A COMPARISON OF ERYTHROCYTE SEDIMENTATION RATES AND ELECTROPHORETIC PATTERNS OF NORMAL AND PATHOLOGICAL HUMAN BLOOD

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The suspension stability of blood, as measured by the rate of sedimentation of erythrocytes in plasma, has had wide application in medicine since Fåhraeus (1) published the results of his investigations on the subject some twenty years ago. It is well known that the sedimentation rate is increased markedly in many pathological conditions, notably in acute infections. However, although a voluminous literature has grown, dealing with many aspects of this subject, the mechanism responsible for the observed differences in the sedimentation rates of normal and pathological blood is not well understood.

Various workers have called attention to an apparent correlation between increases in the sedimentation rate and corresponding increases in serum globulin or fibrinogen levels. An excellent survey of the subject was made by Ham and Curtis (2), in which they discussed various techniques for measuring sedimentation rates, the influence of erythrocyte size and total volume on the rate of fall, and correlation with fibrinogen levels.

In this paper, we shall report the results of studies on human blood in a number of pathological conditions, obtained by the method of electrophoresis. These results have been correlated with corresponding observations on the erythrocyte sedimentation rates.

Materials and Methods

Sedimentation rate measurements were performed on samples of blood[1] taken into tubes containing a dry mixture of potassium and ammonium oxalates, as recommended by Heller and Paul (3). In some cases heparin (Connaught Laboratories) was used as an anticoagulant instead of the oxalate mixture; about 0.1 mg. of this material was sufficient for 10 cc. of blood. After thorough but gentle mixing the blood was introduced into Rourke-Ernstene (4) tubes, graduated at 2 mm. intervals over a length of 100 mm., containing about 1.25 ml. The tubes were suspended in an accurately vertical position in a glass cylinder of water, the temperature of which

[1] Fasting samples of blood were drawn from the antecubital vein with as little stasis as possible. The sedimentation rate was measured within 2 hours after collection.

remained quite constant, the work being done in a thermostated room at 25°C. Readings of a clock were taken as the erythrocyte boundary came to succeeding graduation marks. The region of uniform rate of fall determined the (uncorrected) sedimentation rate. The partial volume occupied by the cells (hematocrit) was measured after centrifuging the tubes for 30 minutes at 3000 R.P.M. The corrected sedimentation rate[2] was then obtained with the aid of charts published by Rourke and Ernstene (4).

Electrophoretic studies were carried out in the Tiselius apparatus, using the scanning method of Longsworth (5). The technique has been described in detail by Longsworth, Shedlovsky, and MacInnes (6). All the determinations were made in diethylbarbiturate buffer solutions at pH 7.8–7.9 and an ionic strength of 0.05, on samples of plasma or serum which had been diluted with 3 volumes of buffer solution against which they were then dialyzed.

RESULTS AND DISCUSSION

In the paper to which we have just referred (6), electrophoretic determinations were made on the plasma or serum from a number of normal individuals, as well as on that of patients suffering from aplastic anemia, rheumatic fever, pneumonia, peritonitis, peritonsillar abscess, acute lymphatic leukemia, lymphogranuloma, obstructive jaundice, lipoid nephrosis, and multiple myeloma. The corresponding sedimentation rates were determined by us, but were not published at that time. The present electrophoretic studies have extended the work to include more normals and cases of tuberculosis, coronary thrombosis, duodenal ulcer, nephritis, arthritis, lymphosarcoma, burns, fractures, and chemically induced shock in treating insanity. The results are summarized in Table I, and some of the corresponding electrophoretic patterns are shown in Figs. 1 and 2. In columns 7 to 10 of the table are given the values for the ratios of α globulin, β globulin, γ globulin, and ϕ (fibrinogen) to albumin, respectively. The concentrations of albumin in the plasma (or serum) appear in column 5, and the albumin:globulin ratios in column 6. The corrected erythrocyte sedimentation rates (E.S.R.) are listed in column 11. The concentrations of the various components were obtained from the areas under the corresponding electrophoretic peaks, such as are shown in Figs. 1 and 2. The values found for the normals in the present series agree closely with those found by Longsworth, Shedlovsky, and MacInnes (6). It was pointed out by these authors that the most striking and general change in the electrophoretic patterns of pathological serum is reflected in the α globulin levels, which appear

[2] The rate of fall of particles in a fluid contained in a tube of finite length is determined in part by the partial volume of the particles, since they fall against a countercurrent of the fluid which must rise to replace the space formerly occupied by the particles. The counterforce thus exerted against the falling particles depends on the relative volumes of the particles and of the fluid.

to be significantly increased in cases of various febrile infections. The present results confirm these findings and also indicate that they hold true in cases of

TABLE I

Composition of Normal and Pathological Serum and Plasma

Corrected Erythrocyte Sedimentation Rate

No.	Material	Age	Sex	Albumin	A/G	α/A	β/A	γ/A	φ/A	E.S.R.	Temp. a.m.	Remarks
				per cent						*mm./min.*	°F.	
1	Normal	9	M	3.45	2.27	0.11	0.18	0.15	—	0.10		
2	Normal	40	M	4.06	1.75	0.16	0.25	0.16	—	0.44		
3	Normal	38	F	4.00	2.00	0.14	0.17	0.19	—	0.50		
4	Oligophrenia	38	F	4.66	1.92	0.11	0.18	0.23	0.10	0.40		Phenyl pyruvic acid
5	Psychopathic personality	22	F	5.05	2.00	0.16	0.12	0.22	0.07	0.30	99.6	During metrazol shock
6	Schizophrenia simplex	16	F	4.33	2.00	0.17	0.14	0.19	0.10	0.55	96.0	During insulin shock
7	Fracture of tibia and fibula	72	M	3.92	2.13	0.17	0.16	0.14	0.11	0.70	98.6	3 days after accident
8	Fracture of femur	40	M	4.21	1.39	0.21	0.30	0.21	—	1.40	101.2	2 days after accident
9	Burns, first, second, and third degree	55	F	3.77	1.04	0.36	0.36	0.25	0.14	2.60	101.0	6 days after accident
10	Duodenal ulcer	58	M	4.55	1.56	0.20	0.25	0.19	—	1.20	98.6	Complicated with hemorrhage and obstruction
11	Coronary thrombosis	42	M	3.34	1.07	0.34	0.32	0.27	—	1.40	99.4	1 week after initial attack
12	Coronary thrombosis	43	M	3.40	1.54	0.19	0.30	0.16	—	1.00	98.4	6 weeks after initial attack
13	Tuberculosis	57	M	4.41	1.11	0.19	0.36	0.35	—	1.80	98.6	Chronic pulmonary
14	Tuberculosis	34	M	3.31	0.90	0.34	0.34	0.43	—	1.30	102.0	Bilateral pulmonary, far advanced
15	Tuberculosis	39	M	2.95	0.83	0.30	0.34	0.57	0.21	1.80	102.5	Bilateral pulmonary, far advanced
16	Tuberculosis	40	M	3.85	1.10	0.17	0.30	0.34	0.10	0.40	100.0	Miliary, tuberculous meningitis
17	Tuberculosis	38	M	3.11	1.10	0.33	0.34	0.56	0.24	2.20	100.0	Bilateral pulmonary, far advanced
18	Neoplastic disease	36	F	3.80	1.01	0.19	0.29	0.51	—	1.20	98.0	X-ray: general skeletal involvement
19	Lymphosarcoma	14	F	3.22	0.70	0.34	0.19	0.91	0.42	1.90	98.0	Febrile. Biopsy
20	Chronic nephritis	30	F	3.30	1.33	0.39	0.20	0.26	0.10	1.40	98.4	Toxemia of pregnancy. Post partum
21	Arthritis	18	F	3.77	1.19	0.34	0.25	0.26	—	3.00	101.4	Gonococcus, 9 day history

extensive tissue destruction, as evidenced by the patterns for coronary thrombosis, burns, and fractures[3] (Figs. 1 and 2). No significant deviation from normal in either sedimentation rate or electrophoretic patterns was found in the cases (4, 5, 6 of Table I) of chemically induced shock in demented patients.

Various authors have reported that good correlation exists between sedi-

[3] Determinations on the plasmas of three normal individuals whose temperatures were raised by artificial fever to 106°F. for 40 minutes showed no increase in α globulin.

mentation rates and fibrinogen levels (2). However, the addition of purified fibrinogen to normal blood fails to increase the rate of sedimentation of eryth-

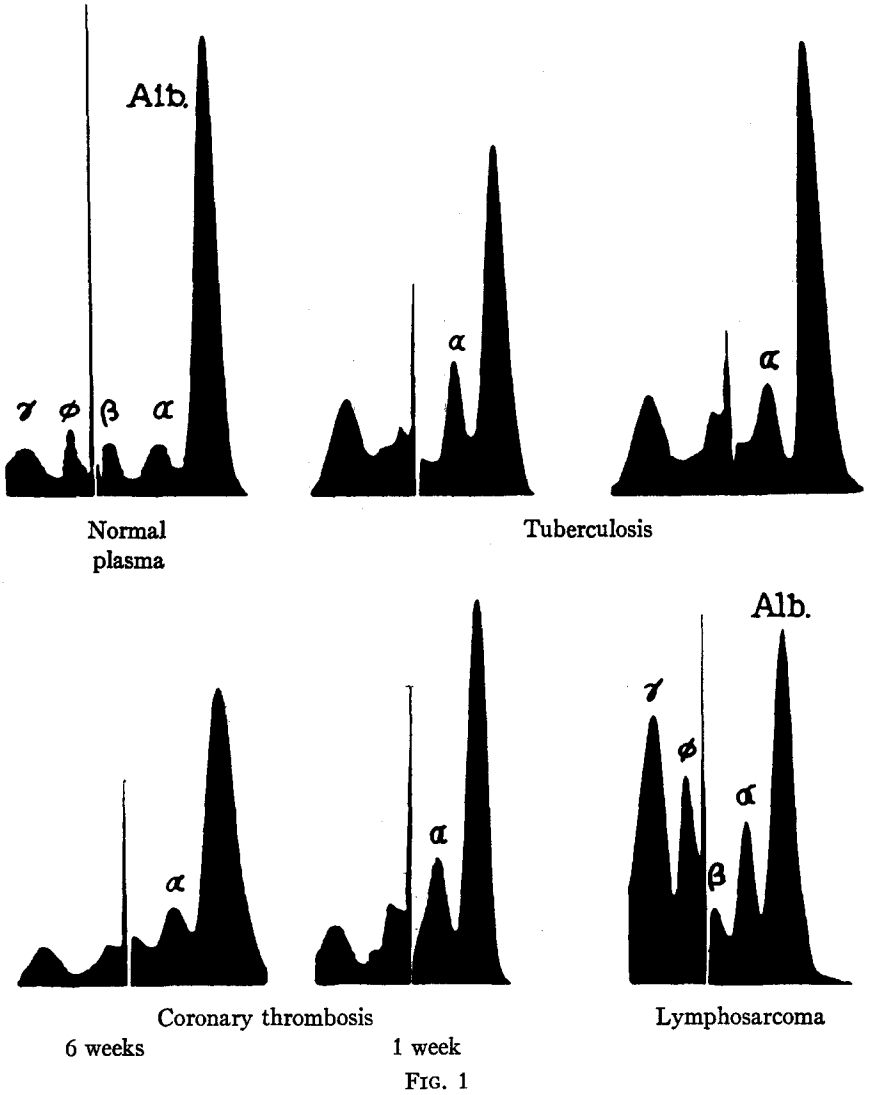
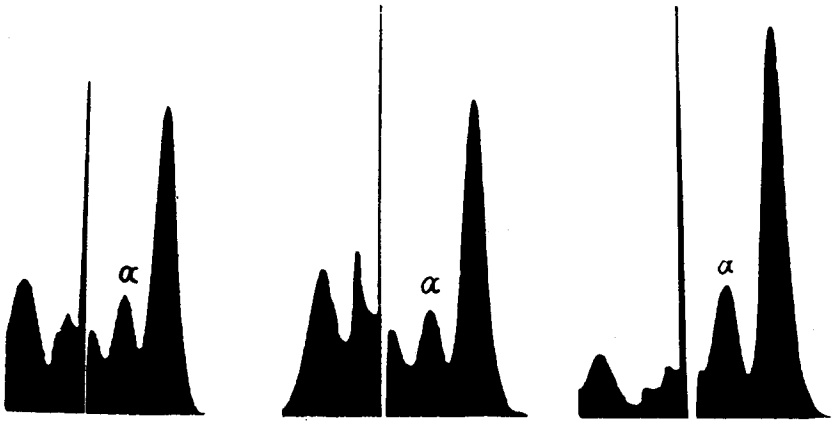


FIG. 1

FIGS. 1 and 2. Electrophoretic patterns on normal human plasma and on plasma from individuals with tissue injury from various causes.

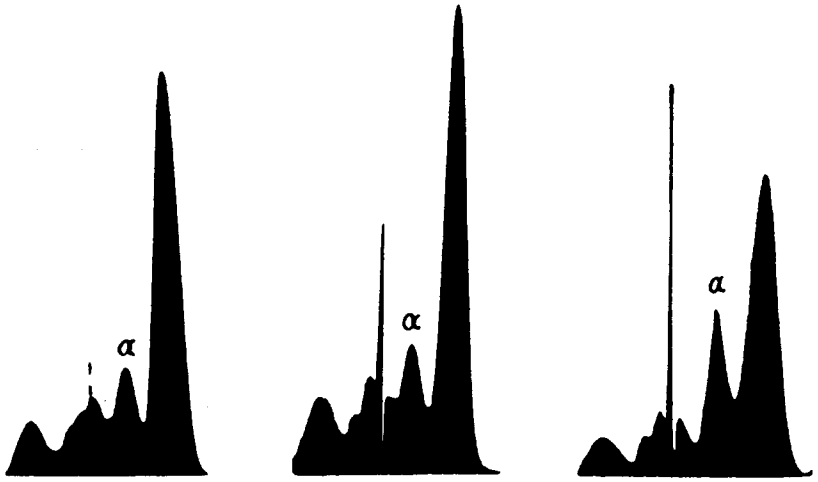
rocytes to the same extent as one observes in pathological blood containing comparable quantites of this protein. In Fig. 3, we have plotted the sedimentation rate (E.S.R.) against corresponding levels of fibrinogen expressed as fibrinogen:albumin ratios (ϕ/A), using the results of Ham and Curtis (2).

It will be observed that only a qualitative correlation exists between these two factors.



Tuberculosis

Toxemia of pregnancy



Bleeding duodenal ulcer

Fracture of femur

Multiple burns

FIG. 2

We have found no satisfactory similar correlation with either β globulin, γ globulin, or albumin:globulin ratios. However, as will be shown below, a significant correlation can be demonstrated between sedimentation rates and the corresponding α globulin present in the blood.

In Fig. 4 are plotted the α globulin levels, expressed as α globulin:albumin

ratios, (α/A), against the corresponding corrected sedimentation rates (E.S.R.). In this graph have been included points corresponding to the results reported by Longsworth, Shedlovsky, and MacInnes (6) as well as our more recent determinations. Although it is not possible to draw a smooth curve through the points indicated, any more successfully than in Fig. 3, an in-

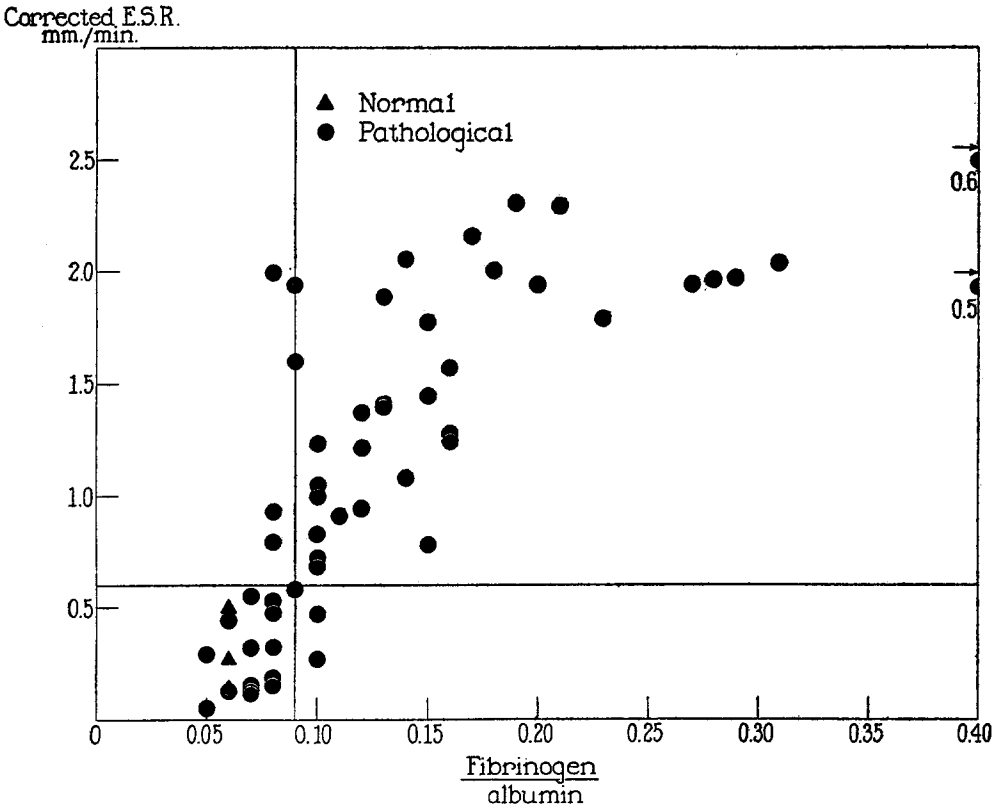


FIG. 3. Variation of corrected erythrocyte sedimentation rates with fibrinogen levels.

teresting relationship appears. By drawing a horizontal line corresponding to the upper limit of normal sedimentation rate and a vertical line corresponding to the upper limit of normal α/A values (α/A = 0.17), we find all the points corresponding to the normals, as well as those for nearly all the pathological conditions which yield normal electrophoretic patterns, in the lower left hand quadrant. The other points, corresponding to elevated sedimentation rates, fall in the upper right hand quadrant. If we were to draw another vertical line at about α/A = 0.20, all the points in this more restricted upper right

hand quadrant, with one exception, correspond to febrile conditions. The significance of fibrinogen and of α globulin in the mechanism responsible for increased sedimentation rates in blood will be discussed in another paper.

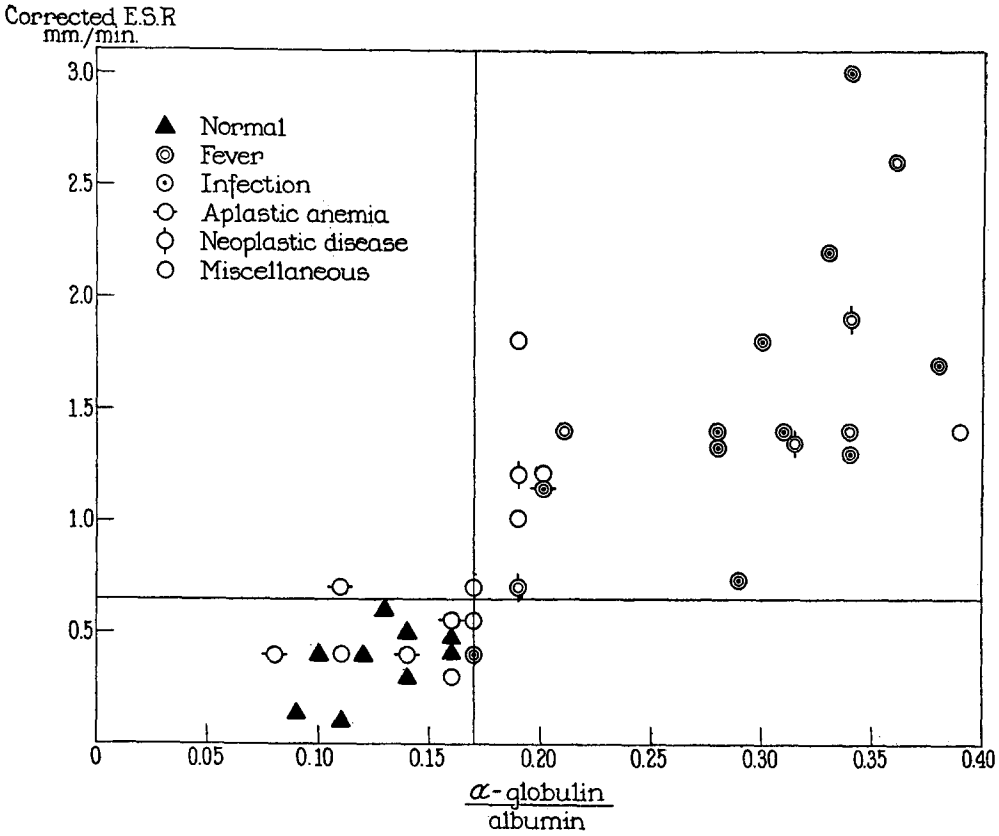


FIG. 4. Variation of corrected erythrocyte sedimentation rates with α globulin levels.

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SUMMARY

Electrophoretic studies and erythrocyte sedimentation rate measurements were carried out on normal and pathological human blood. An increase in α globulin levels appears to take place, as well as an increase in sedimentation

rates, when there is present any considerable inflammation or tissue destruction, irrespective of its cause. A graphic correlation is presented between sedimentation rates and α globulin levels, which is at least as good as a similar correlation involving fibrinogen levels.

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