allowing the effect of the thromboplastin to become the dominant one in the dialysed urine.

Summary. The clear protein and cell-free urine from normal men and women and from hemophilic men has thromboplastic activity when tested on normal and hemophilic blood. The thromboplastin in dialysed and lyophilized urine seems more resistant to heat than that in fresh urine. The lyophilized dialysed urine residue has a clot accelerating action on hemophilic blood *in vitro* and *in vivo*. The clots which form in the ureter of hemophilics with hematuria are probably due to contact of the extravasated blood with the thromboplastin of the urine.

15861 P

"Control" Erythrocytes for Hemolysis Studies.*

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In several previous experiments the permeability of chicken erythrocytes at 37.5°C was studied over periods of many hours' duration. It was noted that the time for hemolysis in glycerol of untreated cells became less after several hours (Hunter).[1] Since no attempt was made in these experiments to control bacterial contamination, it was thought that a part of the change in these control cells might have resulted from bacterial action. In another series of experiments, however, in which aseptic technics were followed it was also observed that the time for hemolysis of control cells decreased with lapse of time (Hunter and Larsh).[2] Results of this nature might have been predicted in view of previous experiments. Jacobs and Parpart[3] have shown that erythrocytes are quite sensitive to environmental changes while Harris[4] showed that under certain conditions of standing, erythrocytes lose potassium.

First a series of experiments were performed in which blood was allowed to stand at 37.5°C and time for hemolysis in glycerol was measured at varying intervals of time. Aseptic technic was observed throughout and tests were made for bacterial contamination as before (Hunter and Larsh).[2] Four stock suspensions were made. (1) Two cc of freshly drawn, heparinized chicken blood, (2) 1 cc of blood plus 1 cc of Ringer Locke, (3) 1 cc of cells plus 1 cc of plasma and (4) 1 cc of cells plus 1 cc of Ringer Locke. The results obtained using whole blood are shown in Fig. 1. Essentially similar results were obtained with all 4 solutions except the Ringer Locke appeared to exert a "protective effect" especially in (4). A similar series was run using blood which had been in the refrigerator for 27 hours. Hematocrit readings indicated that these cells had swollen although the initial hemolysis times and subsequent

TABLE I.
Effect of Standing on Hemolysis Time and Volume.

Time of exposure in hr	Time in sec for 50% hemolysis	Cell volume in μ^3
0	285	90
12⅓	245	104
0	395	112
23½	200	123
0	300	118
29¾	160	138

* The authors are indebted to the Faculty Research Fund of the University of Oklahoma for grants in aid.

1 Hunter, F. R., *J. Cell. and Comp. Physiol.*, 1947, in press.

2 Hunter, F. R., and Larsh, Howard W., to be published.

3 Jacobs, M. H., and Parpart, A. K., *Biol. Bull.*, 1931, **60**, 95.

4 Harris, J. E., *J. Biol. Chem.*, 1941, **141**, 579.

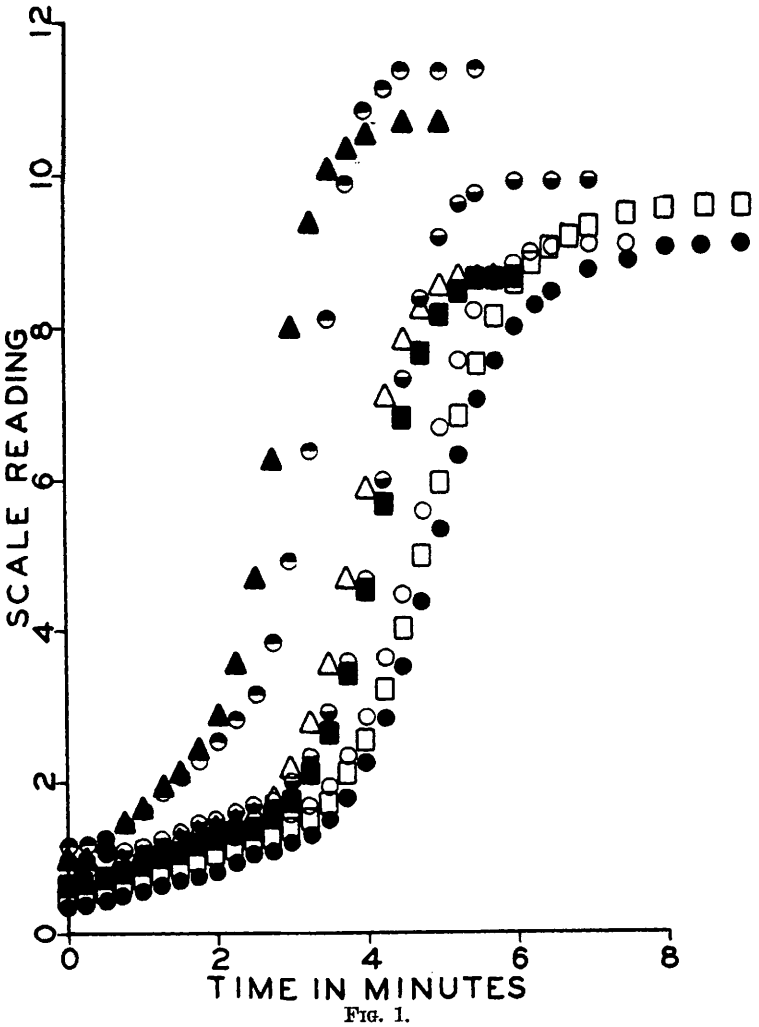

FIG. 1.

Effect of time on the hemolysis of chicken erythrocytes in glycerol. □—0 hr; ●—¾ hr; ○—3½ hr; ■—7½ hr; △—10¼ hr; circle with solid bottom—12¾ hr; circle with solid top—28¾ hr; ▲—51¾ hr.

changes paralleled those obtained with the freshly drawn blood.

A second series of experiments involved hemolysis measurements, cell counts (Parpart),[5] and hematocrit determinations using an air turbine. These data are presented in Table I. It can be seen that with the passage of time at 37.5°C the cells swell as well as hemolyze more rapidly.

To determine whether the swelling could account for the change in rate of hemolysis, equal volumes of freshly drawn blood and 80% Ringer Locke (80 cc of Ringer Locke plus 20 cc of water) were mixed. At the end of 10 minutes the measurements were made. These data are presented in Table II.

As expected, the cells in the 80% Ringer Locke swelled but the hemolysis time at 0 hours was unaffected. After a lapse of 20-24 hours, these cells increased in volume but to a lesser extent than the untreated cells. The change in hemolysis times of these treated cells was less than for the untreated.

The fact that erythrocytes in whole blood maintained at 37.5°C under aseptic condi-

[5] Parpart, A. K., *Biol. Bull.*, 1931, **61**, 500.

TABLE II.
Effect of Initial Swelling on Hemolysis Time.

Time of exposure in hr	Time in seconds for 50% hemolysis		Cell volume in μ^3	
	Plasma	80% R.L.	Plasma	80% R.L.
0	345	350	112	128
20	200	230	126	139
0	365	370	123	132
23¼	190	220	136	139
0	340	340	125	143
24	205	240	146	160

tions change so markedly indicates additional factors which must be taken into consideration in hemolysis experiments. No satisfactory explanation is available at present for the apparent lack of effect of a slightly hypotonic environment on hemolysis time.

In summary it may be said that control chicken erythrocytes after standing at 37.5°C for several hours hemolyze more rapidly. An increase in the volume of these cells was observed but this cannot be the sole explanation for the change in rate of hemolysis.

15862

A Simple Inexpensive Pump for Perfusion of Organs with Preservatives or with Physiological Solutions.*

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In an attempt to investigate the secretory properties of human testes by means of perfusion *in vitro*, 3 types of perfusion pumps were employed but none were found to be suitable for these purposes. The Carrel-Lindbergh pump[1] proved to be so readily subject to disturbances upon change in the resistance offered by the organs during perfusion that maintenance of a proper flow of blood was difficult. The pump devised by Kleinberg and associates[2] is less complex, but in our experience the valves were prone to stick, and during long experiments trouble was encountered in stabilizing the flow of blood through the shunt and in preventing changes in the level of fluid in the compression chamber. The Fischer "Volustat"† came nearest to filling the requirements of our experiment but afforded no means for regulation of the rate of pulsation.

Study of the mechanical principles of these and other pumps, *e.g.*, the Dale and Schuster apparatus,[3] suggested the need for a pump with the following characteristics: (a) ready construction at small cost; (b) sufficient simplicity in operation as to provide rapid and satisfactory perfusion of an organ with physiological solutions or with preservative fluids; (c) no need of continuous adjustment during prolonged periods of perfusion; (d) the capacity to mimic the action of the heart by providing any desired rate of pulsation

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1 Carrel, A., and Lindbergh, C., *The Culture of Organs*, Paul B. Hoeber, Inc., New York, 1938.

2 Kleinberg, W., Gordon, A., and Charipper, H., *J. Lab. Clin. Med.*, 1943, **28**, 1484.

† Fischer, "Volustat," Eimer and Amend catalogue 90, No. 13-684.

3 Dale, H., and Schuster, E., *J. Physiol.*, 1928, **64**, 356.