

citrate on the rate of disintegration of virus in 6 M urea at about pH 7.5 was determined by separating the high from the low molecular weight material by high-speed centrifugation after different periods of time. Disintegration proceeded most rapidly in 0.1 M phosphate buffer, slightly less rapidly in 0.05 M phosphate, 0.05 M borate and 0.02 and 0.04 M citrate, and less rapidly in 0.01 and 0.3 M phosphate and 0.004 and 0.12 M citrate. The rate was noticeably slower in 0.024, 0.12, 0.24 and 0.71 M sodium chloride and in 0.24 M citrate, and quite slow in 0.6 M phosphate, in 1.4 M sodium chloride and in the absence of electrolytes. The half-life periods of the native protein in the various urea-electrolyte solutions cover the range from less than 2 hours to more than a week. The effect of 4 different hydrogen ion concentrations on the rate of disintegration in 6 M urea and 0.1 M phosphate buffer was also studied. The rate was greatest at pH 8.2, slower at pH 7.4, much slower at pH 6.4 and almost negligible at pH 5.5. The disintegration of virus protein in 6 M urea and 0.1 M phosphate buffer at pH 7 is similar to the urea denaturation of egg albumin<sup>9</sup> in proceeding more rapidly at 0° than at 25° C., but differs from it by proceeding more rapidly at 40° than at 25° C. The rates of reaction during the degradation of about 90 per cent. of the high molecular weight material at 25° or 40° C. in 6 M urea and dilute buffers at about pH 7 may be described fairly satisfactorily by the equation of a first order reaction. However, the amount of high molecular weight active material remaining after about 50 hours, though quite small, may be as much as 10<sup>6</sup> times that predicted by the first order equation.

It may be concluded that tobacco mosaic virus is rapidly disintegrated in 6 M urea and 0.1 M phosphate buffer at pH 7, with appearance of free sulfhydryl groups, into low molecular weight protein components which contain no nucleic acid, exhibit no double refraction of flow, are insoluble in dilute buffers, and, most important, possess no virus activity. The rate of degradation varies widely with the concentration of urea, the concentration of electrolyte, the type of the electrolyte, the hydrogen ion concentration and the temperature. These results, especially the demonstration of the great effect of small changes in pH or in electrolyte concentration on the rate of disintegration, as well as the earlier work on the degradation of virus in solutions of sodium dodecyl sulfate,<sup>10</sup> may provide information concerning the nature of the forces which hold together the large virus molecule. The conclusion of Frampton and Saum<sup>11</sup> that virus activity is associated with the low molecular weight products obtained

from tobacco mosaic virus upon solution in concentrated urea has not been confirmed.

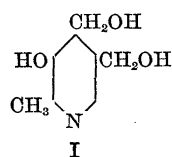
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### SYNTHETIC VITAMIN B<sub>6</sub>

THE isolation of vitamin B<sub>6</sub> was reported by Keresztesy and Stevens,<sup>1, 2</sup> of this laboratory, and also by other workers.<sup>3-6</sup>

The structure of vitamin B<sub>6</sub> was studied at this laboratory, and the evidence which led to the structure is described fully in two papers<sup>7, 8</sup> which are about to appear. The structure of vitamin B<sub>6</sub> was shown to be 2-methyl-3-hydroxy-4,5-di-(hydroxymethyl)-pyridine,



I. Kuhn and coworkers<sup>9-11</sup> have announced the results of researches which led to the same structure for the vitamin.

In this laboratory, the complete synthesis of the vitamin B<sub>6</sub> has been accomplished, and we wish to describe the results of the comparison of the synthetic vitamin B<sub>6</sub> hydrochloride with the natural vitamin B<sub>6</sub> hydrochloride.<sup>1</sup>

M.p. 206-208° C., mixed melting point with natural vitamin B<sub>6</sub> hydrochloride, 207° C. Positive ferric chloride test.

Anal. Calc. for C<sub>8</sub>H<sub>12</sub>O<sub>3</sub>NCl: C, 46.72; H, 5.84; N, 6.81. Found: C, 46.55, 46.64; H, 5.57, 5.69; N, 6.83, 6.75. The biological assay also confirms these findings.

A complete report of this work will be published in the near future.

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<sup>1</sup> Keresztesy and Stevens, *Proc. Exp. Bio. Med.*, 38: 64, 1938.

<sup>2</sup> Keresztesy and Stevens, *Jour. Amer. Chem. Soc.*, 60: 1267, 1938.

<sup>3</sup> Lepkovsky, *SCIENCE*, 87: 169, 1938; *Jour. Biol. Chem.*, 124: 125, 1938.

<sup>4</sup> Kuhn and Wendt, *Ber.*, 71: 780, 1118, 1938.

<sup>5</sup> Ichiba and Michi, *Sc. Papers Inst. Phys. Chem. Research*, 34: 623, 1014, 1938.

<sup>6</sup> Gyorgy, *Jour. Amer. Chem. Soc.*, 60: 983, 1938.

<sup>7</sup> Stiller, Keresztesy and Stevens, *ibid.*, 61, May, 1939.

<sup>8</sup> Harris, Stiller and Folkers, *ibid.*, 61, May, 1939.

<sup>9</sup> Kuhn and Wendt, *Ber.*, 72: 305, 1939.

<sup>10</sup> Kuhn, Andersag, Westphal and Wendt, *ibid.*, 72: 309, 1939.

<sup>11</sup> Kuhn, Wendt and Westphal, *ibid.*, 72: 310, 1939.

<sup>9</sup> F. G. Hopkins, *loc. cit.*

<sup>10</sup> M. Sreenivasaya and N. W. Pirie, *Biochem. Jour.*, 32: 1707, 1938.

<sup>11</sup> *Loc. cit.*