Hemolysis in Simple Shear Flows

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Whole human blood was sheared in the flow field of a concentric cylinder Couette viscometer in the hope of isolating the physical traumas specifically related to the escape of intracellular hemoglobin. The experimental results indicated that laminar shear stresses per se were not instrumental within the investigated range (up to 600 dynes/sq.cm.). This, plus the hematocrit dependence, evidenced the importance of interaction of individual red cells with a surface in the hemolytic event.

One of the problems to be considered in the design of extracorporeal circuits (such as blood pumps, continuous blood centrifuges, cardiopulmonary bypass machines, and hemodialyzers) and phosthetic devices such as heart valves is the escape of intracellular hemoglobin from the red blood cells into the surrounding plasma. This phenomenon, known as *hemolysis*, must be kept below a certain tolerance level. Plasma hemoglobin concentrations above 160 mg./100 ml. can be toxic to the human, as the body degrades these molecules and the kidneys cannot dispose of the waste in such quantities (*9*). Also, the decrease in intracellular hemoglobin concentration can cause patients to become anemic.

The aim of this investigation was to determine the specific type of physical interaction mainly responsible for hemolysis in simple shear flows. To accomplish this, whole human blood was sheared in a fixed geometry concentric cylinder Couette viscometer. The variables investigated were the shear stress τ, the revolutions per minute (and thus the cell velocity v), the hematocrit $\mathcal{H}$ (volume fraction of red blood cells), and the time of traumatization.

BACKGROUND

The normal human red blood cell is a nonnucleated cell whose shape is best described as a biconcave disk with a diameter of 8.5μ (see Figure 1). It consists of a highly oriented lipoprotein framework (membrane) 75Å. thick, surrounding a fluid heavily concentrated (~33 wt. %) with hemoglobin molecules. The fluid may have some orientation, as opposed to the isotropy and randomness characterizing a simple solution of hemoglobin.

Through details of the fine structure of the membrane are unknown as yet, its composition can be inferred from the complex mixtures of fibrous protein, lipid, and small quantities of mucopolysaccharides which which can be prepared from cell ghosts, the remnants of complete hemolysis. Whether hemoglobin is bound in part to membrane substances is uncertain. There is no evidence for the existence of membrane pores except for those so small as to allow the passage of lipid-insoluble materials such as water and small ions. Rheological studies have determined that the membrane is viscoelastic in its mechanical properties (*10, 16*).

From observations of red cells in Poiseuille flow, it is learned that the red cells are extremely flexible structures, being continuously deformed into a variety of shapes and occasionally becoming folded into U shaped conformations (*8*). It has sometimes been suspected that local fluid shear stress determines the extent of hemolysis by causing the red cell membrane to stretch beyond a yield point (*2*).

PREVIOUS WORK

A variety of experiments have been performed to isolate the parameters specifically related to the escape of hemoglobin. Fok and Schubothe subjected blood to a standardized amount of trauma in an Erlenmeyer flask containing a fixed number of 4 mm. glass beads (*6, 18*). The entire flask was rotated about a horizontal axis through its center line. Although the exact cause of the hemolysis is impossible to ascertain in such a complex flow, some valuable results were obtained. The hemolysis was found to be directly proportional to both the number of beads in the flask (suggesting the possible importance of solids surface area) and also to the time of traumatization.

Numerous results of hemolysis in tube flow have been reported since 1959. It is difficult to interpret these data, too, owing to the variable shear field (characteristic of all capillary flows) and the complex interactions of the deformable red cells with such a field. In addition, near the center of the tube where stresses can be low, the cells can stack and move as a plug.

In two separate studies of hemolysis in laminar flow, a circular loop of tubing (half filled with blood) was closed on itself and the loop mounted on a vertical disk which was rotated (*11, 21*). Stewart and Sturridge found that tube surface roughness (maximum roughness peaks of 15μ) was associated with increased hemolysis, suggesting the possible effect of increased tube surface area per unit blood volume. When the inside diameter of the tube was doubled, the flow velocity could be approximately doubled before the same amount of hemolysis was incurred. This would support a postulated shear stress mechanism, since $\tau = \mu\ (dv)/(dr)$, and both μ and dv/dr would be approximately the same under both conditions. It also admits the possibility of a mechanism involving the surface. That is, doubling the diameter alone would have led to less hemolysis, corresponding to the tube surface area per unit blood volume, $\dfrac{\pi Dl}{\pi \dfrac{D^2 l}{4}} \approx \dfrac{1}{D}$ also being less. This interpretation is consistent with the work of Kusserow and Kendall, who found that decreasing the internal tube diameter for the same rotational speed increased the hemolysis rate. In a separate set of experiments, they held the tube velocity $V = \Omega\ R_c$ constant while varying the angular velocity Ω and the radius of curvature of the loop R_c. When Ω was doubled (and hence R_c halved), the hemolysis was tripled. If, indeed, the blood velocity and velocity distributions were identical in all cases, this result would rule out a stress mechanism as well as a mechanism involving collisions with the tube surfaces. However, owing to laminar secondary flows and different ratios of free (blood-air) surface to blood vol-

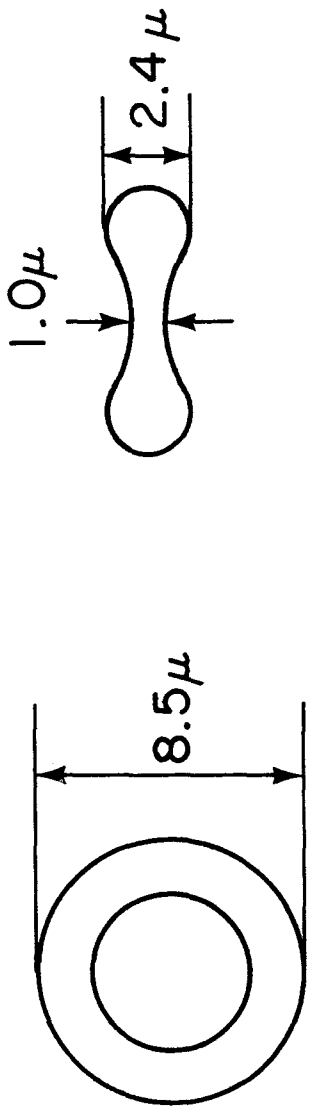

Fig. 1. Human red blood cells: (A) top view, (B) cross section from a plane through the cell center. Dimensions from Ponder (15).

ume, the flow characteristics were probably significantly different in the various individual experiments, and hence few definite conclusions can be drawn.

In a couple of papers on erythrocyte destruction (3, 4), Blackshear et al. have attempted to isolate the hemolytic actions of various physical traumas. Their work with a Couette system, capillary flow systems, and circular turbulent free jets is not described clearly in these publications. The reported data seem limited, and the applicability of various theories utilized to interpret the results is not at all certain. Nevertheless, Blackshear et al. were apparently the first both to postulate and attempt to prove that hemolysis in shear flows may be associated with red cell-surface interactions. A subsequent publication by Bernstein, Blackshear, and Keller surveys this work as well as other pertinent studies (2).

Nevaril and Hellums attempted to isolate the effect of shear stress on hemolysis by imposing extremely high stresses (up to 4,500 dynes/sq.cm.) in a concentric cylinder viscometer machined to a cone-on-plate at the bottom so that the same uniform shear rate would be applied throughout the fixed-geometry flow field (12, 13). These severe conditions were sufficient to cause the red cells to fragment, and the hemolysis appeared to be correlatable

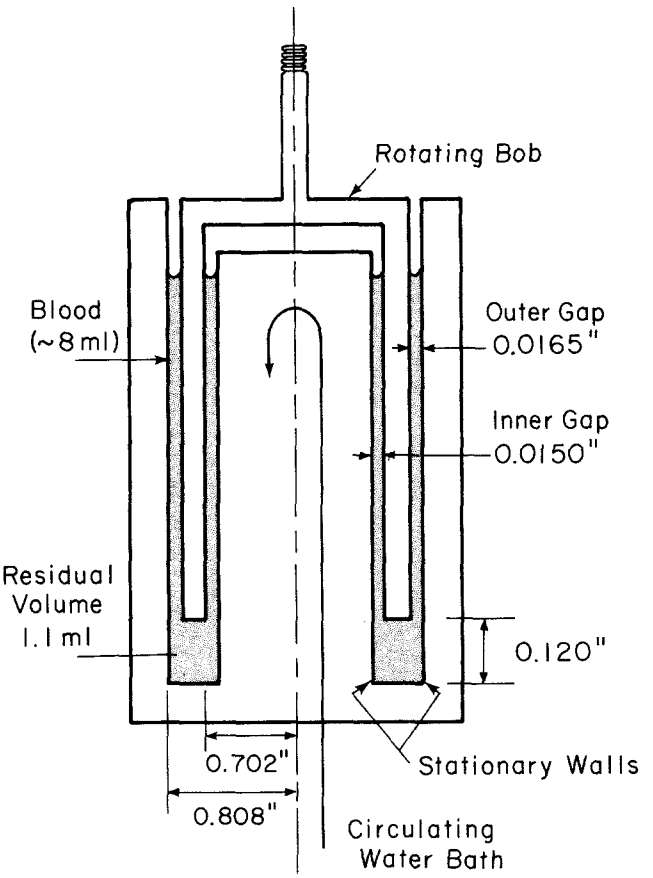

Fig. 2. Double-gap viscometer.

with τ alone. The present investigation involved the lower range of shear stresses commonly achieved in the circulation and in extracorporeal circuits. There was no fragmentation observed in this study.

APPARATUS

A Haake Rotovisco cylindrical rotating viscometer with the NV measuring system (made of nickel plated brass) was the basic unit which was modified to meet the requirements of this study. The blood to be sheared was contained within the gaps between the surfaces of the rotating bob and the inner and outer stationary walls (see Figure 2). A residual volume of 1.1 ml. of blood was not contained within the uniform shear gap spaces. The fraction of the hemolysis occurring in this volume was separately determined and accounted for in the calculation of hemolysis (see results).

The rotating bob is connected to a measuring head which consists of two coaxial shafts, a creep resistant spring, and a potentiometer. Torque on the rotating member causes a proportionate displacement of the spring. The angle of displacement is converted by a potentiometer into an electrical resistance which controls the electrical signal and hence the scale reading on a control cabinet.

In order to extend the revolutions per minute range to 1,500 (corresponding to shear rates of 7,500 sec.$^{-1}$), the original motor in the control cabinet was replaced by a more powerful, nonsynchronous one. A variable transformer was used to control the current to the motor in order that the final rotational speed could be approached gradually.

Blood temperature during shearing was fixed by a circulating water bath which was controlled within ± 0.5°C. In most experiments, the bath was near 30.5°C. Owing to the low viscosity of blood and the short duration of an individual run, viscous heating was negligible. From an approximate formula, the maximum (steady state) temperature difference across the system was calculated to be less than 1/10°C. for the highest revolution per minute and blood viscosity used in this study.

PROCEDURE

The same source of human blood was used in most experiments, a hemachromatotic patient whose blood was being drawn to the extent of three pints every 2 weeks. (One effect of this frequent blood drawing is to skew the age distribution curve of the red blood cells towards the lower age level.) The blood was drawn into heparin solution and was stored at 5°C. with no sugar source for 5 to 6 days under sterile conditions.

The blood was allowed to reach room temperature prior to an experiment. The samples, withdrawn slowly from a bottle through a glass pipet, were 8-ml. volumes in most experiments. Pipets and viscometer surfaces were washed out and rinsed with isotonic saline before each individual run.

A sample which served as the reference was pipetted directly into a test tube, falling slowly down the tube wall. Another sample was then pipetted similarly into the viscometer. Bob rotational speed was gradually raised to the value under investigation in a fixed time, usually 15 sec. Speed was held constant at this maximum level for a fixed amount of time, and then the applied stress was quickly removed. Stress was read in arbitrary units from the instrument scale, and the maximum rotational speed was determined by using a Strobetac.

The viscometer cup was slowly lowered and removed from the assembly and the blood poured into a test tube. After the sample was gently mixed by overturning the test tube, part of the blood was used to determine the hematocrit by using a micro method. The remaining blood was centrifuged for 10 min in a standard International Clinical Centrifuge at 1,000 g. and the supernatant plasma withdrawn. This solution was analyzed for hemoglobin by combining the hemoglobin with benzidene and by performing a spectrophotometric analysis of the resulting complex (see reference *14* for details). The percentage hemolysis H was then calculated (see Appendix) and plotted vs. the pertinent variable in the experiment.

In the variable-hematocrit experiments, the different samples were withdrawn from different depths in the blood bottle after some settling had occurred. Hematocrits up to 0.75 could be obtained in this way.

Experiments were also performed to determine the effect of the time of traumatization. While the hematocrit and acceleration characteristics were held constant for all runs, the time for which the blood was subjected to the shear value under investigation was varied from 0 to 20 min.

EXPERIMENTAL RESULTS

The measure of hemolysis being presented, denoted by H, is the percentage of red blood cell hemoglobin released by the red cells due to controlled shearing (see Appendix for the method used to calculate H).

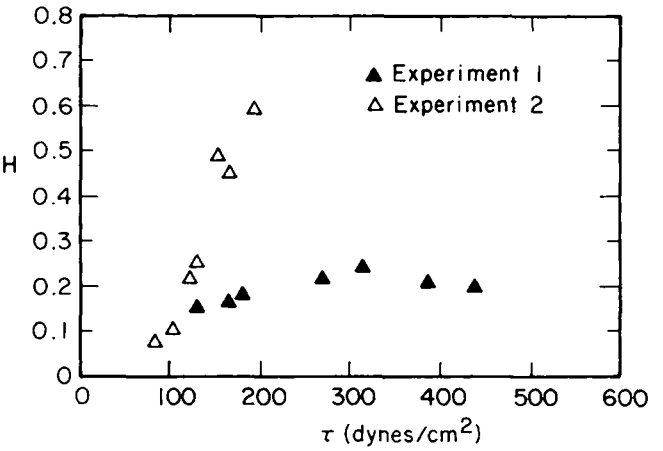

Fig. 3. Experiment No. 1: rev./min. 650 for 3 min., blood stored for ~ 5 days at ~ 5°C. Experiment No. 2: $\bar{H}$ = 0.40, 2-min. runs, blood stored for ~ 6 days ~ 5°C. Data from reference 19.

It was necessary to establish that most of the hemolysis was occurring in the uniform gap spaces and not near the base of the rotating bob or in the 1.1 ml. residual volume. To accomplish this, the sample volume was varied from 2 to 7 ml. for the same nominal shearing conditions. A plot of total hemoglobin released (milligrams) vs. sample volume was extrapolated to show that the amount of hemolysis attributable to the lower regions of nonuniform shear was about the same as, or somewhat less than, in the Couette uniform-shear spaces (on a per unit volume basis). Thus the lower regions contributed less than 15% of the total hemoglobin release in subsequent runs, when the viscometer contained 8 ml.

In the initial experiments, the hematocrit was essentially constant for all blood samples in a given set of tests, and the apparent dependence of H on τ (or rotational speed) was thereby determined. Typical results are given by the data of experiment 2 in Figure 3. In such experiments it is impossible to separate the effect of τ from that of rotational speed, since the two are proportional under these constant hematocrit and thus constant viscosity conditions.

In another series of tests, in which the rotational speed was constant for all samples, τ was varied by selecting samples of different hematocrit. Remarkably different results were obtained, as shown by the results of experiment 1 in Figure 3. (It should be pointed out that blood samples represented in Figure 3 were quite similar; they were drawn from the same donor and treated essentially the same prior to experimentation.) Comparison of the

TABLE 1. RESULTS OF CONSTANT REVOLUTIONS PER MINUTE EXPERIMENTS

Exp't. No.		Sample No. 1	2	3	4	5	6	7	8	9	10	11	12	13	14
1	$\bar{H}$	0.72	0.62	0.45	0.67	0.55	0.40								
	τ (dynes/sq.cm.)	440	316	182	387	269	165								
	H	0.19	0.23	0.15	0.20	0.20	0.13								
3	$\bar{H}$	0.74	0.75	0.61	0.30	0.72	0.54	0.37	0.70	0.56	0.33	0.32	0.68	0.67	0.62
	τ (dynes/sq.cm.)	525	554	335	132	488	246	145	434	298	127	127	420	382	307
	H	0.45	0.47	0.51	0.26	0.39	0.36	0.34	0.36	0.51	0.25	0.21	0.41	0.37	0.34

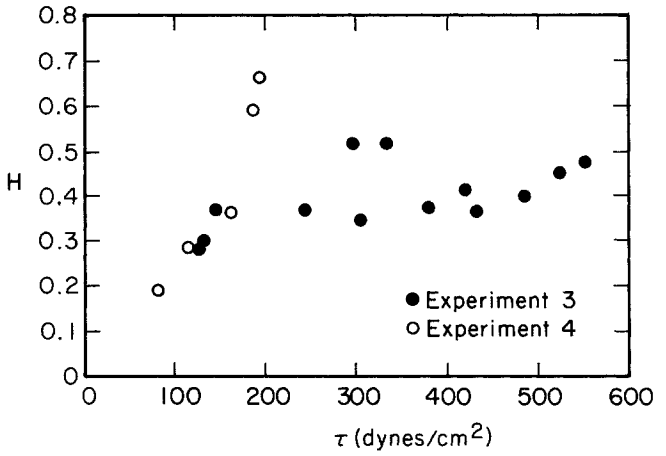

Fig. 4. Experiment No. 3: rev./min. = 590 for 3 min., blood stored for ~ 6 days at ~ 5°C. Experiment No. 4: $\mathcal{H}$ = 0.31, 2-min. runs, blood stored for ~ 5½ days at ~ 5°C. Data from reference 19.

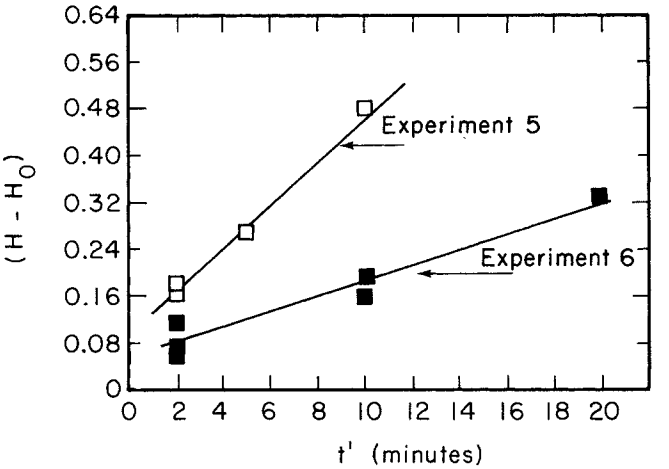

Fig. 5. The time dependence of hemolysis. $\mathcal{H}$ = 0.35 in experiment 5. $\mathcal{H}$ = 0.34 in experiment 6.

two sets of data in Figure 3 demonstrates conclusively that laminar stress in the bulk fluid is not the parameter that correlates hemolysis data in this stress range. Similar sets of data are presented in Figure 4, and the hematocrits of the individual samples in experiments 1 and 3 are presented in Table 1.

Since the hemolysis data are not correlated by a stress parameter, mechanisms which can be related conceptually to fluid velocity appear to be favored. For example, the hemolytic event could involve cell-cell collision phenomena and/or interaction of the cells with the viscometer surfaces. Of these two possibilities, the latter is strongly favored on the basis of a further inference drawn from the constant revolutions per minute experiments. In these, H was relatively independent of τ and thus $\mathcal{H}$, and, therefore, according to the definition of H, the total amount of hemoglobin released was proportional to $\mathcal{H}$ to a power near 1. This means that a single cell is involved in the hemolytic event, and that cell-surface (as opposed to cell-cell) interaction must be predominantly responsible for the hemolysis. If cell-cell interaction were of major hemolytic significance, a very strong dependence of total hemoglobin release on $\mathcal{H}$ would have been expected, owing to the concentrated nature of the suspensions (hematocrits varied from 0.30 to 0.75). Such high order interactions do occur, for example, in the strong dependence of blood viscosity on hematocrit in this same hematocrit range (5), where multibody effects are acknowledged to be responsible.

In another set of experiments, the time dependence of hemolysis was studied under steady shearing conditions. Data were taken with ACD-preserved blood obtained from the Red Cross after its 21 day expiration date and results are plotted in Figure 5 as $(H - H_0)$ vs. time. The term H_0 is subtracted in an effort to remove from the H data the hemolysis occurring during acceleration and deceleration of the bob, so that $(H - H_0)$ would reflect only the effects of steady shear. H_0 was evaluated, for each blood, by shearing a separate sample in a run which consisted of the standard (15 sec.) acceleration to the shear level being investigated, followed immediately by deceleration. Figure 5 shows clearly that hemolysis under steady shear is an increasing function of time, essentially linear in this low range of H. Such behavior is consistent with many possible mechanisms, although not with the concept of a particular kind of stress induced destruction in which a certain small fraction of susceptible cells would hemolyze almost immediately upon exposure to sufficient (characteristic) stress. The failure of the data in Figure 5 to extrapolate to zero is probably due to the approximate nature of the H_0 correction, although if real it would indicate a much higher rate of hemolysis at short times. The latter interesting point, which has additional biomedical implications, requires much more data before it can be resolved completely.

DISCUSSION

It was intended that the hemolysis takes place in a flow characterized as laminar simple shear, so that a uniform stress distribution would prevail. Deviations from this flow can be due to several reasons: Reynolds turbulence, Taylor vortices (both of these may exist even in the presence of excellent geometrical alignment), and other secondary flows produced by the pumping action of a misaligned geometry. Reynolds numbers calculated for the most severe conditions were about an order of magnitude below the usual turbulence requirement, so ordinary turbulence was absent. The Taylor stability criteria indicate no vortex formation within the inner gap but that some vortices may have been present in the outer gap under the more severe conditions encountered here. In the latter case, the numerical value cited (17) as characterizing the onset of secondary flow was exceeded upon occasion by factors up to 3. Data points obtained under these conditions are included in Figures 3 and 4 and appear entirely unexceptional. Certain geometrical misalignments were also present, amounting to several percent of the gap spacing, but it seems likely that the dominant flow was always a tangential velocity component and that bulk secondary flows had negligible effect on hemolysis.

The experimental data constitute strong evidence for importance of interaction between a single red cell and a surface in the hemolytic event. The nature of the interaction and the actual mechanism by which the hemoglobin leaves the red cell are uncertain. Examination of stained films of the sheared blood showed that no cell fragmentation had occurred, and no cell ghosts were seen by using phase contrast microscopy. (However, owing to the small levels of hemolysis considered in this study, a small fraction of cell ghosts could easily have been overlooked.) If ghosts were truly absent, it would be necessary to envision a new mechanism for hemolysis, which has traditionally been thought to involve gross damage to the cell. Alternative mechanisms, incorporating

a more gradual transport of hemoglobin from the nonfragmenting cell during cell-wall interaction, would then become attractive.

The hemolytic response of an individual cell to mechanical trauma is surely a complex function of bulk and surface properties, rheological, chemical, and perhaps dielectric. Moreover, in studies such as these, an experimental problem exists in the change of these properties over a long period of time. In particular, the cells become more susceptible to shear induced hemolysis with in vitro storage time in the absence of additional glucose.* During this period of being exposed to a nonphysiologic environment and exhaustion of available glucose, the cell experiences property changes, which can account for the altered hemolytic behavior. For example, it becomes slightly spherocytotic (*7*) which affects its rheology, and the cell lipid layer undergoes transformation.

These complexities suggested that a useful parameter for characterizing the hemolytic susceptibility of a given cell relative to that of the total population might be its physiological age, which presumably could correlate many physical and chemical properties of the cell. This postulate was tested with samples of fresh rat blood, in which the age of radioactively labeled red cells were known. Results were negative, and, as reported in a separate communication (*19*), it must be concluded that in vivo aging of blood cells does not alter their hemolytic response to the type of trauma studied here.

The findings of this study are reconciled with those of Nevaril, who found that hemolysis data were correlatable with shear stress, with the understanding that these works emphasized two very different shear regimes indeed. In Nevaril's study the much larger shear stresses caused the cells to fragment (supposedly by stretching them beyond the yield point), and the amount of hemolysis was consequently much greater than in the present study. Little time dependence was observed, probably because fragmentation occurs within very short times after application of the requisite stress. With the smaller cell fragments present, viscosity and stress levels actually decreased under constant shear rate (*13*), and little hemolysis due to further fragmentation would be expected. Cell-surface interaction was probably a factor in Nevaril's study as well, and indeed he reported some small increase of hemolysis with time, but this mechanism apparently contributed only a minor fraction of the total amount of hemoglobin released. The stresses imposed in the present study were well below the value of 1,500 dynes/sq.cm., above which Nevaril found fragmentation to occur.

This study was undertaken in the hope of further understanding the phenomenon of hemolysis. Such an understanding is necessary for the development of blood handling techniques and for the rational design of prosthetic and extracorporeal devices. Although most of these systems are not hydrodynamically similar to circular Couette flow, the ideas presented here can hopefully be extended.

ACKNOWLEDGMENT

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NOTATION

D = inner tube diameter, cm.

H = percentage of hemoglobin at risk released into the plasma by the trauma imposed

Hgb = hemoglobin

$\mathcal{H}$ = hematocrit, the red blood cell volume fraction of whole blood

l = length of tube

MG% = number of milligrams of hemoglobin/100 cc. solution

R_c = radius of curvature of circular loop

r = radial coordinate, cm.

r_i = radial coordinate of stationary surface, cm.

t' = time for which maximum stress was applied, min.

v = fluid velocity

V = tube velocity

Greek Letters

Δr = radial distance from stationary surface, $r - r_i$

γ = shear rate, sec.$^{-1}$

μ = shear viscosity, poise

τ = shear stress, dynes/sq.cm.

Ω = angular velocity, rad./sec.

LITERATURE CITED

1. Altman, P. L., ed., "Blood and Other Body Fluids," Fed. Amer. Soc. Exp. Biology, Washington, D. C. (1961).
2. Bernstein, E. F., P. L. Blackshear, and K. H. Keller, *Am. J. Surg.*, **114**, 126-138 (1967).
3. Blackshear, P. L., F. D. Dorman, and J. H. Steinbach, *Trans. Am. Soc. Artificial Internal Organs*, **2**, 112 (1965).
4. ———, E. J. Maybach, A. Singh, and R. E. Collingham, *ibid.*, **12**, 113 (1966).
5. Burton, A. L., "Physiology and Biophysics of the Circulation," Year Book Medical Publishers, Inc., Chicago, Ill. (1965).
6. Fok, F. P., and H. Schubothe, *Brit. J. Haematol.*, **6**, 355 (1960).
7. Gabrio, B. W., and C. A. Finch, *J. Clin. Invest.*, **33**, 242 (1954)
8. Goldsmith, H. L., *Federation Proc.*, **26**, No. 6 (1967).
9. Guccione, E., *Chem. Eng.*, 107 (Jan., 1967).
10. Katchalsky, A., O. Kedem, C. Klibansky, and A. De Vries, in "Flow Properties of Blood and Other Biological Systems," A. L. Copley and G. Stainsbt, ed., Pergamon Press, New York (1960).
11. Kusserow, B. K., and L. W. Kendall, *Trans. Am. Soc. Artificial Internal Organs*, **9**, 262 (1963).
12. Nevaril, C. G., E. C. Lynch, C. P. Alfrey, and J. D. Hellums, *J. Lab. Clin. Med.*, **71**, 784 (1968).
13. Nevaril, C. G., Ph.D. thesis, Rice Univ., Houston, Tex. (May, 1967).
14. Page, L. B., and P. J. Culver, "A Syllabus of Laboratory Examinations in Clinical Diagnosis," Harvard Univ. Press, Boston, Mass. (1960).
15. Ponder, E., "Hemolysis and Related Phenomena," Grune & Stratton, New York (1948).
16. Rand, R. P., *Biophys. J.*, **4**, 303 (1964).
17. Schlichting, H., "Boundary Layer Theory," p. 440, McGraw-Hill, New York (1960).
18. Schubothe, H., and F. P. Fok, *Brit J. Haematol.*, **6**, 350 (1960).
19. Shapiro, S. I., M.S. thesis, Univ. Calif., Berkeley (1968).
20. ———, S. Landaw, H. S. Winchell, and M. C. Williams, *Proc. Soc. Expt. Biol. Med.*, **131**, 1206 (1969).
21. Stewart, J. W., and M. F. Sturridge, *Lancet*, **1**, 340 (1959).

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APPENDIX

The hemoglobin concentration in the plasma was analyzed by using the benzidene method (*14*). This analysis determines the number of milligrams of hemoglobin per 100 cc. plasma,

* The acid citrate dextrose (ACD) added to Red Cross blood acts as both an anticoagulant and as a sugar source. In most of this study, the anticoagulant was heparin, and no sugar was added.

known as the MG % of the plasma solution. To calculate the percentage hemolysis due to controlled testing H, it is necessary to subtract from the final value the plasma hemoglobin concentration that was present prior to shearing. This reference value reflects the hemoglobin release due to the blood drawing and pipetting techniques and also the autohemolysis during the blood storage period. The method of calculating H is outlined below:

$$H \equiv \left[\frac{\text{MGM Hgb in final plasma} - \text{MGM Hgb in Ref. plasma}}{\text{MGM Hgb at risk}} \right] \times 100$$

$$H = \left[\frac{\left\{ \frac{(\text{MG \%})_f - (\text{MG \%})_{\text{Ref}}}{100} \right\} \times \left(\frac{\text{cc. plasma}}{\text{in blood sample}} \right)}{325^{*} \frac{\text{MGM Hgb}}{\text{cc. RBC}} \times \left(\frac{\text{cc. RBC}}{\text{in blood sample}} \right)} \right] \times 100$$

$$H = \left[\frac{(\text{MG \%})_f - (\text{MG \%})_{\text{Ref}}}{325} \right] \left(\frac{\text{cc. plasma}}{\text{cc. RBC}} \right)$$

or

$$H = \left[\frac{(\text{MG \%})_f - (\text{MG \%})_{\text{Ref}}}{325} \right] \left(\frac{1 - \mathcal{H}}{\mathcal{H}} \right)$$

* From reference 1.

The Calculation of the Critical Locus Curve of a Binary Hydrocarbon System

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A method of calculating the critical properties of binary hydrocarbon systems is presented which is based upon the rigorous thermodynamic equations for the critical point of a binary mixture. By using these equations together with an equation of state, a completely analytical procedure was developed with the aid of a digital computer. The Redlich-Kwong and the Dieterici equations of state were chosen for study. The Redlich-Kwong equation was found superior for predicting critical pressures and temperatures by this method, although the Dieterici equation was better for critical volumes. The two interaction parameters arising from the equation of state were calculated from combining rules or from available experimental critical data on binary systems. To facilitate the latter approach, a mathematical optimization routine was used to find the best values of the interaction parameters for twenty-one binary hydrocarbon systems for which critical data were experimentally determined. The optimum values of the interaction parameters were correlated as functions of the ratio of molecular weights of the components. These correlations enable one to predict quite precisely the critical properties of the binary systems from the pure component data alone.

The critical properties of chemical compounds and their mixtures are of major importance in engineering calculations and design. The critical point defines the temperature and pressure where the liquid and vapor phases have identical properties and is, therefore, a key point in the construction of the phase diagram of a mixture. Also, with a knowledge of the critical properties of the pure components and with the aid of van der Waals' theorem of corresponding states, it is possible to predict the thermodynamic properties of the compounds when these properties have not been determined experimentally. For a number of years, this laboratory has had a continuing research program to determine the factors affecting the critical properties of binary systems. Most of these studies have been carried out on hydrocarbon systems which were chosen to investigate the effect of molecular size, structure, and chemical nature of the components. For this paper, the critical locus curves of twenty-one binary systems composed of mixtures of paraffinic, naphthenic, and aromatic hydrocarbons have been used to develop a method of calculating the critical properties of a mixture of known composition from a knowledge of the critical properties of the pure components. The systems and references

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