

At the end of the pepsin period of 96 hours there was an average of 17% amino nitrogen but no tryptophane liberated. During the trypsin period the liberation of tryptophane and the establishment of equilibrium was the same as in the experiment with trypsin alone without any pepsin action. Erepsin action for 48 hours showed an additional liberation of 15-18% of amino nitrogen but no change in the tryptophane concentration. Thus, trypsin or a trypsin type of enzymes alone are involved in the liberation of tryptophane from the proteins studied.

The effects of sodium and chloride ions on the precipitation of tryptophane by mercuric sulfate were studied. A 0.3% concentration of chloride ion interferes with and a 0.77% entirely prevents the precipitation of tryptophane under the conditions as given in the indirect Vanillin-HCl reaction. Sodium in concentrations up to 2% has no effect.

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<sup>1</sup> Kraus, Ida, *J. Biol Chem.*, 1925, lxiii, 157.

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#### Demonstration of Rapid Pepsin-Hydrochloric Proteolysis in Vitro.

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Hydrolysis of the more complex protein molecule into simpler forms carried on with the aid of proteolytic enzymes ordinarily requires considerable time even at body temperature. Some time ago it was observed that the addition of a very small amount of solid pepsin to fibrin jelly causes an almost immediate solution of the fibrin at room temperature. The jelly was produced by treating 20 gm. of washed fibrin with 250 cc. of .04% hydrochloric acid. If the jelly is stirred or shaken after the addition of the pepsin, 5 minutes usually suffices to put the fibrin into solution. An observation on this point is recorded in literature but no definite data is given as to condition of the experiment, the speed of the solution or the nature of the end products.

Recently the nature of the soluble protein products has been studied. Dr. Hektoen determined by means of an anti-fibrinogen serum that the soluble protein, resulting from this treatment, was immunologically different from fibrinogen. This result definitely

eliminated the idea that this reaction might be a reversal of the one which causes conversion of fibrinogen into fibrin. A further study on the soluble products resulting from this treatment revealed the fact that they are largely proteose in character and that the larger percentage of the compounds falls between the limits of 50 to 100 saturation with ammonium sulphate. We have then, here, a definite demonstration of extremely rapid proteolysis at room temperature.

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### Some Effects of Histamine on the Acid-Base Balance.

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The "alkaline tide" in the urine during the digestive period is commonly regarded as a compensatory process, eliminating the excess of base left in the blood after the formation of HCl in the stomach. Some of the observed variations in urine reaction, however, may be indirectly dependent on the absorption of food. So far as we are aware the alkaline tide has not been shown to accompany gastric secretion in the fasting subject. Ackman<sup>1</sup> studied the urine reaction in man, giving a test meal followed immediately by the administration of histamine. The food factor was therefore not eliminated. It seems that the use of histamine alone might give more valuable information as to the cause of the tide.

Female dogs, weighing from 9.0 to 12.5 kg., and having fasted for 16 to 20 hours, were used in our experiments. Measurements of pH were made on urine by the potentiometer, and on blood plasma by the method of Hastings and Sendroy.<sup>2</sup> In the studies on urine, each animal was as a rule used for a single experiment of 4 to 6 hours, in order to avoid the effects of a possible cystitis from the use of the catheter. Histamine dichloride was injected subcutaneously, the usual dose corresponding to 0.7 mg. of histamine base.

Such a dose of histamine invariably produces a rise in urinary pH, lasting for about 2 hours, after which the reaction returns to near the original level. The duration of this effect suggests that it is related to the gastric secretory activity. The following table is a typical record from a Pavlov pouch dog, showing the gastric and urinary effects.