THE PHOTO-EFFECT OF SERUM ALBUMIN.

Chiefly on Its Viscosity and Solubility.

By Nagao Hayami.

In the course of his research on binding materials, the author found that serum albumin presented turbidity in its exposure of light and at the same time showed some decrease of its binding force. In order to make clear this phenomenon, he mode the measurement of the viscosity by means of the viscosimeter he had devised and the solubility change of serum albumin with time. Here will be reported the effect in question with cow serum albumin.

(A) Materials and Experimental Method.

(I) Preparation of serum albumin.

The composition of various animals' blood have hitherto been reported and there has not been found much difference among them, as found by the author and Lewinsky.

The sample, dried serum albumin, was prepared thus: serum albumin was separated from cow's blood through decantation and dried in vacuum. The drying treatment was performed by the two methods—in the dark and in the light. The former was done in a dark room, while the latter was as follows. The vacuum drying system is shown in Fig. 1.

About 100 c.c. of serum albumin on a watch glass (6) was put in a desiccator (5). On its lid (4) was placed a Wratten light filter (3), on which the filter (2) to absorb the heat rays was surmounted. All of these kept in a dark box (1), to which the connecting tube to a vacuum pump (7) and a water pipe (8) were connected. Then the lid of the dark box (9) being removed, the box was exposed to the sunlight (10). After the drying course, every sample was stored in a dark room.

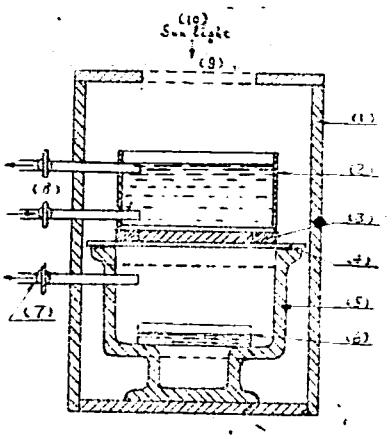

Fig. 1.

(II) Apparatus for measuring viscosity.

(1) Characteristics of the viscosimeter.

For the present experiment, the author

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devised a viscosimeter. There are many kinds of viscosimeters and no viscosimeter suitable for any photochemical reaction. But the author's viscosimeter satisfied the following conditions:—

(a) Measurement without being exposed to light.
(b) Wide range of measurement.
(c) The materials of the viscosimeter itself free from any chemical reaction and also from the action of light.
(d) Its reliability higher than hitherto proposed.

(2) Structure and measuring method of the viscosimeter.

The viscosimeter is graphically shown in Fig. 2.

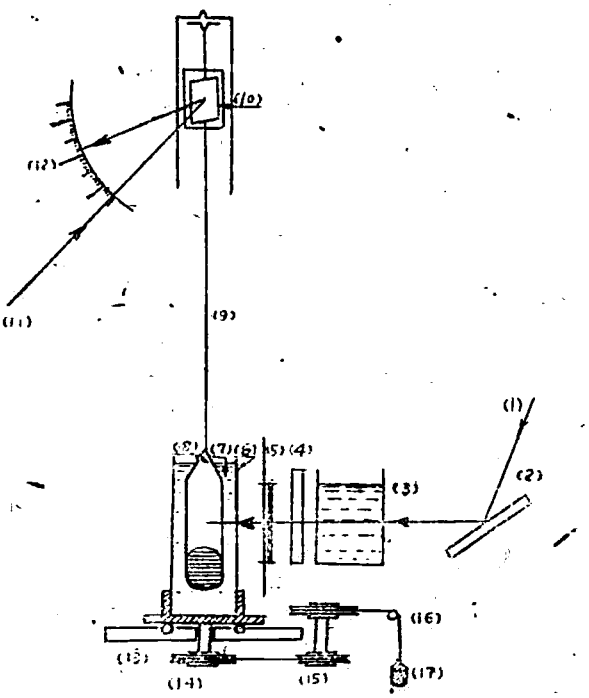

Fig. 2.

The Sample (7) is filled in the cylinder (6). For the exposure to light, the shutter (5) is opened and the light radiated from the light source (1) is reflected on a mirror (2) and passed through a filter (3) to absorb the heat rays. In the case of monochromatic light, Wratten light filter (4) is used. The rotation of the cylinder (6) is smoothly made on a rotatory stand with ball bearings (13) by the pulleys (14), (15) and (16) with a load (17).

The measurement of relative viscosity by means of a viscosimeter requires preliminary determination of the rotation velocity for the standard solution and

involves the measurement of its density. In this experiment, ethylalcohol, water and its aqueous solution are employed as the standard solution. In the present viscosimeter, the rotation velocity of the cylinder (8) suspended by the thread (9) is measured by the light image on the scale (12) reflected from the mirror (10) with a stop-watch. Therefore, the viscosity change of the sample can be readily detected. The characteristics of this viscosimeter is that the viscosity change with time is relatively easy to measure.

(B) Experimental Results.

The effect of light on the viscosity and solubility of serum albumin.

Taking into account the preparation of serum albumin, much attention was paid to the drying treatment. The changes of solubility and viscosity were measured for the samples treated under different conditions—solvent, temperature, atmosphere of different gases and pressures, light intensity, wave-length, etc.

(I) The effect of the sunlight on the solubility of dry blood in water.

As seen in Table I, albumin exposed to the sunlight contains the greatest amount of the substances insoluble in water. The sample dried in the sunlight, when dried in the atmosphere of nitrogen, is nearly similar to that in the dark room. This suggests that a dominant influence of light is produced in the air.

Table I. Effect of Sunlight on Solubility of Dry Blood into Water
Drying conditions: Temperature, 18°C.; Time, 48 hours; 1 atmosphere.

Atmosphere	Condition of Light	Amount of Insoluble Substances (%)
Air	Exposed directly to Sunlight	10~15
	Exposed indirectly to Sunlight	3.5~5.5
	Dark	2.0~3.0
Nitrogen	Exposed directly to Sunlight	3.5~5.0
	Exposed indirectly to Sunlight	3.0~5.0
	Dark	3.5~5.5

(II) The effect of monochromatic light.

Table II shows the amounts of the insoluble substances contained in serum albumin dried by the exposure to various kinds of monochromatic light.

The longer the wave-length of light is, the smaller the amount of the insoluble substances is. In the case of the drying in the dark, the amount is scarcely changed.

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Table II. Effect of Wave-length of Light on Solubility of Dry Blood into Water
Drying conditions: Temperature, 18°C.; Time, 48 hours;
1 atmosphere of the air.

Wave-length	Amount of Insoluble Substances (%)
~ 390	17.5 ~ 22.0
390 ~ 450	15.5 ~ 21.0
450 ~ 530	15.0 ~ 20.0
530 ~ 560	15.5 ~ 21.0
560 ~ 600	14.5 ~ 19.5
600 ~ 650	12.5 ~ 15.0
650 ~ 800	10.5 ~ 13.0
800 ~	9.5 ~ 10.5
In the dark	1.5 ~ 4.0

(III) The measurement of specific gravity of serum albumin dried in the sunlight and that dried in the dark.

The specific gravity was measured by the Ostwald's method. Three kinds of commercial dry albumin were measured for comparison. Various percentage dry albumins were immersed in water for more than 2 hours and stirred without bubbling. Then insoluble substances being filtered, the albumin thus clarified was measured. The preparation of the samples was carried out in the dark.

It is found that according to the drying process of albumin, the specific gravity

Table III. Specific Gravity of Aqueous Solution of Blood Albumin.

Albumin Concentration %	Specific Gravity (18°C)				
	Treatment in Dark (D)	Commercial Samples			Treatment in Light (L)
		M_1	M_2	M_3	
0.25	1.0012	1.0006	1.0005	1.0004	1.0003
0.50	1.0026	1.0018	1.0009	1.0008	1.0007
1.0	1.0034	1.0024	1.0011	1.0010	1.0008
2.0	1.0066	1.0045	1.0019	1.0017	1.0015
3.0	1.0105	1.0085	1.0034	1.0031	1.0026
5	1.0178	1.0134	1.0055	1.0050	1.0045
10	1.0300	1.0225	1.0095	1.0090	1.0080
15	1.0450	1.0375	1.0223	1.0218	1.0125
20	1.0655	1.0545	1.0370	1.0345	1.0270
25	1.0773	1.0635	1.0434	1.0398	1.0285
30	1.0922	1.0763	1.0515	1.0475	1.0384
35	1.1108	1.0858	1.0556	1.0505	1.0405
40	1.1225	1.0955	1.0660	1.0590	1.0425
45	1.1304	1.1105	1.0855	1.0785	1.0505
50	1.1455	1.1204	1.0995	1.0950	1.0605

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differ. The sample dried in the sunlight shows far smaller specific gravity than that dried in the dark and any commercial samples.

(IV) The measurement of the relative viscosity of albumin dried by different treatments.

Table IV. Viscosity of Albumin Aqueous Solution.

Albumin Concentration %	Viscosity (Water=1, Temperature, 18.5°C.)				
	D	M_1	M_2	M_3	L
0.25	60	30	25	20	15
0.50	65	45	23	20	17.5
1.0	76	54	24.6	22.4	15.7
2.0	95	64.8	27.4	24.4	21.6
3.0	106	86	34.4	31.2	26.2
5.0	119	89.5	36.8	33.2	30
10	126	95	40.1	38	33.8
15	125	104	62	60.6	34.7
20	125	104	70.8	66	51.6
25	124	102	69.7	64	45.8
30	120	99.5	67.2	61.9	50.0
35	116	86.1	55.8	50.7	40.7
40	111	85.8	59.3	53.0	38.2
45	107	91	70.2	64.5	41.5
50	101	83.7	68.5	66	42.3

The relative viscosity of the sample dried in the dark is the highest; that of the sample dried in the sunlight is decreased so much that it is lower than any commercial albumin.

(V) The effect of monochromatic light on viscosity.

It was found from the measurement of viscosity that the effect of the sunlight

Table V. Variation of the Viscosity of Albumin Solution by Wave-length of Light.
10% Albumin aqueous solution.
Drying conditions of the samples: 0.45~0.37 atmospheric pressure.
Time of exposure: 2.2~3.8 hours.

Wave-length	Viscosity (Water=1)	Specific Gravity (18°C)
~ 390	29	1.0068
390 ~ 450	36.8	1.0085
450 ~ 530	41.5	1.0098
530 ~ 560	63	1.0150
560 ~ 600	118	1.0283
600 ~ 650	124	1.0295
650 ~ 800	135	1.0311
800 ~	128	1.0305

was remarkable. Therefore, using Wratten light filter for monochromatic light, the viscosity was measured for prepared different wave-length exposure.

As seen in the table, the wave-length of light affects the viscosity: the shorter the wave-length is, the more the viscosity falls. This shows that the monochromatic light in the drying process of albumin gives much effect.

(VI) The effects of light intensity and the wave-length of light on the change of viscosity with time.

If a reaction is in progress by light, viscosity is to be changed according to the reaction rate. Therefore, it is a matter of importance to measure the change of viscosity in the course of light exposure.

(1) The effect of light intensity.

The light intensity used was in such an order as this: sunlight> 1000^{W} bulb> 100^{W} bulb.

Table VI. The Change of Relative Viscosity of 10% Albumin Aqueous Solution by the Intensity of Light at 18.5°C.

Course of Measurement	Relative Viscosity of the Samples for Light Source		
	Dark		
1	126.0	126.0	126.0
2	129.0	129.0	129.0
3	132.1	132.1	132.1
4	135.1	135.1	135.1
5	138.0	138.0	138.0
	100 Volt Tungsten Filament Lamps		Sunlight
	100 Watts	1000 Watts	
6	128.7	126.5	115.0
7	125.0	118.3	90.0
8	120.0	109.4	65.0
9	100.5	100.0	45.0
10	89.4	92.1	30.0
11	85.0	85.7	19.0
12	81.0	80.7	17.6
13	81.0	80.7	17.6

As seen in the table, the viscosity strikingly decreased in the order of light intensity with the lapse of time.

(2) The effect of the wave-length of light with time.

The light source was the sunlight, using Wratten light filter for monochromatic light.

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Table VII. The Change of Relative Viscosity of 10% Albumin Aqueous Solution by the Wave-length of Light at 18.5°C.

Course of Measurements	Relative Viscosity of the Samples for Wave-length ($\mu\mu$) of Light							
	Dark							
1	126	126.3	126.3	126.3	126.0	126.3	126.0	126.1
2	129	129.4	129.4	129.4	129.0	129.4	129.0	129.0
3	132	133.0	133.0	133.0	132.0	133.0	132.0	132.0
4	135	135.5	135.5	135.5	135.0	135.5	135.1	135.1
5	138	138.5	138.5	138.5	138.0	138.5	138.0	138.0
	~390	390~450	450~530	530~560	560~600	600~650	650~800	800~
6	125	128.0	130.0	131.5	132.0	133.5	134.0	136.0
7	95.6	97.4	100.5	105.5	108.0	110.0	115.0	126.5
8	68.5	70.5	72.5	75.5	77.0	80.0	95.5	108.5
9	47.5	49.5	51.5	53.4	55.5	60.6	87.5	95.5
10	35.0	37.3	39.5	41.0	43.5	50.5	75.0	87.0
11	29.5	30.4	33.4	35.4	36.8	49.6	66.5	69.5
12	26.0	27.5	28.9	30.5	33.5	41.9	65.0	67.5
13	25.5	26.5	27.7	29.5	32.4	45.1	63.5	66.0
14	25.5	26.5	27.7	29.5	32.4	45.1	63.5	66.0

The table shows that the shorter the wave-length of light is, the more the viscosity is affected, i.e. the sooner the solution presents turbidity. In every case, the viscosity gradually decreased and reached its apparently stable state.

In conclusion, the author wishes to express his appreciations to coworkers of the laboratory.

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