

# SCIENCE

VOL. 100

FRIDAY, DECEMBER 22, 1944

No. 2608

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SCIENCE: A Weekly Journal devoted to the Advancement of Science. Editorial communications should be sent to the editors of SCIENCE, Lancaster, Pa. Published every Friday by

## THE SCIENCE PRESS

Lancaster, Pennsylvania

Annual Subscription, \$6.00

Single Copies, 15 Cts.

SCIENCE is the official organ of the American Association for the Advancement of Science. Information regarding membership in the Association may be secured from the office of the permanent secretary in the Smithsonian Institution Building, Washington 25, D. C.

## VITAMINS IN OUR FOOD<sup>1</sup>

By Professor A. E. MURNEEK

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My appearance on this program, I wish to assure you, is not of my choice. I am the object of a well-established punishment to those who have been "honored" due to previous servitude and an annual or perennial display of the older folks for the amusement or encouragement of the younger generation.

The particular subject, however, is of my selection in order to fit into the general program of "Nutrition—Some Current Views." Vitamins must have gained an alarming popularity, when a non-specialist, like myself, desires to discuss them before a group of assorted specialists. Plants and vitamins, however, are so closely linked that, as a horticulturist and plant physiologist, I, too, have been obliged to deal with

them more intimately—the "little things" in nutrition that now count so much. If in this discussion I may be unduly critical, kindly forgive me by ascribing it to no greater fault than that due to emotional simplification, personal prejudice or perchance conservatism come with age. In agreement with Thorstein Veblen, I do not wish to criticize but merely to understand. If disparagement is involved in this quest for certainty, I hope for your graciousness.

### DEMONSTRATION AND DISTRIBUTION OF VITAMINS

According to an older definition, a vitamin is a catalytic substance indispensable in animal or human nutrition. It can not be synthesized in their organisms but must be obtained from plants. Now we know that all plants, likewise, require certain vitamins and that many of them are heterotrophic, must obtain their vitamins from an external source. This certainly is

<sup>1</sup> Address of the retiring vice-president and chairman of Section O (Agriculture), read before the joint meeting of Sections O and G (Botany) of the American Association for the Advancement of Science, Cleveland, Ohio, September 14, 1944.

test whether the dosage-response curves for standard and unknown are parallel. Computing  $y_3$  in two ways checks the correctness of the original differences. The computing units are then summed for each column to obtain  $T_1$ ,  $T_2$  and  $T_3$ , respectively.

The potency of the unknown is computed in logarithmic units. If the unknown is assumed to have the same potency as the standard, values such as those in the right hand and lower portion of Table 1 lead directly to the logarithm of relative potency,  $M'$ , by the formula

$$M' = \frac{IT_1}{T_2} = \frac{0.5 \times 3.8}{39.4} = 0.0482 \quad (1)$$

where  $I$  is the dosage interval in logarithms and the numerical values are those obtained from Table 1. To express the results in Oxford units, the unitage assumed in carrying out the assay is used. The estimated log-potency ( $M$ ) is given by the equation

$$M = \log (\text{assumed unitage or potency}) + M' \\ = 2.6021 + 0.0482 = 2.6503. \quad (2)$$

From the antilog of  $M$  the potency of the unknown has been assayed at 447.0 U/mg.

The above calculation differs from a similar proposal<sup>3</sup> in leading directly to an easy estimate of the standard error,  $s_M$ . While some factors in the determination of potency are not tested in a single assay—and these may be of first importance—the precision of a given technique is no greater than that indicated by the standard error. The latter depends upon several intermediate terms. The first of these, the slope of the dosage-response curve, is computed as

$$b = \frac{T_2}{2IN}, \quad (3)$$

where  $N$  is the number of sets or plates in the assay. With  $I = 0.500$ , this may be simplified to

$$b = \frac{T_2}{N} = \frac{39.4}{4} = 9.85. \quad (3a)$$

The standard deviation of a single response as computed from all relevant data is

$$s = \sqrt{\frac{S(y^2) - (T_1^2 + T_2^2 + T_3^2)/N}{12(N-1)}} \\ = \sqrt{\frac{392.20 - 391.71}{36}} = 0.1167, \quad (4)$$

where  $y$  refers to all individual values of  $y_1$ ,  $y_2$  and  $y_3$  and  $S(y^2)$  is the sum of their squares. The standard deviation in the response is divided by the slope to transform it from units of response ( $y$ ) to units of log-dose ( $x$ ), giving

$$\lambda = \frac{s}{b} = \frac{0.1167}{9.85} = 0.01185. \quad (5)$$

The smaller the term  $\lambda$  the more sensitive is the assay for detecting differences in potency. The standard error of  $M$  or of  $M'$  is then equal to

<sup>3</sup> M. B. Sherwood, E. A. Falco and E. J. de Beer, *SCIENCE*, 99: 247, 1944.

$$s_M = \lambda \sqrt{\frac{1}{N} \left\{ 1 + \frac{T_1^2}{T_2^2} \right\}} = 0.01185 \sqrt{\frac{1.0093}{4}} = 0.0060. \quad (6)$$

To realize the full precision of a given method, the assumed potency should not differ widely from the true value. Changes in technique which increase the inherent precision of an assay will reduce  $\lambda$ . With a standardized procedure,  $\lambda$  should be relatively stable from one assay to another.

Most tests of statistical significance are made in units of  $M \pm s_M$ . In original units an average standard error may be computed as

$$\text{standard error of potency} = 2.30s_M (\text{antilog } M). \quad (7)$$

In the present case the potency of the unknown would be reported as 447.0  $\pm$  6.2 U/mg. This agreed well with a parallel assay of the same preparation, which gave a potency of 451.7  $\pm$  5.6 U/mg.

The above procedure assumes that the dosage-response curves for standard and unknown are parallel. This assumption may be tested by the t-test for the significance of a difference where

$$t = \frac{T_3}{2s\sqrt{N}} = \frac{0.2}{2 \times 0.1167 \times 2} = 0.43 \quad (8)$$

The observed value is referred to a suitable table<sup>4</sup> with 3(N-1) degrees of freedom and should not exceed the value for  $P = 0.05$ . In the present case no divergence from parallelism is indicated. The above design for an individual assay holds equally for those repeated frequently in the same laboratory, but in the latter case a modified calculation may be preferred.<sup>5,6</sup>

C. I. BLISS

#### CONNECTICUT AGRICULTURAL EXPERIMENT

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<sup>4</sup> R. A. Fisher and F. Yates, "Statistical Tables for Biological, Agricultural and Medical Research," Oliver and Boyd, 1943.

<sup>5</sup> C. I. Bliss, *Jour. Amer. Statistical Assn.* (in press).

<sup>6</sup> C. I. Bliss and L. C. Miller (in preparation).

#### BOOKS RECEIVED

- BROOKS, SUMNER CUSHING and M. MOLDENHAUER BROOKS. *The Permeability of Living Cells* (Protoplasma-Monographien Bd. 19). Pp. xviii + 395. J. W. Edwards, Ann Arbor, Mich. \$5.00. 1944.
- CANTAROW, ABRAHAM and MAX TRUMPER. *Lead Poisoning*. Pp. xiii + 264. Williams & Wilkins Co. \$3.00. 1944.
- CONRAD, VICTOR A. *Methods in Climatology*. Illustrated. Pp. xx + 228. Harvard University Press. \$4.00. 1944.
- GARRETT, S. D. *Root Disease Fungi*. Illustrated. Pp. 177. Chronica Botanica Company. \$4.50. 1944.
- MOVIUS, HALLAN L. *Early Man and Pleistocene Stratigraphy in Southern and Eastern Asia*. Illustrated. Pp. 125. 6 tables. Peabody Museum, Cambridge 38, Mass. \$3.75. 1944.
- NEVILLE, ERIC HAROLD. *Jacobian Elliptic Functions*. Illustrated. Pp. xiii + 331. Oxford University Press. 1943.
- SELTZER, CARL C. *Racial Prehistory in the Southwest and the Hawikuh Zunis*. Pp. 37. 15 tables. Peabody Museum, Cambridge 38, Mass. \$3.75. 1944.
- WILLET, HURD C. *Descriptive Meteorology*. Illustrated. Pp. viii + 310. Academic Press, Inc. \$4.00. 1944.

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