consisted in a reduction of the enzyme; the inactive enzymes were supposed to contain disulfide groups which were reduced by the activators to sulfhydryl groups.[3]

Recently it has been possible to synthesize substances of low molecular weight which are split by the following catheptic enzymes: cathepsin, papain, and bromelin of the pineapple. Reports on the substrates of papain have already been published.[4] The present authors have found that cathepsin from pig liver splits the following substrates: Carbobenzoxy glycyl-*l*-glutamyl glycine amide, Carbobenzoxy leucyl glycyl glycine, Carbobenzoxy diglycyl glycine and Carbobenzoxy triglycyl-*l*-leucyl glycine. It was further found that bromelin splits carbobenzoxy glycyl-*l*-glutamyl glycine amide and carbobenzoxy triglycyl-*l*-leucyl glycine.

It is thus possible to study the activation and specific action of each catheptic enzyme with the aid of synthetic substrates of known structure. The possible detailed investigation of cathepsin is of some significance in human physiology in view of its rôle in intracellular protein metabolism.

The application of the synthetic substrates has already yielded the result that cathepsin, papain and bromelin each consists of two partial enzymes with quite different specificities and different behaviors toward phenylhydrazine. One of the enzyme components of cathepsin, papain and bromelin is inactivated by phenylhydrazine, the other enzyme component is not. It is remarkable that the three hitherto investigated catheptic enzymes, the origins of which are so different, show the same activation phenomena and reveal themselves to be dual enzyme systems with analogous properties of the partial components. It seems probable therefore that the dual enzyme system is necessary for the physiological task of this group of enzymes, and that it is related to the mechanism of activation.

This expectation finds support in the following observations: When, in a non-activated preparation of cathepsin, one of its partial components is combined with phenylhydrazine, simultaneously the other partial component of the enzyme is decisively activated. The experiments with papain[5] and bromelin show quite analogous results. These and other experiments speak in favor of an interdependence of the two partial enzymes of each catheptic enzyme and prove that activation and inactivation can be performed without resorting to oxidation-reduction processes.

At present the following hypothesis seems therefore to be the most probable interpretation of the dual enzyme system of catheptic enzymes: Catheptic enzymes are systems of two enzyme components which are in combination with each other and thus mutually inactivate each other. The process of activation thus involves a dissociation of the inactive dual enzymes into its two component enzymes having different specificities. The known natural activators which effect the dissociation and thus the activation of catheptic enzymes are oxidizable and lose through oxidation their ability to effect the dissociation. In metabolism the activity of catheptic enzymes may thus be influenced by the oxidation-reduction potential of the milieu through the medium of oxidizable activators.

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CONCENTRATION AND PARTIAL PURIFICATION OF BACTERIOPHAGE

A PROTEIN preparation which possesses the properties of bacteriophage has been isolated from lysed staphylococcus cultures. 1×10^{-10} mg of this preparation will cause lysis when added to growing staphylococci cultures. During the reaction more of the bacteriophage protein is formed. The loss in activity of a solution of the protein is proportional to the denaturation of the protein at various temperatures and at different pH. The protein is adsorbed by susceptible living or dead bacteria to the same extent as is the bacteriophage activity. Neither the protein nor the bacteriophage activity is adsorbed by resistant bacteria. The rate of diffusion of the protein is the same as that of the active agent and the diffusion coefficient is 0.02 cm²/day, corresponding to a molecular weight of about 500,000.

The protein is not digested nor is the activity decreased by trypsin or papain. Chymo-trypsin inactivates the preparation and this inactivation is accompanied by the formation of protein insoluble in one quarter saturated ammonium sulfate. There is no detectable hydrolysis of protein during this reaction.

The ultra-violet absorption spectrum agrees with that calculated from Gates' inactivation experiments with ultra-violet light and bacteriophage.

The method of preparation is briefly as follows: Culture media used was prepared by extracting 2½ kilo dried yeast with 200 liters boiling water. The media was adjusted to pH 7.6 and 37° C. and inoculated with staphylococci and a small amount of bacteriophage. Sterile air was

bubbled through the solution until growth and subsequent lysis was complete. After lysis the solution was adjusted to pH 9.0, dilute lead sub-acetate added and the supernatant solution from the lead acetate precipitate concentrated in vacuo to one tenth its original volume, digested with crystalline trypsin and the active protein isolated by fractional precipitation with ammonium sulfate between 0.2 and 0.4 saturation at pH 7.0. About 40 mg of protein, representing about 30 per cent. of the original total activity, may be obtained in this way from 200 liters of lysed culture.

The protein preparation obtained in this way is brownish and forms highly viscous, slimy solutions. The brown color may be removed by repeated precipitation with ammonium sulfate, but the protein is always mucin-like in character. It contains about 1 per cent. reducing sugar and small amounts of glucose amine.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

SIMPLE SYNCHRONOUS MOTOR FOR THE HARVARD KYMOGRAPH

THE synchronous motor described below was devised to operate a small Harvard kymograph with two drums, the original spring drive being found to produce a very inconstant drum speed. It is simple to construct, inexpensive and durable. Its speed is constant when the frequency of the alternating-current supply is accurately controlled (*i.e.*, when the current is suitable for the operation of the ordinary variety of electric clock), and is independent of variations of the line voltage or of the load applied. It requires manual starting, will run in either direction and uses no belts or external gears.

The apparatus consists essentially of a toothed soft-iron disk rotating between similarly toothed pole-pieces which are energized by alternating current. The speed depends on the number of teeth in the disk and the frequency of the supply current, according to the following formula:

$$\text{Speed (r.p.m.)} = \frac{120 \times \text{frequency (cycles/sec.)}}{\text{number of teeth}}$$

It was found that the power and stability of the motor was much improved by the addition of a small flywheel, which fits loosely on the rotor spindle and is attached to the latter by a coil spring.

A satisfactory design is shown in Fig. 1. Considerable deviation from this design is possible, providing the principles mentioned are retained. The 24-toothed rotor shown rotates at 300 r.p.m. and results in a drum speed of approximately 2.8 cm. per minute at the low speed setting of the kymograph. This rotor has a tapered hole in its shaft, and replaces the fans ordinarily used to control the kymograph speed. The pole-pieces are attached to the top plate of the kymograph by machine screws, and the field coil projects over the edge of the kymograph. This coil consists of approximately 2,000 turns of No. 26 enameled copper wire, and the cross-section of the core is $\frac{3}{8}$ inch by 1 inch inside the coil. The flywheel is attached to the

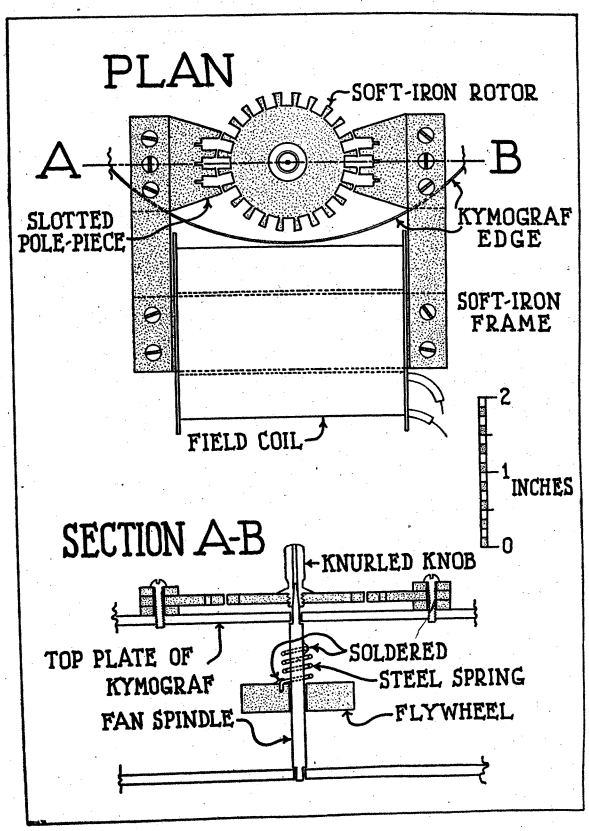

FIG. 1

rotor spindle by its spring inside the kymograph base. The winding lever and ratchets on the spring drum and brake lever on the fan spindle are removed. Care must be taken that the spring is entirely run down when the kymograph is taken apart, else the gears may be injured by the suddenly released spring.

The design might be improved by the use of silicon-steel laminations for the core of the field coil, and it is possible that the coil and core of a cheap electric clock would be satisfactory. The coil will require more turns for operation on 25-cycle current than described for 60-cycle current. If a considerably