

A deficiency of yeast or various B vitamins has been reported to produce histological changes in the liver of rats,^{4,5,14} rabbits,⁷ and guinea pigs.¹⁶ Rhoads and Miller¹⁷ found that there was a lowered capacity of the liver of dogs fed a black-tongue diet to excrete injected bilirubin. The amount of yeast in the diet is also related to the hepatic damage produced by lead arsenate,¹⁸ *p*-dimethylamino-azo-benzol¹⁹ and thyroid feeding.^{10,20} An increased food intake may be a factor in preventing liver damage in such cases.^{21,22}

Conclusions. Five dogs fed a vitamin

complex-free diet showed a gradual loss of appetite which culminated in anorexia. Four of these dogs developed an abnormal liver function, as judged by an increased retention of bromsulphalein. The hepatic damage appeared before, or slightly after, the time that anorexia appeared, and as the period of anorexia continued the dye retention returned to normal in 2 dogs. Histologically, the liver of these dogs showed mainly a mild to a marked chronic passive congestion.

¹⁴ Gyorgy, P., and Goldblatt, H., *J. Exp. Med.* 1942, **75**, 355.

¹⁵ Rich, A. R., and Hamilton, J. D., *Bull. Johns Hopkins Hosp.*, 1940, **66**, 185.

¹⁶ Spellberg, M. A., and Keeton, R. W., *Am. J. Med. Sc.*, 1940, **200**, 688.

¹⁷ Rhoads, C. P., and Miller, D. K., *J. Exp. Med.* 1938, **67**, 463.

¹⁸ Von Glahn, W. C., and Flinn, F. B., *Am. J. Path.*, 1939, **15**, 771.

¹⁹ Sugiura, K., and Rhoads, C. P., *Cancer Research*, 1941, **1**, 3.

²⁰ Drill, V. A., and Hays, H. W., *Am. J. Physiol.*, 1942, **136**, 762.

²¹ Drill, V. A., Shaffer, C. B., and Overman, R., *Am. J. Physiol.*, 1943, **138**, 370.

²² Post, J., Earle, D. P., Patek, A. J., and Victor, J., *Am. J. Path.*, 1942, **17**, 661.

1143

Ultracentrifugation Studies on the Blood of Normal and Jaundice-diseased Silk worms.

MAX A. LAUFFER. (Introduced by W. M. Stanley.)

From the Department of Animal and Plant Physiology of The Rockefeller Institute for Medical Research, Princeton, N.J.

Through the courtesy of Drs. R. W. Glase and W. M. Stanley, the author was afforded an opportunity to make a limited number of observations with the ultracentrifuge on the blood of normal and of jaundice-diseased silk worms and on an infectious material isolated by high-speed quantity centrifugation from the fluid of diseased worms. Because it is unlikely that it will be possible to extend this study in the near future, it was thought desirable to describe briefly the results obtained thus far.

A Bauer-Pickels type ultracentrifuge^{1,2} equipped with a Svensson-Philpot optical

system^{3,4} was used in this investigation. The results of sedimentation studies on 4 samples of pooled blood from healthy worms are presented in Table I (Samples I, II, III, and IV). It may be observed that 2 major components were found. As judged by the areas under the schlieren diagrams, the amount of material associated with the slower component was from one-half to one-third of the total.

Three of the samples of normal blood listed in Table I were treated by dilution with an 0.18 M sodium chloride-0.02 M sodium phosphate buffer solution at pH 7, by dialysis against an excess of the buffer, or by both. These treated materials, as well as a prepara-

¹ Bauer, J. H., and Pickels, E. G., *J. Exp. Med.*, 1937, **65**, 565.

² Pickels, E. G., *Rev. Sci. Instruments*, 1938, **9**, 358.

³ Svensson, H., *Kolloid-Z.*, 1939, **87**, 181.

⁴ Philpot, J. St. L., *Nature*, 1938, **141**, 283.

TABLE I.
Sedimentation Data on Undiluted Silkworm Blood.

Sample	Characterization	Sedimentation constants (Svedberg units)*		Temp., °C
		uncorrected		
I	Fresh normal blood	6.4	10.9	25°
II	" " "	6.0	10.8	25°
III	3 yr " "	6.6	13.6	27°
IV	" " "	6.4	11.8	26°
V	Fresh diseased blood	6.9	13.3	27°
VI	" " "	9.4	14.2	—
VII	" " "	9.5	15.3	—

* A Svedberg unit, symbolized by S, is a sedimentation rate of 10⁻¹³ cm per second per unit field.

tion obtained by Glaser and Stanley⁵ by centrifuging the fluid of normal silkworms for 2 hours at 60,000 R.P.M. in a Masket⁶ type rotor, were examined in the ultracentrifuge. Each was found to contain a fairly homogeneous component and a small amount of

TABLE II.
Sedimentation Data on Diluted and Dialyzed Normal Silkworm Blood.

Description of material		S_w^{20} Svedberg units
I of Table I	Dialyzed 24 hr	15.2
I " " "	Dialyzed 24 hr, then	
	diluted 1:4 with buffer	16.8
III " " "	Diluted 1:4 with buffer,	
	then dialyzed 24 hr	
	against buffer	16.5
IV " " "	Diluted 1:4 with buffer	16.4
Glaser and Stanley's material isolated from normal worms		15.5

more slowly sedimenting material which was too inhomogeneous to give the appearance of a definite boundary. The sedimentation constants corrected to water at 20°C, S_w^{20} , of the homogeneous component are listed in Table II. The average of the values listed there is about 16 S. This component is the same as the faster component of the undiluted and undialyzed blood. The values obtained in the untreated whole blood are lower because they were not corrected for the effects of temperature, solvent, and concentration. The inhomogeneous material is evidently composed of the same particles which make up the slower com-

ponent observed in undiluted and undialyzed silkworm bloods. The addition of buffer and dilution seem to favor the spreading of the boundary of the more slowly moving material. From all of these experiments it can be concluded that the blood from healthy silkworms contains a relatively homogeneous, very stable component with a sedimentation constant, S_w^{20} , of about 16 S and a variable amount of very inhomogeneous, more slowly sedimenting material. If the particles with S_w^{20} of 16 S are assumed to be spheres of density 1.35, a molecular weight of 325,000 and a particle diameter of 9.1 $m\mu$ may be calculated for them.

The blood from the diseased worms differed from that of the healthy worms in containing polyhedral bodies⁷ and a small amount of fatty material in suspension and in being infectious. The fatty material and the polyhedral bodies were separated from the blood by low-speed centrifugation, and the clarified blood was then examined in the ultracentrifuge. The results obtained with 3 samples of fresh pooled diseased blood clarified as described are presented in Table I (Samples V, VI, and VII). No difference between the clarified blood from diseased worms and the undiluted blood from healthy worms could be observed by this means.

Five different preparations of highly infectious material obtained by Glaser and Stanley⁵ by quantity high-speed centrifugation were also examined in the ultracentrifuge. The solvent was 0.10 M borate buffer at pH 7. In every case a single fairly homogeneous com-

⁵ Glaser, R. W., and Stanley, W. M., *J. Exp. Med.*, 1943, **77**, 451.

⁶ Masket, A. V., *Rev. Sci. Instruments*, 1941, **12**, 277.

⁷ Glaser, R. W., and Lacaillade, C. W., *Am. J. Hyg.*, 1934, **20**, 454.

ponent was observed. The sedimentation constants corrected to water at 20° are shown in Table III. The average of the 6 values

TABLE III.
Sedimentation Data on Infectious Material from Diseased Silkworms.

Sample	S_{w}^{20} Svedberg units	Sample	S_{w}^{20} Svedberg units
A	17.7	D	19.9
B	17.9	D*	16.8
C	14.4	E	16.4

* After 3 months' storage at 4°C.

reported in Table III is about 17 S. This figure is obviously not significantly different from that obtained for the homogeneous constituent of normal fluid. The material it represents must be, of course, the more rapidly sedimenting constituent of the undiluted blood of diseased worms.

The components found in this study to be present in the blood of both healthy and diseased worms are entirely different from the 2 components extracted by Glaser and Wyckoff⁸ from the frozen and ground bodies

⁸ Glaser, R. W., and Wyckoff, R. W. G., *Proc. Soc. Exp. Biol. and Med.*, 1937, **37**, 503.

of both healthy and jaundice-diseased silkworms. The blood components have corrected sedimentation constants of 16 or 17 S and about half of that value; the extracts of whole worms showed 2 components with corrected sedimentation constants of 53 and 92 S. The blood constituents are extremely stable, for they are apparently the same in fresh blood and in blood stored in the cold for 3 years; the components extracted from whole worms disintegrate within a few days.

Summary. The blood of normal and of jaundice-diseased silkworms and a highly infectious purified material isolated by Glaser and Stanley⁷ from the blood of diseased worms were studied in the ultracentrifuge. No difference between the bloods of healthy and diseased worms was observed by this method. Both contained a homogeneous component with a corrected sedimentation constant of 16 or 17 S and a very inhomogeneous, more slowly sedimenting component. The only component present in an optically detectable amount in the highly infectious purified material was found to have a corrected sedimentation constant of about 17 S.

14144

Inhibition of Bacterial Growth by Glucose in Media Devoid of Nicotinic Acid.

I. J. KLIGLER, N. GROSSOWICZ AND S. BERGNER.

From the Department of Hygiene and Bacteriology, Hebrew University, Jerusalem.

In preceding publications¹ it was shown that *Sal. paratyphi* A. and *Sh. dysenteriae* give appreciable growth in a synthetic or semi-synthetic medium in the absence of nicotinic acid, provided that no sugar or alcohol fermented by these organisms is present. The addition of glucose to such a substrate inhib-

ited growth, while the non-fermentable sucrose (or lactose) had no such unfavorable effect.

The present paper presents data showing that this inhibitive effect obtains with other bacteria requiring nicotinic acid for their utilization of carbohydrates and related substances. In these experiments we tested *B. proteus* XK, three strains of *Sh. dysenteriae* (Schmitz, Flexner and Hiss), and *Staphylococcus aureus*.

The proteus strain was grown in a medium consisting of the salt solution recommended by Fildes (without glucose or nicotinic acid),

¹ Fildes, P., *Brit. J. Exp. Path.*, 1938, **19**, 240; Kligler, I. J., and Grossowicz, N., *J. Bact.*, 1939, **38**, 309; Kligler, I. J., and Grossowicz, N., *Nature*, 1940, **146**, 652; Kligler, I. J., and Grossowicz, N., *J. Bacteria*, 1941, **42**, 173; Knight, B. C. J. G., *Biochem. J.*, 1937, **31**, 731.