

In preliminary chemical studies, fractionation by the procedures of Kirk<sup>4</sup> and Kirk, Page and Van Slyke<sup>5</sup> showed that about 20 per cent. of the material were soluble in petroleum ether. Of this, 5.7 per cent. were in the form of neutral fat, 10.2 per cent. were phospholipid and 5.1 per cent. cholesterol. The alcohol-ether insoluble fraction constituted about 65 per cent. of the material and contained phosphorus to the extent of 0.27 per cent. of the whole virus. If all this phosphorus were present in nucleic acid, the whole virus complex would contain about 3 per cent. nucleic acid.

The yield of the purified material in all experiments averaged about 2 mg per 100 cc of chorio-allantoic fluid. On the basis of nitrogen precipitable with trichloroacetic acid the degree of concentration for practical conditions was about 20 times and on the basis of volume it was 150 times. The specific 50 per cent. point infectivity for chick embryos varied from  $10^{-11.2}$  to  $10^{-12.1}$  grams per 0.1 cc inoculum with an average of  $10^{-11.4}$  grams corresponding to 6,600 particles. The quantity of virus giving the 2<sub>+</sub> red blood cell agglutination end point was  $10^{-6.5}$  grams.

The findings indicate that the influenza virus B is a relatively large particulate complex consisting of lipoprotein with which is associated nucleic acid of the desoxy-pentose type. From electron micrography, the average diameter of the spherical or ovoid bodies was 98 m $\mu$ . The diameter calculated from the sedimentation constant,  $S_{20}^0 = 832 \times 10^{-13}$ , the specific volume, 0.865, and an assumed spherical shape was 100 m $\mu$ . The influenza virus B (Lee strain) thus appears to be significantly larger than the influenza virus A (PR8 strain).

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<sup>5</sup> E. Kirk, I. H. Page and D. D. Van Slyke, *Jour. Biol. Chem.*, 106: 203, 1934.

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## THE ETHER SOLUBLE FRACTION OF NAVY BEANS AND THE DIGESTION OF STARCH

DURING the course of experiments dealing with the digestibility of dried navy beans it was observed that the oil of these beans retards the digestion of soluble starch by pancreatic amylase *in vitro*. The degree of this action appears to be of sufficient magnitude to warrant detailed study and comparison with other fats. It was also found that the retarding influence of the bean oil can be overcome to a large degree by a treatment with yeast. The object of this preliminary note is to briefly describe some of these findings.

Oil having these interfering properties can be readily obtained by extracting finely ground navy beans with ether for about one week. The total fat-soluble fraction when added to soluble starch in the same concentration in which it occurs in the beans retards the digestion of the starch more than some of the other edible fats. The amount of pancreatic amylase which completely digests 50 cc of a 1 per cent. solution of untreated soluble starch at pH 7 in less than thirty minutes *in vitro* leaves the treated starch incompletely digested after forty-eight hours.

Starch impregnated with the same amount of butter, lard or olive oil shows a negative starch-iodine test at about the same time as the untreated control. These findings are summarized in Table I. The oil which is

TABLE I

Fat added to starch in concentration of 1.5 per cent.	Starch-iodine test	
	½ hour digestion	48 hours digestion
bean oil	strongly positive	strongly positive
lard	negative	negative
butter	negative	negative
olive oil	negative	negative

quickly extracted from coarsely ground beans instead of allowing the usual period of one week for extraction does not retard the digestion. The difference in digestibility can not be attributed to the action of the ether on the oil.

The amount of starch which remains undigested in the presence of the bean oil varies with the age of the starch-oil preparation. In fresh preparations about half of it is undigested after two hours under the above conditions. As the age of the preparation increases the starch becomes less digestible until the inhibition is essentially complete in those preparations which have stood several months. A similar preparation of starch and olive oil becomes less digestible after standing for an equal period but is considerably more digestible than the starch impregnated with bean

oil. The relative difficulty with which the interfering factor in the bean oil is extracted from the beans and its increased effectiveness as the starch-oil preparations age suggests that the firmness of the association of the oil with the starch may account for its action.

Various means of overcoming this interference were tried. The method which appears to be most practical from a nutritional point of view involves a preliminary treatment with yeast. After incubating the undissolved starch-oil preparation with yeast and heating the preparation at 100° for 30 minutes the starch treated with bean oil is then digested by pancreatic amylase at about the same rate as untreated starch. This is true for both new and old starch-oil preparations. The yeast employed does not significantly digest the starch nor does the heated yeast hydrolyze maltose. Likewise, a similar treatment of whole navy

beans approximately doubles the rate of hydrolysis of the bean starch by pancreatic amylase. In this respect the treated beans compare more favorably with other common starchy foods. Heating the beans in dilute acid followed by neutralization further compensates for the delayed digestion. It is believed that a similar fat-soluble factor occurs in some other foods and preliminary studies indicate that other enzymes are influenced by this substance. It appears to be especially prominent in soybeans.

In view of some of the present nutritional problems and the increased use of soybeans the significance of this factor is being studied in detail.

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## SCIENTIFIC APPARATUS AND LABORATORY METHODS

### THE PROTECTIVE ACTION OF GLUCOSE IN BOVINE PLASMA AGAINST HEAT COAGULATION<sup>1</sup>

BEILINSSON<sup>2</sup> found that sucrose and glycerol inhibited the heat coagulation of egg albumin. Newton and Brown<sup>3</sup> showed that sucrose and glucose prevented coagulation of plant sap proteins by freezing. The protective action of other sugars against heat coagulation of egg albumin and the nature of the protection have been investigated.<sup>4</sup>

Van Der Scheer and co-workers<sup>5</sup> have studied the changes occurring in horse serum when it is heated to 60° to 65° C. for short intervals. They were able to show the presence of a new component by electrophoretic analysis. The new component was labeled "C." Evidence was presented to show that this component was a colloidal aggregation product resulting from the denaturation of constituents in the serum.

The present study demonstrates the protective action of glucose against the formation of the "C" component in heated bovine plasma by using the moving boundary technique of electrophoretic analysis.<sup>6</sup>

Normal bovine plasma obtained from citrated blood was utilized. One aliquot of the plasma was saturated

at room temperature with glucose, stoppered and then heated at 65° C. for one hour in a constant temperature water-bath. At the end of this period of heating, the sample was perfectly clear and no coagulation was evident. The glucose was removed by dialysis against physiological salt solution. A second aliquot was heated in its native form under the same conditions. However, in this case a coagulum formed during the heating period which was later removed by centrifugation. The remaining aliquot was not heated and

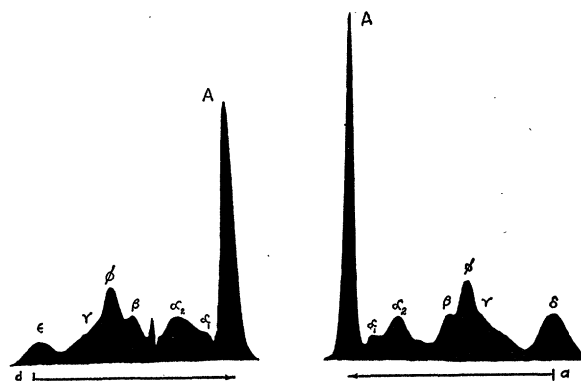


FIG. 1

served as a control. The three samples were then adjusted to a protein concentration of 2 per cent. and dialyzed for 3 days against large volumes of a diethyl barbituric acid—sodium diethyl barbiturate buffer of pH 8.6 and 0.1 ionic strength. Electrophoretic analyses of the samples were then carried out in a manner previously described at 0° C. for 10,000 seconds at a potential gradient of 6 volts.<sup>7</sup>

<sup>7</sup> C. L. San Clemente and I. F. Huddleson, *Mich. Agr. Exp. Sta. Bul.* 182, p. 3, 1943.

<sup>1</sup> From the Bacteriology Section of the Michigan Agricultural Experimental Station and the Kedzie Chemical Laboratory, East Lansing, Michigan. Journal article No. 652 from Michigan Agricultural Experiment Station.

<sup>2</sup> A. Beilinson, *Biochem. Zeits.*, 213: 399, 1929.

<sup>3</sup> R. Newton and W. R. Brown, *Can. Jour. Research*, 5: 87, 1931.

<sup>4</sup> C. D. Ball, C. R. Hardt, R. J. Westfall and W. J. Duddles. In process of publication.

<sup>5</sup> J. Van Der Scheer, R. W. Wyckoff and F. L. Clarke, *Jour. of Immunol.*, 40: 39, 1941.

<sup>6</sup> A. Tiselius, *Trans. Faraday Soc.*, 33: 524, 1937.