should submit the title and abstract of about 200 words to Farrington Daniels before June 21. The program of papers approved by the committee will be announced on June 21 at the symposium.

The symposium papers will be distributed as preprints before June 1. Together with the discussions they will be published in the *Journal of Chemical Physics*. An attempt will be made to record most of the discussions, but any one may withdraw his discussion from publication.

Chadbourne Hall, one of the university dormitories near the Chemistry Building, will be available for the symposium at rates of $1.50 per day. One floor will be reserved for families. In order to ensure accommodations, those intending to come should make reservations in advance.

Meals will be served in the cafeteria at the Memorial Union on the lake shore. Informal discussions will be continued at noon and in the evenings at the Memorial Union.

The Symposium Committee for the Division of Physical and Inorganic Chemistry is composed of E. J. Cohn, Farrington Daniels, H. Eyring, J. H. Hildebrand, L. S. Kassel, C. A. Kraus, V. K. LaMer, P. A. Leighton, S. C. Lind and G. Scatchard. The committee for the University of Wisconsin is composed of Farrington Daniels, J. O. Hirschfelder, W. E. Roseveare and J. E. Willard.

An informal dinner and launch ride are planned for the early evening on Tuesday and Wednesday.

HAROLD C. UREY,
Secretary

SPECIAL ARTICLES

THE SYNTHESIS OF A POLYSACCHARIDE FROM GLUCOSE-1-PHOSPHATE IN MUSCLE EXTRACT[1]

IT has been shown in previous papers[2, 3] that dialyzed extracts of muscle, heart, liver, brain and yeast contain a phosphorylating enzyme which catalyzes the reaction

glycogen + H_3PO_4 → glucose-1-phosphate ..... (1).

Glycogen is esterified with inorganic phosphate on carbon atom one of each glucose unit. The entire molecule is thereby disrupted into uniform fragments which consist of glucose-1-phosphate. Reaction (1) does not occur unless a small amount of adenylic acid (1 mM) is added to the dialyzed extracts.[4]

Another enzyme which is present in the extracts catalyzes the reaction

glucose-1- → glucose-6-phosphate ..... (2).

The migration of the phosphate group is greatly accelerated by Mg^{++} or Mn^{++} ions but takes place also without their addition, so that with time all the 1-ester formed is changed to 6-ester.[5] Attempts to reverse reaction (2) have not been successful, and it became obvious that for a further study of reaction (1) the phosphorylating enzyme had to be separated from the conversion enzyme.

Adsorption with aluminium hydroxide, followed by elution with disodium phosphate, yields an enzyme solution which is rich in phosphorylase and contains relatively little of the conversion enzyme. A second adsorption and elution results in an almost complete separation of the two enzymes. These elutions, after removal of the inorganic phosphate by dialysis, are suitable for a study of reaction (1).

When natural or synthetic 1-ester and 1 mM of adenylic acid are added to these dialyzed elutions, inorganic phosphate is liberated and a polysaccharide is formed. This substance has been isolated by a method similar to that used for the preparation of glycogen from liver or muscle. Without addition of adenylic acid the enzyme remains inactive, showing that adenylic acid is necessary for reaction (1) in both directions. Inosinic or adenosinetriphosphoric acid can not be substituted for adenylic acid. Galactose-1- and mannose-1-phosphate[6] are not transformed into a polysaccharide, and their phosphate group is not split off.

The polysaccharide formed by the muscle enzyme from added 1-ester is not destroyed by heating for one hour in 20 per cent. NaOH at 100°, is insoluble in 50 per cent. alcohol in the presence of electrolytes and does not show measurable reducing power with the alkaline copper reagent of Shaffer and Somogyi. The rate of hydrolysis in 0.2 N HCl at 100° is similar to that of glycogen, and the sugar formed is glucose. When the polysaccharide is added to muscle extract with inorganic phosphate and adenylic acid, it is converted back to the 1-ester. The polysaccharide differs from glycogen by the color it gives with iodine, which is blue. Under certain conditions, particularly after prolonged incubation, the formation of a polysaccharide which gives a purplish-brown color with iodine, can be demonstrated. It is not yet possible to give an explanation for these different color re-

[1] This work was aided by a research grant from the Rockefeller Foundation.

[2] Cori and Cori, *Proc. Soc. Exp. Biol. and Med.*, 36: 119, 1937; Cori, Colowick and Cori, *Jour. Biol. Chem.*, 121: 465, 1937.

[3] Cori, Colowick and Cori, *Jour. Biol. Chem.*, 123: 375, 1938.

[4] Cori, Colowick and Cori, *Jour. Biol. Chem.*, 123: 381, 1938.

[5] Cori, Colowick and Cori, *Jour. Biol. Chem.*, 124: 543, 1938.

[6] Colowick, *Jour. Biol. Chem.*, 124: 557, 1938.

actions with iodine. Determinations of the molecular weight of the synthetic polysaccharide may offer a clue.

Fig. 1 shows the general character of the reaction.

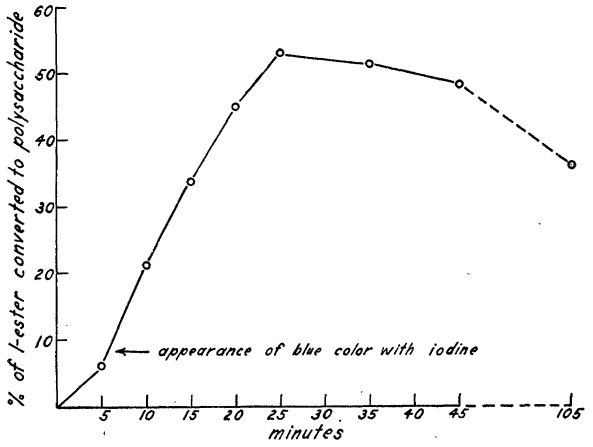

FIG. 1. The reaction mixture consisted of 2.5 cc of enzyme solution and 1.5 cc of additions; it contained 25 mM of synthetic glucose-1-phosphate and 1 mM of adenylic acid and was incubated at 30°. Aliquots of the reaction mixture were removed at the times indicated, deproteinized with trichloroacetic acid and analyzed for inorganic phosphate.

There is a definite lag, followed by a rapid attainment of an equilibrium. During the lag period the reaction mixture remains perfectly clear and the iodine reaction is negative. Often within one minute, the mixture becomes strongly opalescent, and the iodine reaction becomes positive. A liberation of inorganic phosphate occurs simultaneously. Both these points were demonstrated at a meeting of the Missouri branch of the Society for Experimental Biology and Medicine held on April 12, 1939. When the polysaccharide is determined according to Pflueger's method for glycogen, up to 91 per cent. of the calculated amount is found (calculated from the inorganic P liberated simultaneously). If some conversion enzyme is still present, as was the case in the experiment shown in Fig. 1, the reaction is gradually reversed, as shown by disappearance of inorganic phosphate between 25 and 105 minutes. This indicates that one is dealing with a reversible equilibrium.

In a recent note Kiessling[7] states that he has obtained a protein fraction from yeast juice by repeated fractionation with 0.3 saturated ammonium sulfate, which catalyzes reaction (1) in a reversible manner. This protein fraction contains the phosphorylase, the presence of which in yeast has been demonstrated before.[3] According to Kiessling the product formed from 1-ester by the yeast enzyme is a polysaccharide indistinguishable from glycogen. Kiessling's enzyme solutions remained active after prolonged dialysis against 0.3 saturated ammonium sulfate, which he interprets to mean that adenylic acid is not required. It is too early to say whether this indicates a difference between the phosphorylase in muscle and in yeast. An effect of addition of adenylic acid on the yeast phosphorylase could not be demonstrated in this laboratory.[3] It was then believed that the dialysis had not been effective in removing the nucleotide.

The interest of these findings lies in the fact that reaction (1) represents a reversible enzymatic equilibrium and that adenylic acid acts as coenzyme in both directions. Besides, the enzymatic synthesis of a high-molecular polysaccharide from a phosphorylated monosaccharide has been shown to occur *in vitro*. From the physiological point of view it seems important that glucose-1-phosphate has been shown to be the substrate for glycogen synthesis and that the same enzyme—the phosphorylase—brings about glycogen synthesis and glycogenolysis.

The difficulty experienced in obtaining glycogen synthesis in perfused organs or tissue slices is obviously due to the sensitiveness of the mechanism by which glucose is phosphorylated, a process about which nothing is known beyond the fact that oxidative energy is necessary. In yeast extract phosphorylation of glucose occurs anaerobically, linked with the oxidoreduction of cozymase. It is generally assumed that the phosphate is introduced in position 6, since only hexose-6-phosphate has been isolated. If reaction (2) should prove to be irreversible, this phosphorylation would not be the one required for glycogen synthesis.

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THE EFFECT OF ULTRA-VIOLET LIGHT ON BUILT-UP MULTILAYERS

A MULTILAYER of acid barium stearate deposited on chromium-plated steel as a built-up film to a thickness which shows interference colors changes in color after a short exposure to ultra-violet light. Optical measurements show a decrease in apparent thickness of the multilayer.

When glass having a refractive index of n = 1.5 is used as a base, a built-up acid barium stearate film is nearly invisible, since its refractive index is nearly equal to that of glass. Upon irradiation the multilayer becomes visible with bright interference colors having characteristics similar to those of a skeleton film.[1]

[7] Kiessling, *Naturwissenschaften*, 27: 129, 1939; see also Schäffner and Specht, *Naturwissenschaften*, 26: 494, 1938.

[1] K. B. Blodgett and I. Langmuir, *Phys. Rev.*, 51: 964, 1937.