

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.

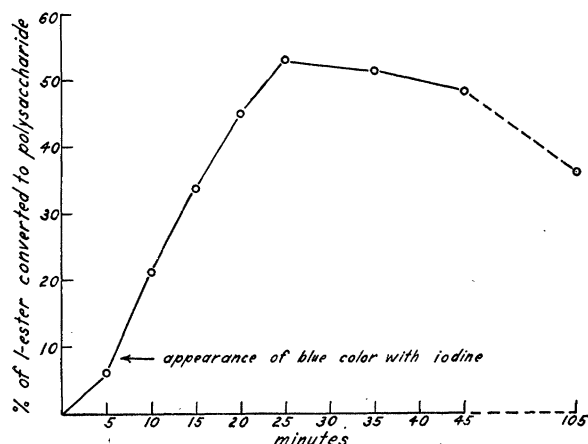


FIG. 1. The reaction mixture consisted of 2.5 cc of enzyme solution and 1.5 cc of additions; it contained 25 mM of synthetic glucose-1-phosphate and 1 mM of adenylic acid and was incubated at 30°. Aliquots of the reaction mixture were removed at the times indicated, deproteinized with trichloroacetic 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 Kiessling⁷ 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

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

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

A film made up of 48 layers, each containing equal parts of stearic acid and barium stearate (referred to as acid barium stearate), placed 25 cm from a quartz high-pressure Hg Uviarc lamp² operating at 300 watts, lost optical thickness equivalent to one molecular layer after 5 minutes' irradiation. Without further exposure the film continued to lose optical thickness for several hours, the final loss amounting to more than two layers. A multilayer of pure stearic acid undergoes a similar change in apparent thickness, although it is not skeletonized. Pure barium stearate shows only one twentieth of this loss under the same conditions of irradiation.

The decomposition products formed evaporate completely within ten minutes following five minutes' irradiation if the exposure is made on a film sealed within a highly evacuated quartz tube. In air or an atmosphere of Ar, N₂ or O₂ the change in thickness following irradiation continues for several hours.

By irradiating a film held at 0° C. the photochemical effect can be separated from the subsequent evaporation of the volatile constituent. When this is done the film shows no loss in thickness at the end of 20 minutes of irradiation. With no further exposure it starts to lose thickness as soon as its temperature is raised, and evaporation continues for several hours.

The photochemical effect is produced by a definite region of short-wave radiation and is related to such factors as exposure time, temperature of the film, distance from the source and intensity of the light.

Ozone seems to have little if any effect on the decomposition of the multilayers. A continuous stream of it directed for an hour against a step film produced a slight increase in optical thickness.

The radiations from a Hg arc lamp responsible for most of the photochemical decomposition were of wavelengths between 2,300 Å and 2,700 Å. This was determined by placing a multilayer at the focal plane of a concave Wood's grating. The light after passing through a narrow slit and falling on the grating 60 cm away was focused as discrete lines on the multilayer. The effective radiation produced visible effects on the multilayer, which after measurement and comparison with the calibrated output of a similar lamp² directly established the wave-length region responsible for most of the photochemical decomposition. Twenty-three lines (ranging from 1,949 Å to 3,983 Å) could be seen on the film following six hours of irradiation.

Light of shorter wave-lengths also decomposes acid barium stearate multilayers. A plate bearing a multilayer was sealed in a hot cathode, low-pressure discharge tube and exposed in successive runs to the radiation from Xe, Kr, Ar, Ne and He. The optical

loss following each exposure was of the same magnitude. The discharge in 2,000 microns of helium decreased the optical thickness by 16 per cent. in 2 minutes of irradiation. The helium discharge gives a resonant line at 584 Å.

The rate of evaporation of the volatile products formed by irradiation was compared to that of pure straight chain hydrocarbons. Decane, tetradecane and hexadecane were placed successively in a step film of barium stearate which had been skeletonized and the decrease in optical thickness measured as the hydrocarbons evaporated. The evaporation rate of the decomposition product when compared to that of the hydrocarbons showed a rate such as would be expected for heptadecane. This indicates that ultra-violet irradiation splits the molecules of stearic acid at the carboxyl group.

Ultra-violet light passing through a sheet of clear fused quartz coated with 700 layers of acid stearate produces a decrease in thickness which is less than when the light passes through uncoated quartz. This difference measures the absorption of the effective radiation by the acid stearate film. A comparison shows that the absorption due to the 700-layer film reduces the effective radiation by 13 per cent. This indicates an absorption coefficient for the film of 2×10^{-4} per layer.

A similar value is obtained by passing part of the light through a film and observing the reduction in brightness of lines on a fluorescent screen representing radiations from the Hg lamp in the 2,500 Å region. The light was separated into discrete lines by the Wood's grating and focused on a fluorescent screen at the focal plane. A 1,000-layer film showed an absorption per layer of 8×10^{-4} in the region of 1,800 Å which decreased to about 5×10^{-4} at 1,970 Å and 2×10^{-4} at 2,537 Å. At wave-lengths longer than 2,652 Å hardly any absorption was observed.

Irradiation by the Hg lamp of an acid stearate multilayer immersed in distilled water showed a loss after withdrawal of the same order as when irradiated in air. Very little of the decomposition product diffused into the water except after irradiation for ten minutes or more. With films made up of the shorter chain fatty acids, however, containing 14, 15, 16 or 17 carbon atoms the film after a five-minute exposure at 25 cm is partially skeletonized before removal from the water and is found to be strongly hydrophobic when withdrawn.

A paper describing further details of this work will be published in the near future.

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² W. E. Forsythe, B. T. Barnes and M. A. Easley, *Jour. Optical Society of America*, 24: 7, 1934.