in better than 98% purity by the dehydration of 3,3-dimethyl-1-butanol over this catalyst. Previous methods of dehydrating this alcohol were accompanied by appreciable rearrangement.[5]

The mechanism suggested here seems to be the most reasonable. However, one involving participation by a neighboring group is not excluded.[6]

Further work with these and other alcohols and with bases other than ammonia and the study of the details of the mechanism are currently being carried out.

(5) V. N. Ipatieff, W. W. Thompson and H. Pines, THIS JOURNAL, **73**, 553 (1951).

(6) S. Winstein, B. K. Morse, E. Grunwald, K. C. Schreiber and J. Corse, *ibid.*, **74**, 1113 (1952), and other papers in that series.

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RECEIVED MARCH 14, 1960

THE BIOSYNTHESIS OF TETRACYCLINE ANTIBIOTICS

Sir:

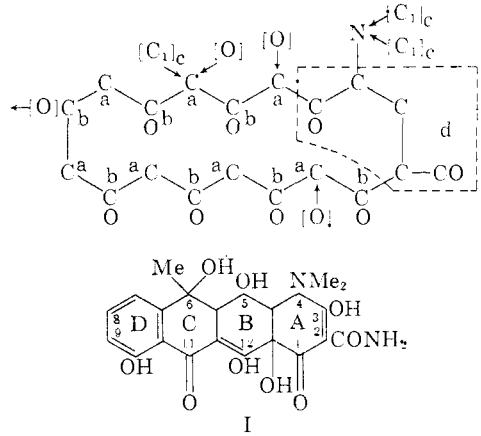
Inspection of the formulas of tetracycline antibiotics, *e.g.*, oxytetracycline, I, suggests that a considerable part of the molecule may arise biosynthetically by the acetic acid route,[1,2,3] with the probable introduction of one methyl group bonded to carbon and two methyl groups bonded to the amino nitrogen group.

The first experimental support for this idea was provided by Snell, *et al.*,[2] who showed that sodium acetate-2-C^{14} was a fermentation precursor (incorporation 5–10%) by a fairly direct route, since the specific activity of the product was directly proportional to the quantity of tracer added. Degradation[2] showed less activity in ring A than in rings B, C and D. Glutamate has been found to be a unique amino-acid source of oxytetracycline in a semi-synthetic medium, and addition to a fermentation of (±)glutamic acid-2-C^{14} gave an active oxytetracycline. Degradation of the product showed that terracinoic acid (from rings B, C, D) contained only 5% of the activity, suggesting that glutamate may be a fairly direct precursor of part at least of ring A.[3]

Detailed degradations have now been carried out on oxytetracycline derived from (i) $C^{14}H_3$-methionine and (ii) 2-$C^{14}H_3CO_2H$. (i) Oxytetracycline (r.m.a.[4] 50.9×10^3) gave by Kuhn–Roth oxidation CH_3CO_2H (r.m.a. of $BaCO_3$ from the CH_3 group, 17.3×10^3; r.m.a. of $BaCO_3$ from the CO_2H, 0) and by standard degradation[5] to tetramethylammonium iodide (r.m.a. 34.8×10^3). All the activity, as expected, is in the C_6-CH_3 and the $N(CH_3)_2$, the activity of all CH_3 groups being the same. (ii) Oxytetracycline (r.m.a. 42.2×10^3) from $C^{14}H_3CO_2H$ after degradations[5] gave products with these r.m.a. $\times 10^{-3}$: terracinoic acid, 27.1; 7-acetoxy-3-methylphthalide 18.7; decarboxyterracinoic acid 26.4; $BaCO_3$ from C_{11}, 0.3; $BaCO_3$ from C_6, 4.0; $BaCO_3$ from C_6-CH_2, 0.3; $BaCO_3$ from C_2-$CONH_2$, 0.6; tetramethylammonium iodide, 0.5.

These results are quantitatively in accord with the production of the molecule by the head to tail linkage of acetic acid units at least from C_5-C_{12} (except C_6-CH_3). It is necessary to assume that the incorporated unit from $C^{14}H_3CO_2H$ has its activity randomized to the extent of 5% and that the methyl-pool has become slightly labelled from this source. It is likely that the glutamate portion extends from the C_2-$CONH_2$, which is differently labelled to an acetate carboxyl, through C_2 to C_{4a}. Degradations to deal with this part of the molecule are inefficient.

The results are in accord with the distributions of label shown in I: (i) r.m.a. contributions of a, b, d = 0, c = 17.3×10^3; (ii) r.m.a. contributions of a = 4×10^3; b = 0.3×10^3; c = 0.25×10^3; with glutamic acid-2-C^{14} as source the r.m.a. contribution of d is much greater than a, b and c.



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We are indebted to Dr. Herchel Smith for assistance with some of the tracer measurements; financial aid from the Rockefeller Foundation and Chas. Pfizer & Co., Inc., supported parts of the work.

(6) A. J. Birch, R. A. Massy-Westropp, R. W. Rickards and H. Smith, *J. Chem. Soc.* (*London*), 360 (1958).

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RECEIVED MARCH 21, 1960

A NEW CLASS OF ACTIVE STEROIDS: THE 19-NOR-$\Delta^{4,9}$-3-KETOSTEROIDS

Sir:

Since the initial demonstration that Δ^1-unsaturation of the corticoids results in enhanced biological and therapeutic activity,[1] numerous investigations have been made of other conjugated systems. The $\Delta^{4,6}$-unsaturated derivatives of the

(1) H. L. Herzog, A. Nobile, S. Tolksdorf, W. Charney, E. B. Hershberg, P. L. Perlman and M. M. Pechet, *Science*, **121**, 176 (1955).

(1) A. J. Birch, Fortschr. Chem. Org. Naturstoffe, Springer, Vienna, **14**, 186 (1957); R. Robinson, "Structural Relations of Natural Products," Oxford Press, New York, N. Y., 1955.

(2) J. F. Snell, R. L. Wagner and F. A. Hochstein, "Internat. Conf. on Peaceful Uses of Atomic Energy," **12**, 431 (1956).

(3) J. F. Snell, in "Radioactivity for Pharmaceutical and Allied Research Laboratories," Academic Press, Inc., New York, N. Y., in press.

(4) Relative molar activity, proportional to molar activity *cf.*[6]

(5) F. A. Hochstein, C. R. Stephens, L. H. Conover, P. P. Regna, R. Pasternak, P. N. Gordon, F. J. Pilgrim, K. J. Brunings and R. B. Woodward, THIS JOURNAL, **75**, 5455 (1953), and references cited therein.