditions may promote the formation of large and 3-D complex structures (clumping). Under this flow condition, no angular dependence was present. At moderate shear rates (17 to 51 s^{-1}), smaller and deformed (cone-shaped) aggregate structures may be present at the center of the tube, and this shape may have promoted the angular dependence. At high shear rates (102 s^{-1}), no rouleaux was formed and no angular dependence was observed. The three situations are illustrated in Fig. 8.

V. Conclusion

Recent technological improvements in color Doppler flow imaging allowed a color map display of the integrated power of the Doppler signal. In preliminary reports, Rubin [44], [45] compared this new imaging modality, called power Doppler, to the conventional color Doppler ultrasonography that is based on the estimation of the mean frequency shift. It was reported that the most important advantages of the power Doppler over the conventional color Doppler were its frequency aliasing free capability, its increased sensitivity to flow due to the extended dynamic range of the method, and its angle independence (except for orthogonal low flow velocity where the near zero frequency shift signals are eliminated by the clutter filter).

The theoretical angle-independent nature of this new modality was verified in the present study by evaluating, in some specific shear rate conditions, the effect of the insonification angle on the Doppler backscattered power from porcine whole blood, saline suspension of calf red cells, and carbon fibers. It was shown that an angular dependence exists for porcine whole blood and carbon fibers. These results were not, however, predicted by the actual scattering models for red blood cells that assume no anisotropic effect. Our results were also not conveniently explained by the scattering models developed for tissues having cylindrical oriented fibers. Further work will be necessary to get a better understanding of the anisotropic effect observed in the present study and a more precise evaluation of its possible impact on the Doppler power imaging measurements performed *in vivo*.

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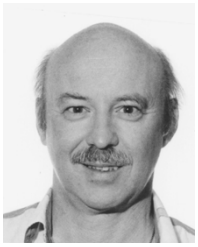

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