tallization was present, but after heating at 100° *in vacuo* the anhydrous form was obtained. This anhydrous product darkened at 205° and melted with decomposition at 220–226° (micro-block).

Assays were carried out by Dr. H. B. Woodruff and Mr. D. Hendlin, of the Microbiological Department. Employing *B. subtilis* as a test organism in a cup assay method which they developed, they found an activity of about 350 units/mg for streptomycin helianthate. The various other salts described in this paper were assayed in the same manner.

The results of some microanalytical determinations on the dried streptomycin helianthate follow: Found: C, 50.38, 50.29; H, 5.86, 5.56; N, 14.48, 14.64; S, 5.76.

Streptomycin helianthate was converted to the hydrochloride by treating the salt with a mixture of methyl alcohol and hydrochloric acid. The liberated helianthine was removed and the hydrochloride was precipitated with ether from the filtrate. The hydrochloride was obtained as a white powder. When dried at 25° *in vacuo* over phosphorus pentoxide, streptomycin hydrochloride showed a specific rotation $(\alpha)_D = -84°$ (C, 0.5 per cent. in water), and an activity of about 800 units/mg. The results of microanalytical determinations on a sample dried at 100° *in vacuo* follow: Found: C, 36.60, 36.42; H, 6.04, 6.20; N, 13.42; Cl, 14.80; ash, none. Tests for the presence of sulfur and phosphorus gave negative results. The ultraviolet adsorption spectra of streptomycin in phosphate buffer at pH 7, in glycine buffer at pH 2 and in borate buffer at pH 9 showed only end absorption below about 2,300 Å.

The above analytical data alone are not sufficient for establishing firmly the true empirical formulae of streptomycin, its hydrochloride and its helianthate. These results need to be interpreted in conjunction with molecular weight determinations and other information. Furthermore, the possible molecular weight of about 700 for streptomycin hydrochloride adds to the difficulties of exact determination of empirical formulae. After more complete data are obtained, the results will be presented in detail.

The crystalline salt of streptomycin and *p*-(2-hydroxy-1-naphthylazo)-benzenesulfonic acid was prepared from streptomycin hydrochloride and Orange II. This salt showed an activity of about 300 units/mg. The preparations of streptomycin sulfate, which have been obtained crystalline, have shown about 520 units/mg.

Streptothricin helianthate was prepared using the same method. The crystals melted with decomposition at 220–225° (micro-block) and had an activity of about 400 units/mg.

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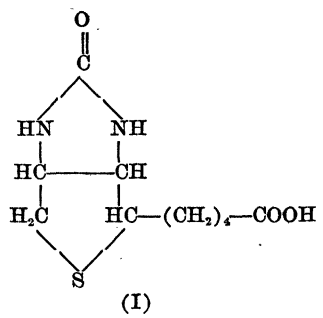
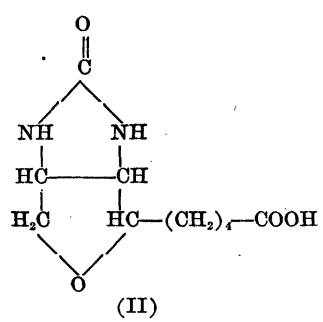
THE MICROBIOLOGICAL ACTIVITY OF AN OXYGEN ANALOG OF BIOTIN

THE synthesis of a *dl*-hexahydro-2-oxo-l-furo [3, 4] imidazole-4-valeric acid, an oxygen analog of biotin, has been recently described.[1] The structural relationship between biotin (I) and this new compound (II) is indicated by the following formulae.



(I)



(II)

The growth-stimulating activity of the oxygen analog under conditions in which biotin is the limiting factor has now been determined for three microorganisms: *Lactobacillus arabinosus*,[2] *Lactobacillus casei*[3] and *Saccharomyces cerevisiae*.[4] The methods were, with slight modifications, those described in the literature cited.

On a weight basis, the *dl* oxygen analog is one half as active as natural *d* biotin when assayed with *L. arabinosus*. Its activity for *L. casei* is slightly lower, approximately 40 per cent. that of *d* biotin. With

[1] K. Hofmann, *Jour. Am. Chem. Soc.*, 67: 694, 1945.
[2] L. D. Wright and H. R. Skeggs, *Proc. Soc. Exp. Biol. and Med.*, 56: 95, 1944.
[3] M. Landy and D. M. Dicken, *Jour. Lab. and Clin. Med.*, 27: 1086, 1942.
[4] R. Hertz, *Proc. Soc. Exp. Biol. and Med.*, 52: 15, 1943.

each of these microorganisms the shapes of the growth curves are identical for the two compounds and the same growth maxima are attained.

However, with *S. cerevisiae* the growth curve of the oxygen analog differs somewhat from that of biotin. Different activity ratios at various portions of the curves are, therefore, obtained. Thus, at one-half maximum growth, the activity of the *dl* oxygen analog is 25 per cent, that of *d* biotin, whereas at maximum growth this compound is only 8 per cent as active. The same growth maximum is obtained with both compounds. Whether the oxygen analog is active as such or exerts activity because of its transformation into biotin remains to be elucidated.

The procedures employed in the synthesis of the oxygen analog have demonstrated the *cis* configuration of the two fused rings. Therefore, only two racemic forms differing in the spatial orientation of the side chain are possible. The *dl* oxygen analog represents one of these racemic forms. Because of its structural relationship to biotin and its comparable biological activity, the name *dl*-oxybiotin is proposed for this compound (II). Since only one of the enantiomorphs of *dl* biotin is biologically active,[5] it appears likely that a similar situation exists for *dl* oxybiotin.

Studies now in progress on the activity of oxybiotin and related compounds, both for animals and for microorganisms, will be presented in subsequent publications.

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OBSERVATIONS ON THE EFFECT OF PREFLIGHT MEALS UPON ALTITUDE TOLERANCE[1]

ALTITUDE tolerance represents only one aspect of preflight and inflight feeding of aviation personnel, but under many circumstances it represents an important consideration. Both empirical observations and carefully controlled experiments indicate the need for food intake at approximately normal meal-time intervals, in order to maintain optimal performance. Hence timing and attractiveness are important aspects of aviation feeding, in addition to provision for an optimal state of nutrition in terms of general health and the maintenance of optimal performance capacity during special stress periods.

The high protein foods, such as meat, milk, eggs and cheese should be consumed in generous quantities, from a long-time point of view, to provide proteins of high quality, minerals and vitamins, and to maintain good morale.

From the point of view of altitude tolerance, however, meals relatively high in carbohydrate afford a distinct advantage when consumed during flight or at the meal preceding altitude exposure. References to the literature and a more detailed discussion of the physiological background of this relationship have been given in an earlier publication,[2] together with experimental data illustrating the gain in altitude tolerance afforded by meals having a relatively high carbohydrate content. Among the factors that apparently contribute to better altitude tolerance after high carbohydrate meals are (a) lesser oxygen demand, (b) higher carbon dioxide production rate, (c) maintenance of glycogen reserves and (d) lessened impairment from alkalosis. In order to achieve the optimum benefits from a high carbohydrate intake, a fairly high caloric intake is necessary, and there is some indication of a slight additional gain from a normal fluid intake.

Not uncommonly a question arises regarding the risk of inter-meal hypoglycemia subsequent to high carbohydrate meals, as a possible factor that might offset the advantages gained during short exposure periods. In the tests performed thus far, however, there has been no evidence of an appreciable risk of that nature when the caloric intake has been adequate.

Because unnecessary impairment is apt to occur after an interval of 6 to 7 hours or less, irrespective of the nature of the preceding meal, there is good reason to provide a convenient reserve or emergency source of food for consumption by flight personnel when unexpected extensions of flight time are encountered.

EXPERIMENTAL

Visual Field: A section of the upper right quadrant of the visual field of the right eye is plotted for sensitivity to a 0.4 mm white test object against a dark ground. The technique was adapted from Evans's procedure for plotting "angioscotoma," using a Lloyd stereocampimeter. The recorded figure is the area of campimeter surface (square inches) within which the object is not seen.

[5] J. L. Stokes and M. Gunness, *Jour. Biol. Chem.*, 157: 121, 1945.

[1] The work described in this paper was done under a contract recommended by the Committee on Medical Research, between the Office of Scientific Research and Development (Contract OEMcmr-224) and Columbia University, and with the aid of a grant from the Nutrition Foundation.

[2] C. G. King, Hylan A. Bickerman, Winifred Bouvet, C. J. Harrer, James R. Oyler and C. P. Seitz, *Jour. Aviation Medicine* (1945, 16: 69).