Cyclic Variation of the Power of Ultrasonic Doppler Signals Backscattered by Polystyrene Microspheres and Porcine Erythrocyte Suspensions

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Abstract—**Factors affecting the power of the ultrasonic Doppler signal within the flow cycle have been evaluated experimentally using a pulsatile flow loop model. Polystyrene microspheres and porcine red cells suspended in saline solution for hematocrits between 2 and 40% were used as scattering fluid in the flow model. Experiments were performed at mean flow velocities of 11, 64, and 76 cm/s. In laminar flow experiments performed at a mean velocity of 11 cm/s, no variation of the Doppler power was found for both polystyrene microspheres and red cell suspensions (40% hematocrit). When turbulence was induced in the flow model, the power increased during systole, a maximum was observed early after peak systole, and a decrease was obtained in diastole during deceleration of flow. At higher mean flow velocities (64 and 76 cm/s), a significant cyclic variation of the Doppler power was also measured for all values of hematocrits (between 2 and 40%). The power of the signal scattered by microspheres and red cell suspensions at 4% hematocrit dropped in systole, reached a minimum at peak systole, and then increased during early diastole. For red cells suspended in saline at 40% hematocrit, a slightly different pattern of variation was obtained. The cyclic variations observed at high flow velocities and in the presence of turbulence are believed to be associated with cyclic changes in the correlation among particles. In the present study, the effect of red cell aggregation on the cyclic variation has not been addressed.**

I. Introduction

THE controversy as to whether ultrasonic Doppler power from blood changes during a cardiac cycle prompted us to initiate a series of investigations to clarify some aspects of this issue using a pulsatile flow loop model. In a "Letter to the Editor" published in 1987, Luckman *et al.* [1] found no power change within the cardiac cycle from a pulsed-wave (PW) Doppler signal at 8 MHz recorded in the carotid artery of a healthy male adult. The objective of the letter was to clarify an undercurrent of belief that the power contained in ultrasound Doppler signals was in some way velocity dependent. Observations of changes in Doppler backscattering power and echogenicity of blood within the cardiac cycle were made recently in *in vivo* studies [2]–[4]. Using the continuous-wave (CW) Doppler ultrasound technique, Thompson *et al.* [2], [3] showed a rise in the power of the signal at the time of peak systole, thereby suggesting the possibility that useful diagnostic information might be extracted from the variation. These results obtained from 12 normal umbilical arteries were partially attributed to the increase in the number of red cells in the region of insonation during dilation of the vessel. A recent analysis based on intravascular imaging of blood at 30 MHz also showed a cyclic variation of the backscattered signals [4]. However, a different pattern of variation was obtained from measurements performed over the iliac artery of three patients. The echogenicity of blood decreased rapidly in early systole and increased during diastole. In their study, De Kroon *et al.* [4] attributed the variation to changes in the state of erythrocyte aggregation.

With a pulsatile flow loop model. Bascom *et al.* [5] investigated possible variations in Doppler backscattering power from blood using a rigid tube to eliminate possible changes associated with vessel dilation. Fixed heparinized human red cells at a hematocrit of 42% were circulated in the model. The fixation process has the advantage of eliminating red cell aggregation. Results obtained with a CW Doppler flowmeter with a carrier frequency at 5 MHz showed no significant change within the flow cycle. However, a preliminary study carried out by our group [6] showed that such significant variations could be present in pulsatile flow.

In order to resolve this controversy, factors that may contribute to the cyclic variation were studied separately. The effect of vessel dilation is not considered in the present study because a rigid tube and the PW Doppler technique have been used. The effect of red cell aggregation has also been eliminated by using nonaggregating polystyrene microspheres and red cell suspensions as scattering media. We have concentrated our efforts on other factors such as hematocrit, changes in mean and maximal flow velocities, and the presence of blood flow turbulence. It will be shown that the power of the Doppler signal varies as a function of the timing within the flow cycle when flow velocity is high or when there is flow disturbance. It will also be shown that the hematocrit of the blood sample affects the manner how the Doppler power varies during the flow cycle.

II. Background

In the present section, a brief review of the known mechanisms affecting the ultrasonic backscattering power from blood

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will be presented. This review will serve to derive possible factors that may be used later to explain the observed cyclic variation in both laminar and turbulent pulsatile flow.

Different theoretical models have been proposed to explain the echogenicity of blood. Both particle [7], [8] and continuum [9] models were suggested. A comprehensive review of those models along with a new unified theory on the backscattering of Doppler ultrasound from blood has been presented by Mo [10]. In a recent study performed by our group [11], a correction to the assumption commonly use in the literature [12] to characterize the Doppler spectral power density at a given frequency was proposed. Based on the particle model, the power contained in the frequency range from ω to $\omega + d\omega$ in the Doppler spectrum was suggested to be given by

$$P(\omega)\, d(\omega) = \frac{\sigma_b H W V(\nu)\, dv}{V_c} \tag{1}$$

where the backscattering cross section of a red cell σ_b is given by

$$\sigma_b = \frac{\pi^2 V_c^2}{\lambda^4}\left[\frac{K_e - K_o}{K_o} + \frac{\rho_o - \rho_e}{\rho_e}\right]^2. \tag{2}$$

In (1), H represents the hematocrit, W the packing factor which is a variable that considers the interaction among ultrasonic scatterers, V_c the volume of a red blood cell, and $V(v)\, dv$ the volume of red cells moving with a velocity within v and $v + dv$. In (2), λ is the wavelength of the transmitted ultrasonic wave, K the compressibility, and ρ the density of red cells (subscript e) and plasma (subscript o).

Over the last 30 years, several forms of the packing factor W have been proposed in the literature. Twersky's earlier theory [13] which assumed independent scatterers has been proven to be invalid for blood of hematocrits higher than a few percent [14]. Later, assuming pair-correlated particles [7], W was shown to be a function of hematocrit, as predicted by experimental results. Recently, other versions of the packing factor were proposed [15]–[17] to consider: 1) the experimental observations of an increase in the backscattering power in the presence of red cell aggregation and blood flow turbulence, 2) the shift of the scattering peak in turbulent flow from 13% to higher hematocrits [11], [18], and 3) to correct for the discrepancy at high hematocrits between the experimental backscattering power and that predicted by the previous versions of the model [8], [19]. Twersky [16] obtained a modified packing factor based on the characterization of the fluctuations of particle number in the sample volume from scaled particle statistical mechanics theory for mixtures of similar-shaped but different-sized scatterers. He introduced two new parameters in his model, parameter c which considers the shape and correlation among particles, and parameter d which represents the variance of the particle size. This new model indicates that in isotropic fluids suspended with a mixture of hard convex particles ($c > 3$) of different size ($d \neq 0$), W is given by [16], [17]

$$W = \frac{H(1-H)^2}{[1+(c-1)H]^2} \cdot \left[(1-H)^2 + \frac{4cdH(1-H)}{1+5d} + \frac{H^2c^2d}{1+4d}\right]. \tag{3}$$

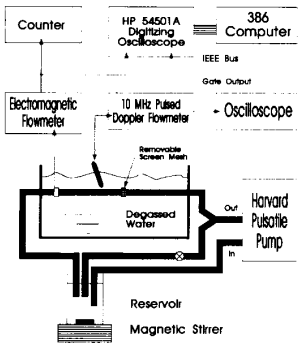

Fig. 1. Experimental block diagram of the flow loop model.

The following observations can be made from (3). Parameter c is first influenced by the shape of the particles. A value higher than 3 was found for convex particles such as red blood cells, $c = 3$ corresponds to hard spheres, and $c < 3$ was suggested to model red cell aggregates [19]. Parameter c is also influenced by the correlation among particles. A reduction of c corresponds to less correlation or higher fluctuation such as seen in turbulent flow, while an increase represents more correlation or lower fluctuation. For a fixed value of the variance of particle size d, reducing c tends to increase the backscattering power and shift the maximum of the power versus hematocrit curve toward a higher hematocrit. A similar effect can be obtained by increasing d for a fixed value of c.

III. Materials and Methods

A. Flow Loop Model

The schematic representation of the flow loop model used in the present study is illustrated in Fig. 1. The pulsed Doppler flowmeter developed by Hartley [20] was used to transmit and receive Doppler signals. The flowmeter was operated with a 10 MHz nonfocused transducer fired at a pulse repetition frequency (PRF) of 40 kHz. The angle between the tube and the Doppler probe was maintained at 70° for all measurements. Bandpass filtering of the received echoes between 100 Hz and 20 kHz was performed by the flowmeter. Within that band, a small decrease with frequency of the amplitude of the frequency response was observed (less than 3 dB). The transfer function of the flowmeter was then corrected numerically by compensating each Doppler spectrum. This was performed by multiplying the Doppler frequency components by the inverse of the transfer function. A flat response was then obtained between 0 Hz and PRF/2 to eliminate possible artifacts, in backscattered power measurements, due to the Doppler system.

A square-wave electromagnetic flowmeter (Carolina Medical Electronics, Model 501) was used along with a blood flow probe (In Vivo Metric, Model K) to monitor the instantaneous flow rate within the tube. A steady laminar flow arrangement as the one described by Shung *et al.* [11] was used to calibrate the flowmeter with red cell suspensions at 4 and 40%

hematocrit, and saline solution mixed with 35% glycerol. The calibration was performed by collecting a measured volume of solution over a known period of time. The output signal from the electromagnetic flowmeter was used to synchronize digitization of the Doppler signal and to monitor the pulsation rate of the Harvard pulsatile pump via a Hewlett–Packard (HP) counter. Only the forward output of the Doppler flowmeter was analyzed in the present study. Both the forward Doppler and flow rate data were digitized by a HP 54501A digitizing scope to 8 b. Acquisition of the Doppler signals at different phases within systole and early diastole was realized by triggering the flow rate signal and adjusting the delay of the HP oscilloscope. An IEEE-488 interface board (Metrabyte, Model MBC-488) was used to transfer the digitized data to a 386 personal computer.

The main flow conduit was a cylindrical high-pressure polyethylene tube with an outer diameter of 0.635 cm, an inner diameter of 0.476 cm, and a length of 90 cm. The distance between the site of measurement of the Doppler signals and the entrance of the polyethylene tubing was approximately 50 cm. The flow conduit was immersed horizontally in a water tank, and plastic fittings were used on both sides of the tube to close the loop with larger sized Tygon tubing. To reduce the high flow rate of the Harvard pulsatile pump (Model 1423), a shunt tube fitted to a valve was connected between the output of the pump and the reservoir to adjust the flow velocity to the value needed at the site of measurement.

B. Position and Size of the Doppler Sample Volume

All Doppler measurements were performed at the center of the tube by gating the returned echoes 19 μs after firing the 0.3 cm diameter nonfocused transducer. The depth of penetration of the ultrasonic pulse was approximately 1.5 cm, which corresponded to the beginning of the far-field zone of the transducer. The gate output of the pulsed Doppler flowmeter was monitored with an oscilloscope, as seen in Fig. 1. To position the sample volume, the zero velocities associated with the near and far walls of the tube were determined from the audio feedback of the Doppler flowmeter and the corresponding range positions evaluated from the gate output. The Doppler probe was moved until the position corresponding to the mean of these two range-gate locations corresponded to the delay of 19 μs (1.5 cm).

Fig. 2 shows a diagram of the position and dimension of the Doppler sample volume. Its three-dimensional size was approximately 0.5 mm^3. The average diameter of the beam width, 1.5 cm from the face of the transducer, was 1.05 mm and the length of the sample volume was 0.55 mm. The -3 dB beam width was determined by using the pulse-Doppler probe as a pulse-echo A-mode probe. The transducer was then fired with bursts of 10 MHz ultrasound signals and the transmitted amplitude was detected in x, y, and z directions with an NP-1000 needle hydrophone (NTR Systems, Model TNU001A) mounted on a three-dimensional step motor arrangement. The length of the sample volume was computed with the following

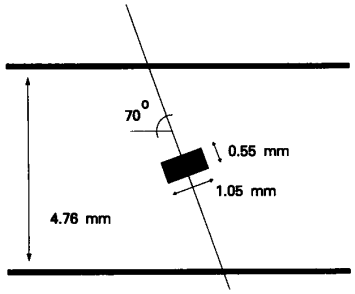

Fig. 2. Position and size of the Doppler sample volume.

equation [21]:

$$Z_r = \frac{c(t_g + t_p)}{2} \tag{4}$$

where c is the speed of ultrasound in blood (1570 m/s), t_g the duration of the gated returned ultrasonic signals (0.3 μs), and t_p the duration of the transmitted ultrasonic bursts (0.4 μs).

C. Generation of Blood Flow Turbulence

A mesh screen similar to that used in a previous study [11] was employed to generate turbulence. The mesh was positioned 2 cm upstream of the Doppler measurement site, as shown in Fig. 1. The mesh was removed for laminar flow experiments. The relative turbulence intensity [22] generated by the screen mesh was 2.74%, which is similar to the relative intensity used in [11].

D. Ultrasonic Scatterers

Polystyrene microspheres (Duke Scientific, diameters of 20–130 μm) and porcine red cells suspended in saline solution at hematocrits between 2 and 40% were used as scattering media. Polystyrene microspheres were suspended in a solution of 35% glycerol and 65% saline at a concentration of 1 g/L. Approximately 15 mL of surfactant was added to facilitate mixing of the particles with the solution. Red cell suspensions were prepared by first adding a solution of 1 g of ethylenediamine tetraacetic acid (EDTA) in 10 mL of saline to fresh porcine whole blood at a concentration of 30 mL per liter of blood to prevent coagulation. The day before the experiment, concentrated erythrocytes were obtained by centrifuging the whole blood and removing the plasma and the top white cell layer. The red cells were then washed and centrifuged two times with 0.9% normal saline buffered to pH 7.4. The concentrated red cells were then stored at 4°C in a hermetic container. Before starting the experiment, the washed concentrated erythrocytes were reconstituted with isotonic saline solution (Celline^{TM} II, pH 6.25) to the desired hematocrit. To prevent crenation of the red cells, a volume of 0.5% bovine albumin was added to the solution. The hematocrit was determined by reading the proportion of red cells from a microcentrifuge tube. Before taking Doppler measurements, the scattering medium was circulated in the flow loop for 1–2 h to eliminate bubbles and to raise the temperature of the solution to 22 ± 1°C. A continuous mixing

of the solution was performed by a magnetic stirrer, as shown in Fig. 1.

E. Doppler Data Acquisition and Computation of the Mean Power

The forward output of the Doppler flowmeter was digitized at a rate of 51.2 kHz over 100–200 cardiac cycles. After synchronizing the acquisition with the flow signal, a 10 ms Hanning window (512 samples) was applied and 1024-sample fast Fourier transforms (FFT) computed. Zero padding was performed before evaluating FFT's. Mean Doppler spectra were then calculated at different phases of the cardiac cycle by averaging spectra over 100 or 200 cycles. Averaging over 100 cycles was done in some situations, as specified later in figure captions, to reduce the duration of the experiments. By using such a large number of cardiac cycles, a very consistent estimate of the spectral properties could be obtained [23].

The mean power of the Doppler signals was determined in the frequency domain. The following equation was used to compute the mean power, in decibels, of the mean spectra:

$$\text{mean power} = 10 \times \log \frac{\sum_{f_k=0}^{\text{PRF}/2} P(f_k)}{N} \qquad (5)$$

where $P(f_k)$ represents the power at frequency f_k, N the number of samples between 0 Hz and PRF/2, and PRF the pulse repetition frequency.

IV. Experimental Results

Two instantaneous mean velocity versus time traces were used to study changes in Doppler backscattering power within the cardiac cycle for polystyrene microspheres and red cell suspensions of different hematocrits. An example of these curves are presented in Fig. 3 for red cell suspensions at 40% hematocrit. The first series of experiments was performed at a mean flow velocity calculated over the cardiac cycle of 11 cm/s and a maximal flow velocity of approximately 60 cm/s (see panel (a) of Fig. 3). This physiological flow profile is similar to that found in the normal human femoral artery [24]. Another series of experiments was performed at higher mean velocities (64 and 76 cm/s). The situation presented in panel (b) of Fig. 3 corresponds to a mean velocity of 64 cm/s and a maximal velocity of approximately 200 cm/s. A pulsation of 70 beats/min was used for all measurements.

Results obtained with polystyrene microspheres are presented in Fig. 4. Each data point corresponds to the average of five experiments performed with different mixtures of microspheres, glycerol, and saline solution. No significant cyclic variation of the mean power (one-way analysis of variance with repeated measurements, $p = 0.994$) was obtained at a mean flow velocity of 11 cm/s [panel (a)]. However, increasing the mean velocity to 76 cm/s significantly influenced ($p < 0.0001$) the mean power of the Doppler signal, as shown in Fig. 4(b). A significant decrease of 2.2 dB of the mean power was obtained during acceleration of flow, reaching a minimum at peak systole, and an increase of 1.9 dB was observed during deceleration of flow.

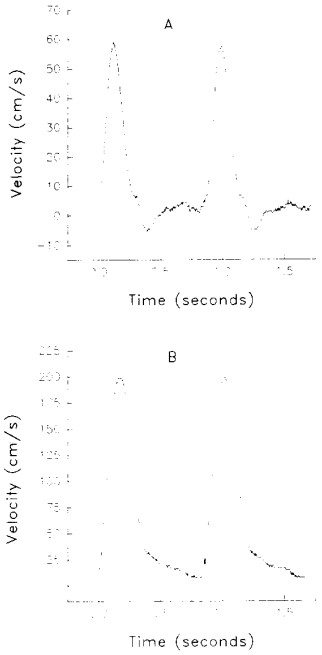

Fig. 3. Typical instantaneous mean velocity versus time traces measured by an electromagnetic flowmeter and used in the present study. The example shows the curves obtained when red cell suspensions at 40% hematocrit were circulated in the flow loop model. Panel (a) corresponds to a mean velocity of 11 cm/s (mean Reynolds number of 191), whereas panel (b) corresponds to a mean velocity of 64 cm/s (mean Reynolds number of 1109).

Measurements were also performed at a high mean flow velocity (64 cm/s) for red cells suspended in saline solution at 4 and 40% hematocrits, the results of which are presented in Fig. 5. Each data point corresponds to the average of five experiments performed with different blood samples. Significant cyclic variations were observed for both concentrations of red cells ($p = 0.001$ at 4% hematocrit and $p < 0.0001$ at 40% hematocrit).

At 4% hematocrit [Fig. 5(a)], the mean power decreased by 0.8 dB in systole and increased by approximately 0.9 dB in early diastole. At 40% hematocrit, a more irregular pattern of variation was obtained. In early systole, the mean power increased by 0.5 dB and then dropped by 1.2 dB. A minimum was measured at peak systole, and an increase of 2.4 dB was found in early diastole. Two notches in the mean power curve can be observed at 40 and 200 ms. The higher mean power values for these two data points were consistent for all five experiments. From panel (b) of Fig. 5, it can be observed that the backscattering power at the end of the deceleration phase (at 440 ms) is higher than that obtained at the beginning of acceleration (0 ms). The 1.5 dB difference suggests that the mean power should decrease during diastole. In the present study, no measurement was performed during mid- and late diastole. Fig. 6 shows an example of mean Doppler spectra obtained during acceleration, peak systole, and deceleration of flow for a mean velocity of 64 cm/s and a hematocrit of 40%.

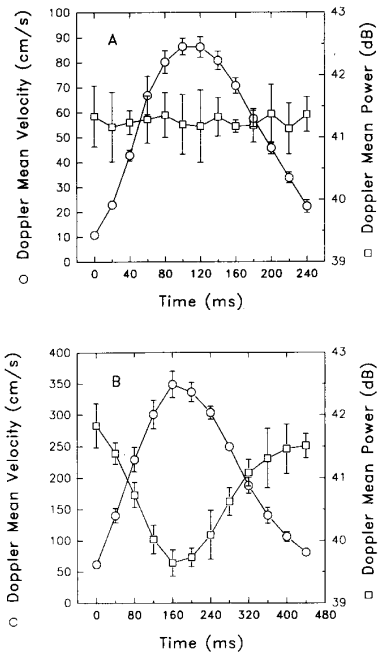

Fig. 4. Mean velocity within the Doppler sample volume of polystyrene microspheres and mean power as a function of the timing within systolic and early diastolic periods. Panel (a) corresponds to a mean velocity of 11 cm/s as measured with the electromagnetic flowmeter, and panel (b) corresponds to a mean velocity of 76 cm/s. Each data point represents the mean ± standard deviation computed over five experiments. The mean spectra were averaged over 200 cycles.

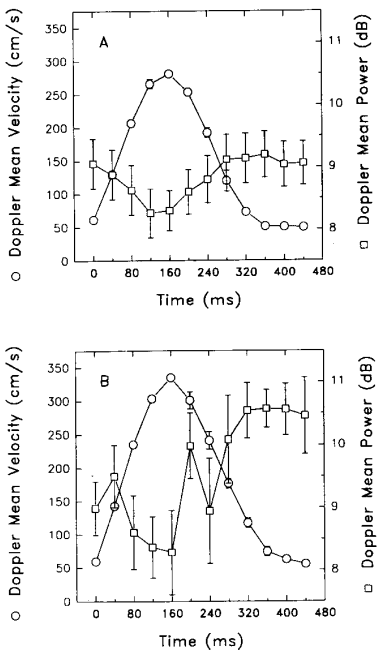

Fig. 5. Mean velocity within the Doppler sample volume of porcine red cell suspensions and mean power as a function of the timing within systolic and early diastolic periods. Panel (a) corresponds to a hematocrit of 4%, whereas panel (b) corresponds to a hematocrit of 40%. Both experiments were performed at a mean velocity of 64 cm/s as measured with the electromagnetic flowmeter. Each data point represents the mean ± standard deviation computed over five experiments. The mean spectra were averaged over 200 cycles.

Additional measurements were performed to study more closely the effect of hematocrit on the cyclic variation of the Doppler power, the results of which are presented in Fig. 7. Three two-way analyses of variance with repeated measurements on one factor, the timing within the flow cycle, were performed to evaluate the statistical significance of these results. Statistical differences ($p < 0.0001$) were measured for all three curves (timing = 0, 160, and 440 ms). However, multiple interaction analyses showed no difference for some values of hematocrit.

No difference ($p > 0.05$) was found between the mean power at peak systole (at 160 ms) and that at the end of early diastole (440 ms) for a mean hematocrit of 2.3%. Insignificant differences ($p > 0.05$) were also measured between the curve at peak systole (160 ms) and that at early systole (0 ms) for mean hematocrits of 2.3, 9.3, and 42.0%. Comparison between the curve in diastole (440 ms) to that in early systole (0 ms) showed that for hematocrits lower than 14.3%, the mean power values were not significantly different ($p > 0.05$). The results presented in Fig. 7 are in accordance with those shown previously in Fig. 5 for 4 and 40% hematocrits.

A. *Effect of Blood Flow Turbulence*

The effect of blood flow turbulence on the cyclic variation of the Doppler power was evaluated at a mean flow velocity of 11 cm/s. Red cells suspended in saline at 40% hematocrit were circulated in the model. Experiments were also performed in laminar flow by removing the screen mesh to compare results with those obtained in turbulent flow. Fig. 8 shows examples of mean Doppler spectra obtained at peak systole for both laminar and turbulent flow. Introducing the mesh in the flow model induced an increase of 433% of the bandwidth at −10 dB and

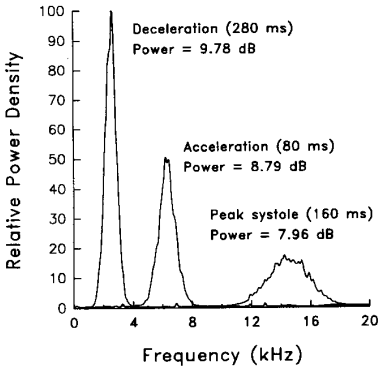

Fig. 6. Examples of mean Doppler power spectra of porcine red cell suspensions at a hematocrit of 40% and a mean velocity of 64 cm/s during acceleration of flow (timing = 80 ms), peak systole (timing = 160 ms), and deceleration of flow (timing = 280 ms). The mean power of each spectrum is also shown. Normalization of the maximal power was performed after computing the mean power in decibels.

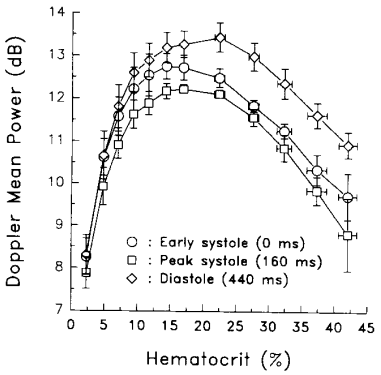

Fig. 7. Mean power of averaged Doppler spectra for porcine red cell suspensions as a function of hematocrit and flow cycle position (time = 0 ms corresponds to the beginning of systole, time = 160 ms to peak systole, and time = 440 ms to the end of early diastole). Experiments were performed at a mean flow velocity of 64 cm/s as measured with the electromagnetic flowmeter. Each data point represents the mean ± standard deviation computed over five experiments. The mean spectra were averaged over 100 cycles.

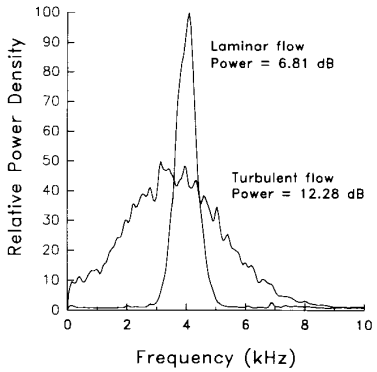

Fig. 8. Examples of mean Doppler power spectra of porcine red cell suspensions at peak systole for 40% hematocrit, a mean velocity of 11 cm/s, for both laminar and turbulent flow. The mean power of each spectrum is also shown. Normalization of the maximal power was performed after computing the mean power in decibels.

an increase of the mean power of 5.5 dB. Fig. 9 summarizes the results on the effect of laminar and turbulent flow on the Doppler power.

No cyclic variation of the Doppler power (one-way analysis of variance with repeated measurements, $p = 0.999$) was obtained in laminar flow. At a mean flow velocity of 11 cm/s, a similar result was observed previously with polystyrene microspheres. In turbulent flow, the Doppler power significantly changed ($p = 0.001$) within the cardiac cycle. As predicted by (1) and (3), increasing the turbulence downstream of the mesh screen by increasing the mean velocity within the tube induced an increase of the Doppler power. The mean power first raised by 1.7 dB to reach a maximum early after peak systole. During early diastole, the mean power decreased by 0.8 dB. As discussed previously, the higher power at the end of the flow deceleration phase compared to that measured at the beginning of systole also suggests that the mean power should decrease during mid and late diastole.

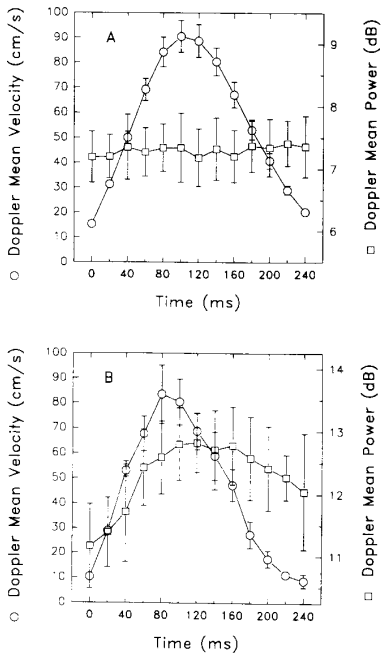

Fig. 9. Mean velocity within the Doppler sample volume of porcine red cell suspensions at a hematocrit of 40% and mean power as a function of the timing within systolic and early diastolic periods. Panel (a) corresponds to the laminar flow experiments, while panel (b) represents measurements performed in turbulent flow. Both experiments were carried out at a mean velocity of 11 cm/s as measured with the electromagnetic flowmeter. Each data point represents the mean ± standard deviation computed over five experiments. The mean spectra were averaged over 200 cycles.

B. Summary

The following points summarize the finding of the present study.

1) No cyclic variation has been observed at a mean flow velocity of 11 cm/s for both polystyrene microspheres [Fig. 4(a)] and porcine red cell suspensions at 40% hematocrit [Fig. 9(a)].

2) A significant and similar pattern of variation has been obtained at high mean flow velocities (76 and 64 cm/s) for dilute polystyrene microspheres [1 g/L, Fig. 4(b)] and dilute red cell suspensions [4% hematocrit, Fig. 5(a)].

3) A significant cyclic variation has been measured for red cell suspensions at 40% hematocrit for high flow velocity [64 cm/s, Fig. 5(b)]. However, the pattern of variation was slightly different from that observed at lower concentrations of red cells [Fig. 5(a)].

4) Higher mean power values were measured at the end of early diastole than values calculated at the beginning of systole for hematocrits higher than approximately 14% (Fig. 7).

5) The presence of turbulence has been found to also affect the power of the Doppler signal within the flow cycle [Fig. 9(b)].

V. Discussion

Our results clearly demonstrated that the Doppler backscattering power from polystyrene microspheres and porcine red

cell suspensions varies cyclically in some experimental situations. It was shown that high-velocity changes and turbulence induce a cyclic variation. In the present study, polystyrene microspheres were used as scatterers mainly to validate results obtained with red cell suspensions. There are certain advantages with the use of microspheres. First, these particles do not form agglomerates because of the repulsive surface tension present on them. This rheological particularity of microspheres is interesting because it corresponds to that of nonaggregating red cell suspensions. The spherical shape of these particles and their nondeformability represent another point of interest because it is known that changes in shape can influence particle–particle interactions, rotation, and possible migration in flowing suspensions [25], [26]. These factors will be considered later when discussing the mechanisms involved in the observed cyclic variation.

From the results obtained with red cell suspensions at 40% hematocrit and presented in Figs. 9(a) and 5(b), it is interesting to note that increasing the mean velocity averaged over the tube and over the cardiac cycle from 11 to 64 cm/s increased the average Doppler power. The average power for a mean velocity of 11 cm/s was 7.3 ± 0.4 dB and that for a mean velocity of 64 cm/s was 9.6 ± 1.0 dB. In *in vitro* steady laminar flow experiments performed with a 5 MHz CW Doppler flowmeter, Bascom *et al.* [5] found no change in the power of the Doppler signal scattered by nonaggregating red cells when the mean flow within the tube varied between 68 and 100 cm/s. However, in another recent steady laminar flow experiment study [27], the PW color Doppler power scattered by sand particles of different concentrations increased as the velocity within the tube increased until a specific velocity of approximately 35 cm/s was reached, but did not change further despite an increase in velocity up to 52 cm/s. This latest result of Parro *et al.* [27] obtained using carrier frequencies at 2.25 and 2.5 MHz, although not performed in pulsatile flow, is in agreement with our results. In the present study, varying the pulsatile mean velocity from 11 to 64 cm/s increased the Doppler power from red cell suspensions by 2.3 dB. However, we also noted that increasing the mean velocity did not significantly change the average Doppler power for polystyrene microspheres. From Fig. 4, the average power for a mean velocity of 11 cm/s is 41.5 ± 0.4 dB and that for a mean velocity of 76 cm/s is 40.8 ± 0.8 dB. The reason for this discrepancy between the results obtained with microspheres and red cell suspensions is not known.

From Fig. 5, it may be surprising to observe that the mean Doppler velocity within the sample volume at peak systole differs for hematocrits of 4 and 40%. Both experiments were performed at a mean velocity averaged over the tube of 64 cm/s. The reason for this difference is as follows. The effect of the hematocrit on the flow profile within a tube has been studied by Goldsmith and Karino [25]. As the concentration of particles increases, the velocity distribution becomes more blunt at the center of the tube. In the present study, only the mean velocity within the tube averaged over the flow cycle was monitored. As the velocity profile probably changed to a more blunt shape, the output of the pulsatile pump was increased to maintain a constant mean flow. Consequently, the velocity at the center of the tube slightly increased as the hematocrit increased.

A. Comparison of the Results with Studies Reported in the Literature

Our results showed that large velocity changes and turbulence can induce a cyclic variation of the Doppler backscattered power. However, for an instantaneous mean velocity versus time curve similar to that found in the normal human femoral artery [see Fig. 3(a)], no cyclic variation of the Doppler power was found for both polystyrene microspheres and porcine red cell syspensions at 40% hematocrit. This last result is in accordance with previous observations by Luckman *et al.* [1] and Bascom *et al.* [5] where no cyclic variation was measured over the carotid artery of a healthy male adult (Luckman *et al.*), and from an *in vitro* model of the internal carotid artery (Bascom *et al* .). De Kroon *et al.* [4] measured a cyclic variation at 30 MHz in human iliac arteries. However, they attributed the variation to the presence of red cell aggregation. The use of a carrier frequency at 30 MHz may have amplified the influence of aggregates on the variation of the Doppler power because at that frequency, Rayleigh scattering may no longer exist. In a preliminary study performed by our group [6] at a carrier frequency of 10 MHz, the presence of aggregates did not induce a cyclic variation at a pulsation rate of 70 beats/min. The lower frequency of the transmitted ultrasonic signal may be the reason for the discrepancy between our results [6] and those of De Kroon *et al.* [4]. In the study by Luckman *et al.* [1] and Bascom *et al* . [5], ultrasonic frequencies at 5 and 8 MHz were used, respectively. In the present study, the frequency of the transmitted signals was 10 MHz.

B. Possible Mechanisms Involved in the Observed Cyclic Variations

Several factors that could theoretically affect the Doppler power were given previously in the Background section. Based on (1), (2), and (3), factors that could explain the observed variations in Doppler power within the cardiac cycle are changes in local hematocrit within the sample volume, variation in scatterer shape and correlation among particles (parameter c), and modification of the variance in particle size (parameter d). It is very unlikely that the particle size distribution changes within the cardiac cycle. On the other hand, it is known that turbulence decreases the correlation among particles. A change in correlation within the flow cycle (variation of parameter c) may explain the cyclic variation observed in turbulent flow. In Fig. 9(b), it is seen that the maximum of the mean power occurs early in diastole. Several authors have reported a higher instability in pulsatile flow during early diastole [28]–[30]. Shen [28] found that decelerating flows are generally less stable than accelerating flows. In the presence of turbulence in both straight tubes [29] and constricted tubes [30], the deceleration of the flow also tends to increase turbulence intensity. Our results then seem to be in accordance with these observations. A higher turbulence intensity in early diastole might have contributed

to the increase in the mean power by reducing the correlation among particles.

It is more difficult to elucidate the mechanisms involve in the observed cyclic variation at high flow velocity. Changes in the shape of the particles when subject to varying shear rates, although observed by Goldsmith [26] for red cells, cannot explain the modifications in the backscattering power within the cardiac cycle. For instance, cyclic variation was observed for both deformable red cells and nondeformable polystyrene microspheres. From (1) and (3), the only remaining possible mechanisms are variations in local hematocrit and modifications in the correlation among particles.

C. Possible Mechanisms for the Cyclic Variation at High Flow Velocity

The possibility of a change in the concentration of particles at the center of the tube within the flow cycle is a very intriguing and interesting phenomenon. Changes in local hematocrit were observed experimentally by Goldsmith [25], [26] in 50–200 μm diameter tubes. Deformable particles in Poiseuille flow (steady laminar flow) at low mean shear rates ($< 25\,s^{-1}$) were shown to migrate away from the wall toward the axis. A more uniform distribution was found for rigid particles. At higher mean shear rates ($> 96\,s^{-1}$), the "tubular pinch effect" was seen to occur [26]. Particles near the wall migrated toward the axis, while those at the center of the tube moved toward the wall. A zone of highly concentrated red cells was observed by Goldsmith [26] at a distance of approximately 0.4–0.6 R, where R is the radius of the tube. The "tubular pinch effect" was observed for both deformable red cells and hardened red cells.

In steady laminar flow experiments performed in a tube of larger diameter (3 mm), a different behavior was observed at hematocrits of 20, 40, and 60% [31]. Experiments were performed at mean velocities of 9.5, 30, and 50 cm/s (mean shear rates of 240, 760, and $1260\,s^{-1}$). For all values of hematocrit and mean velocity, the concentration distribution of ghost red cells was high in the core of the tube and about zero near the wall. Based on these results and as suggested by Goldsmith and Karino [25], it seems that several factors, namely, flexibility and size of the particles, mean shear rate, tube diameter, viscosity of the fluid, and hematocrit, may influence the behavior of cell migration. Another factor not addressed by Goldsmith and Karino [25], and probably significant, is the pulsatility of the flow. Unfortunately, no experimental evidence exists to support the possibility of saline suspended red cell migration in pulsatile flow and in tube diameter of approximately 0.476 cm, the primary reason being a lack of experimental methods for studying the behavior of red cells flowing in a large diameter tube. The availability of high-frequency ultrasonic intravascular imaging devices may be an answer to this problem. However, one may have to consider the interaction of the intravascular probe with the blood flow in such a study.

As seen from the previous discussion, there is considerable uncertainty as to whether cell migration and polystyrene microsphere migration occurred in our experiments. Before going further, it is very important to realize that such migration can have a totally different consequence on the backscattering Doppler power, depending on the value of the mean hematocrit within the tube. As seen in Fig. 7 and as reported in previous studies [11], [14], [32], the power scattered by red cells increases monotonically with hematocrit at low hematocrits up to a value of approximately 13%. For hematocrits higher than 13%, the power drops. To explain the decrease in the Doppler power during the acceleration of flow for red cells at 4 and 40% hematocrits (see Fig. 5), the concentration of red cells at the center of the tube would have to decrease at 4% hematocrit and increase at 40% hematocrit. Moreover, at approximately 13% hematocrit, the cyclic variation would theoretically be minimal because the slope of the mean Doppler power versus hematocrit curves is minimal around that value of hematocrit for laminar flow (see Fig. 7). Based on Fig. 7, a cyclic variation was observed in our experiments around 13% hematocrit, which suggests that red cell migration is not the main determinant in the observed cyclic variation at high flow velocity.

D. Cyclic Changes in the Correlation Among Particles at High Flow Velocity

It is postulated in the present study that the correlation among particles changes as the velocity changes in high flow velocity experiments. When the flow accelerates, the correlation among particles probably increases because the spatial coherence between scatterers becomes more deterministic. As the velocity decreases, the cells and polystyrene microspheres obtain more freedom in their movement and the correlation among particles decreases. The differences in the shape of both curves presented in Fig. 5 may be explained by biorheological and flow dynamics phenomena. The higher backscattering power values at 40 and 200 ms after the beginning of systole for 40% hematocrit [see Fig. 5(b)] are explained as follows. In early systole, when the flow velocity increases, the cells start to rotate and their long axis aligns with the flow field. As discussed by Goldsmith and Karino [25], the cycle of rotation is significantly reduced at high hematocrit, and the cells travel parallel to the axial velocity field most of the time. It may be speculated that this alignment is the cause of the higher power in early systole (at 40 ms). In a study of McMillan *et al.* [33], it was shown that the orientation of the cells influenced the reflectivity of light. It is interesting to observe that such orientation of the cells influenced the reflectivity of light. It is interesting to observe that such orientation of the cells does not seem to be present at 4% hematocrit [see Fig. 5(a)]. At low hematocrit, Goldsmith and Karino [25] observed a constant rotation of the cells and no particular orientation.

The higher power 200 ms after the beginning of the flow cycle is believed to be associated with the presence of instability during flow deceleration [28]. As discussed previously for turbulent flow at 40% hematocrit, the presence of flow instability may have decreased the correlation among particles. It is interesting to observe that no such instability in early diastole was probably present at low concentrations of particles

[microspheres at a concentration of 1 g/L, as seen in Fig. 4(b), and red cell suspensions at 4% hematocrit, as presented in Fig. 5(a)].

The higher power at the end of early diastole (at 440 ms) compared to that measured at the beginning of systole (0 ms) for hematocrits higher than approximately 14% [see Figs. 7 and 5(b)] may also be explained by a lower correlation among red cells in early diastole. Because this phenomenon does not occur at low hematocrits or at a low concentration of polystyrene microspheres, we believe that the interaction among neighboring particles, such as seen at 40% hematocrit, plays a role in changing the correlation among particles.

VI. Conclusion

Changes in the correlation among particles were suggested as a possible mechanism to explain the cyclic variations of the Doppler power in the presence of large velocity changes and turbulence. In normal human arteries, the maximal velocity at peak systole is usually lower than 150 cm/s. Such high velocity can be found in the human ascending aorta [24]. In the present study, maximal velocities at peak systole were approximately 200 cm/s for high flow velocity experiments, as shown in Fig. 3(b). Based on our results, it may be expected that a cyclic variation of the Doppler power can also be found in the normal ascending aorta in the absence of turbulence because a large range of variation of flow velocities is observed in this artery.

The observation of an increase in Doppler power and the detection of a cyclic variation in the presence of blood flow turbulence may have clinical implications. Detecting turbulence by Doppler ultrasound would allow improved characterization of arterial stenosis. The combination of this information with the quantification of Doppler spectral broadening may contribute to the improved diagnosis of patients with minimal-to-moderate arterial obstructions.

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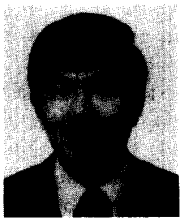

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