## Letters to the Editor

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## Ultra-Violet Absorption Spectrum and Chemical Structure of Vitamin B,

WE have been particularly interested in the report in NATURE<sup>1</sup> by Bowden and Snow regarding the photochemistry of the vitamins, in view of our investigations of the absorption spectra of various vitamin B<sub>1</sub> concentrates, a preliminary report of which was presented at the meeting of the American Chemical Society at Buffalo, September, 1931.<sup>2</sup> At that time we noted that the concentrates examined had absorption maxima at 2600 A., but as the concentrates were admittedly impure, and as some also presented maxima at other wave-lengths, we hesitated to identify any band with vitamin B<sub>1</sub>.

In view of the possible presence of contaminating substances in the Jansen-Donath preparation used by Bowden and Snow, we agree with Morton and Heilbron<sup>3</sup> that their definite conclusion, "The correlation of the 2600 band with the activity of vitamin B<sub>1</sub> has thus been fully established" seems scarcely to have been justified by their reported experiment, in which irradiation of the preparation with the 2560 A. line reduced the intensity of the 2600 A. absorption band and destroyed the B1 activity. Particularly is this true because, as we pointed out in our previous paper, certain biologically inactive purines and pyrimidines (which we have shown to be characterised by an absorption maximum at 2600 A., and to be destroyed by certain regions of the ultra-violet<sup>4</sup>), might be expected to contaminate B<sub>1</sub> concentrates. In view of this difficulty, we believed it advisable, at the time of our earlier report, to carry out parallel spectrographic and biological studies on a number of  $B_1$  preparations before making positive statements about the absorption of the vitamin.

During the course of our later investigations, and prior to the publication of Bowden and Snow's report, Windaus and his co-workers<sup>5</sup> announced a crystalline  $B_1$  preparation, believed by them to be pure  $B_1$ . which had, indeed, a pronounced absorption maximum at 2600 A.

Our later experiments include the comparison of the biological activity and ultra-violet absorption spectra of four B<sub>1</sub> concentrates prepared in this Laboratory, of three kindly supplied us by other workers, and of three reported by Guhas and by Windauss. Full details of the spectrographic technique and biological assays will appear shortly in the Bulletin of Basic Science Research. 'The absorption curves of all these are presented in Fig. 1. The biological activity of three of these preparations (H4, and the two Guha preparations) have not been determined by methods permitting comparison of their activities with those of the others.

In agreement with our previous suggestions, it was found that the two relatively inactive concentrates (H1 and Cerecedo) had very high absorption in the 2600 A. region, presumably because of the presence of inactive purines or pyrimidines. When these were eliminated, a good degree of correlation was found between the activity and the absorption at

No. 3290, Vol. 130]

2600 A., better than that at any other wave-length in the ultra-violet spectrum. If, for example, the activity and the extinction at 2600 A. of the Windaus preparation are each taken as 100, these values are respectively, for the other four concentrates : Heyroth II, 26.7, 29.8; Seidell (30:85), 24.0, 32.8; Heyroth III, 6.7, 7.0; Seidell (30:182), 4.8, 14.4. This. together with the fact that a maximum at or near 2600 A. was found in all of the concentrates, indicated that vitamin B<sub>1</sub> is characterised by a 2600 A. band.

The type of absorption thus attributed to vitamin B<sub>1</sub> resembles most closely, of the nitrogenous heterocyclic compounds which have thus far been investigated, the absorption of pyrimidine-ring-containing compounds, or of compounds of the type of ergo-The absorption curve of the sulphurthioneine. containing Windaus preparation, the most active of



those considered, is in fact very similar to that of uracil<sup>4</sup> or other pyrimidines. The molecular extinction coefficient of the Windaus preparation at 2600 A. as calculated from the ompirical formula of Windaus is 8225, and as calculated from the formula of van Veen<sup>7</sup> is 8925. That of uracil at the same wavelength is 9500.

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- <sup>1</sup> NATURE, 129, 720; 1932.
  <sup>2</sup> Heyroth and Loofbourow, Bull. Bas. Sci. Res., 3, 237; 1931.
  <sup>4</sup> NATURE, 129, 866; 1932.
  <sup>4</sup> Heyroth and Loofbourow, J. Amer. Chem. Soc., 53, 3441; 1931.
  <sup>4</sup> Windaus, Tschesche, Ruhkopf, Laquer, and Schultz, Z. physiol. Chem., 204, 123; 1932.
  <sup>5</sup> Guha, Biochem. J., 25, 941; 1931.
  <sup>7</sup> van Veen, Rec. trav. Chim., 50, 200, 208, 610; 1931.