

blood from dog 38-112, an anemic animal with no circulating radioactive iron, was centrifugalized. Plasma was discarded. The cells were laked in distilled water and the stroma precipitated partially by the addition of 18 per cent. sodium chloride. The supernatant solution, largely hemoglobin, was separated and 2 ml of iron labelled with the radioactive isotope in the form of ferric ammonium citrate were added. After standing in the refrigerator for 24 hours the mixture was poured into aluminum cream to precipitate remaining stroma and any other protein material other than hemoglobin. Perhaps 15 per cent. of the original hemoglobin was lost in this step. The filtrate was poured into an equal volume of 10 per cent. trichloroacetic acid to precipitate hemoglobin and this precipitate was washed and ashed for determination of radioactivity.

EXPERIMENTAL DATA

Iron content of hemoglobin from 90 ml of blood	22 mg
Labelled iron added as ferric ammonium citrate	0.34 mg
Activity of radioactive iron added	66 counts per min.
Net activity of radioactive iron in hemoglobin	0.1 counts per min.

The amount of labelled iron found in the hemoglobin is within the experimental error. It is estimated that radioactive iron equal in activity to 0.4 counts per minute might have been present and escaped detection. This sets an upper limit of 1 per cent. on the amount of exchange which might have taken place and escaped detection under the conditions of the experiment.

These findings are supported by other experiments in which solutions of hemoglobin were tested with artificial solutions of radio-iron in plasma. There was no evidence of any exchange between the hemoglobin iron and the radio-iron in solution. These experiments were done before the final tests recorded in Table I.

P. F. HAHN
W. F. BALE
J. F. ROSS
R. A. HETTIG
G. H. WHIPPLE

VISCOSITY AND THE SHAPE OF PROTEIN MOLECULES

IN some recent publications,^{1, 2, 3, 4} attempts have been made to interpret protein viscosity data in terms of the asymmetry of the molecules. This has been

done by applying equations developed by Burgers⁵ and Eizenschitz,⁶ or by Kuhn,⁷ on the assumption that there is no orientation of the molecules. The discrepancies between these values for the asymmetry and the values obtained by applying the equations of Perrin⁸ to sedimentation velocity and diffusion data are considerable. It has been suggested that the differences arise from the failure to consider the effects of hydration. Recently the hydrodynamic behavior of ellipsoids has been reconsidered by Simha.⁹ The results obtained seem to yield better agreement for the asymmetries of various proteins than previous equations without any assumption of hydration.

In the expression for the specific viscosity at infinite dilution,

$$\eta/\eta_0 - 1 = \nu c \quad (1)$$

(where c is the volume fraction of the solute), ν is a function of the axial ratio, ρ , of an ellipsoid of revolution. Table I gives the numerical values of the factor

TABLE I

$1/\rho$ or ρ	ν		$1/\rho$ or ρ	ν	
	elongated	flattened		elongated	flattened
1.0	2.50	2.50	20.0	38.6	14.80
1.5	2.63	2.62	25.0	55.2	18.19
2.0	2.91	2.85	30.0	74.5	21.6
3.0	3.68	3.43	40.0	120.8	28.3
4.0	4.66	4.06	50.0	176.5	35.0
5.0	5.81	4.71	60.0	242.0	41.7
6.0	7.10	5.36	80.0	400.0	55.1
8.0	10.10	6.70	100.0	593.0	68.6
10.0	13.63	8.04	150.0	1222.0	102.3
12.0	17.76	9.39	200.0	2051.0	136.2
15.0	24.8	11.42	300.0	4278.0	204.1

ν for values of $1/\rho$ and ρ from 1 to 300. These values have been applied to the same viscosity data as those used by Polson¹ and by Mark and Simha.² In Table II the axial ratios are compared with those from sedi-

TABLE II

Protein	f/f_0	ν	$1/\rho$, elongated		ρ , flattened	
			Dif-fusion	Vis-cosity	Dif-fusion	Vis-cosity
Egg albumin ...	1.17	5.7	3.8	5.0	4.0	6.7
Serum albumin..	1.25	6.5	5.0	5.6	5.4	7.7
Hemoglobin	1.16	5.3	3.7	4.6	3.9	6.0
Amandin	1.28	7.0	5.4	6.0	6.0	8.5
Octopus hemo-cyanin	1.38	9.0	7.2	7.3	8.2	11.4
Gliadin	1.60	14.6	10.9	10.5	13.6	21
Homarus hemo-cyanin	1.27	6.4	5.2	5.5	5.8	7.5
Helix pom. hemo-cyanin	1.24	6.4	4.8	5.5	5.2	7.5
Serum globulin .	1.41	9.0	7.6	7.3	8.9	11.4
Thyroglobulin ..	1.43	9.9	7.8	7.9	9.2	12.7
Lactoglobulin ..	1.26	6.0	5.2	5.1	5.7	6.9
Pepsin	1.08	5.2	2.5	4.5	2.6	5.8
Helix hemocyanin pH 8.6	1.89	18.0	16.6	12.0	23.9	26

⁵ J. M. Burgers, "Second Report on Viscosity and Plasticity," Amsterdam, 1938.

⁶ R. Eizenschitz, *Zeits. physik. Chem.*, A163: 133, 1933.

⁷ W. Kuhn, *Zeits. physik. Chem.*, A161: 1, 1932.

⁸ F. Perrin, *Jour. Phys. et Rad.*, 7: 1, 1936.

⁹ R. Simha, *Jour. Phys. Chem.*, 44: 25, 1940.

¹ A. Polson, *Koll. Zeits.*, 88: 51, 1939.

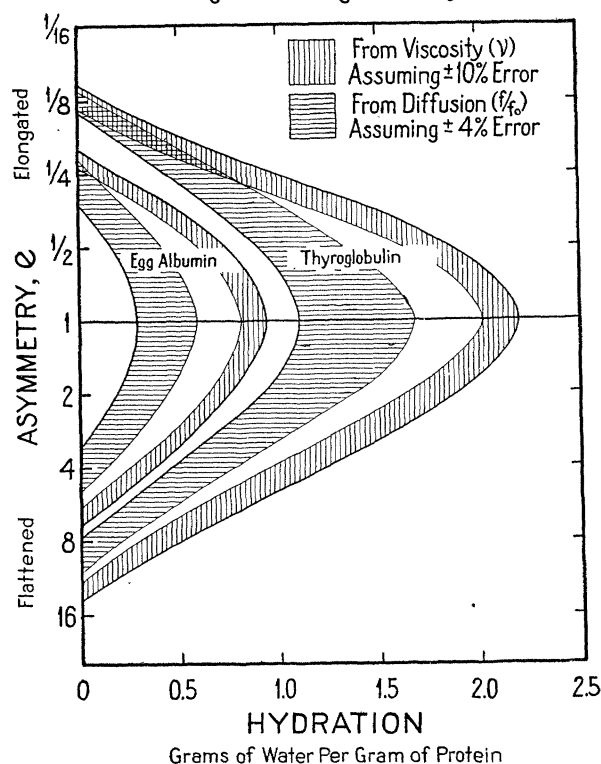
² Mark and Simha, *Nature*, 145: 571, 1940.

³ V. L. Frampton and H. Neurath, *SCIENCE*, 87: 468, 1938.

⁴ M. A. Lauffer, *SCIENCE*, 87: 469, 1938.

mentation and diffusion. The agreement seems to be generally good on the assumption that the protein molecules can be approximated by rigid elongated ellipsoids.

The Asymmetry of Protein Molecules Calculated from Viscosity and Diffusion Assuming Various Degrees of Hydration



In an attempt to evaluate the importance of hydration and to determine whether any choice can be made

between the elongated and flattened shapes, values of ρ were calculated from viscosity and from sedimentation and diffusion, assuming that the protein carried varying amounts of water. It also seemed desirable to consider the influence of experimental error, so that the values of f/f_0 were taken to be subject to ± 4 per cent. error, and values of v to ± 10 per cent. error. The areas included between pairs of such curves would then include the possible choices for shape and hydration. Such curves for egg albumin¹⁰ and thyroglobulin are given in Fig. 1. They were chosen as well-characterized examples of fairly symmetrical and fairly asymmetrical molecules. The shape of these curves is such that they never cross sharply, but it can be said that, for these two cases, hydration greater than 0.5 grams of water per gram of protein, and the flattened shape, seem relatively improbable.¹¹

It should be further emphasized that all the equations mentioned above only apply to measurements of the viscosity which are made in the region of complete Brownian movement. Any orientation will reduce v , and hence $1/\rho$ or ρ if these equations are misused. This may be well illustrated by the case of tobacco mosaic virus, where measurements in capillary tube viscometers give values of v of about 80.^{3, 4} However, this must be much too low, as it is known that double refraction of flow is obtained under such conditions of flow. Robinson¹² has made measurements in a Couette viscometer and found that v approaches 1,500 as the velocity gradient approaches zero. This would correspond to an axial ratio of about 160, assuming a rod-like shape.

J. W. MEHL
J. L. ONCLEY

HARVARD MEDICAL SCHOOL

R. SIMHA

COLUMBIA UNIVERSITY

SCIENTIFIC APPARATUS AND LABORATORY METHODS

AN ELECTRONIC RELAY FOR HEAT CONTROL¹

THERE are a large number of biological and chemical processes whose study requires the temperature of the materials to be held within very narrow limits. While several methods for temperature control have been described they have the disadvantage of being rather elaborate and expensive when the temperature is to be held to 0.02°C . To overcome these disadvantages, we developed the electronic relay whose circuit is given in Fig. 1. It controls a bath temperature to 0.02°

¹ Contribution from the Department of Electrical Engineering and the Research Laboratory of Physical Chemistry, No. 451, Massachusetts Institute of Technology.

$\text{C}.$,² requires only a 115-volt power source which can be either a-c or d-c, and permits only a few microamperes to pass through the thermoregulator contacts. The parts of the apparatus were bought at a radio shop for less than six dollars.

The device operates as follows: When the temperature of the thermostat is too low, the circuit through

¹⁰ The value of v chosen for egg albumin for this calculation was 5.4, rather than 5.7 as used by Polson. The choice of this value is discussed by Oncley, *Proc. N. Y. Acad. Sci.*, in press.

¹¹ For a discussion of solvation on the basis of viscosity measurements, see also H. Mark and R. Simha, *Jour. Phys. Chem.*, in press.

¹² J. R. Robinson, *Proc. Roy. Soc. Lond.*, A170: 519, 1939.

² Heidt, *Jour. Am. Chem. Soc.*, 61: 3455, 1939.