alcohol reduction of the nitro compound. Repetition of their work using racemic camphene gave nitro and amino compounds identical (infrared spectra and other physical constants) with those described above. In addition, lithium aluminum hydride reduction of I yields 3-methylaminoisocamphane (III), b.p. 72° (4 mm.), $n^{25}D$ 1.4881; hydrochloride, m.p. 243–246° dec., *Anal.* Calcd. for $C_{11}H_{22}NCl$: C, 64.83; H, 10.87; N, 6.88. Found: C, 64.54; H, 10.67; N, 6.90, identical with the product obtained in low yield by reaction of methylamine and camphene hydrochloride.

Isolation of 3-formamidoisocamphane from the acid-catalyzed reaction of hydrogen cyanide with camphene led us to re-examine this reaction with a series of simple nitriles. In every case the product was the N-acylisobornylamine to be expected from reaction with Wagner–Meerwein rearrangement, as reported for a few cases by Ritter and Minieri.[1]

The primary amine I shows a significant degree of ganglionic blocking action and this activity becomes pronounced in 3-methylaminoisocamphane (III). In animal experiments the ganglionic blocking properties of III, in terms of potency and specificity, compared favorably with the conventional bisquaternary ammonium drugs of the hexamethonium type. Notwithstanding its chemical dissimilarity, 3-methylaminoisocamphane appears to differ from these only in possessing an inherently longer duration of action and almost quantitative absorption following oral administration.[5]

We have assigned 3-methylaminoisocamphane the generic name mecamylamine, and it is currently undergoing extensive clinical trials.[6]

(5) We are indebted to our collaborators, Drs. K. H. Beyer and C. A. Stone of the Sharp & Dohme Division of Merck & Co., Inc., for the biological data.

(6) Under the Sharp & Dohme Division trade name Inversine ® Hydrochloride.

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RECEIVED FEBRUARY 2, 1956

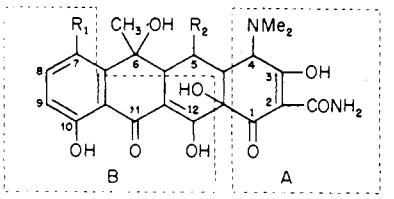
EPITETRACYCLINE—THE CHEMICAL RELATIONSHIP BETWEEN TETRACYCLINE AND "QUATRIMYCIN"

Sir:

In a recent communication[1] from another laboratory, it has been revealed that a reversible isomerization can occur in the antibiotics tetracycline, oxytetracycline and chlorotetracycline. The workers isolated an isomer from tetracycline[2] (I, R's = H) which they called "Quatrimycin." Previous to this report, we had also observed these phenomena and, in addition, had studied the chemical nature of the isomerization reaction. Our studies have established beyond reasonable doubt that racemization at C.4 carbon atom leads

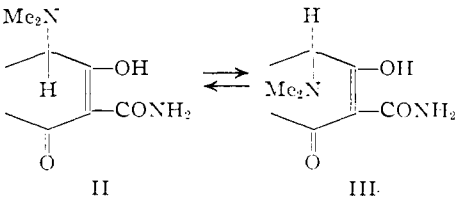
(1) Albert P. Doerschuk, Barbara A. Bitler and J. R. D. McCormick, THIS JOURNAL, **77**, 4687 (1955).

(2) Tetracyn is the registered trade-mark of Chas. Pfizer & Co., Inc., for the antibiotic tetracycline. Achromycin is the registered trade-mark of the Lederle Laboratories Division, American Cyanamid Company, for tetracycline.



I

to the equilibrium mixture previously described. Thus, the relationship between tetracycline and "Quatrimycin" (we have employed the term "epitetracycline" to describe this epimer—m.p. 170–171° (dec.), $(\alpha)^{25}D$ −339° (0.5% in methanol, 0.1 *N* in HCl), *Anal.* Calcd. for $C_{22}H_{24}N_2O_8$: C, 59.45; H, 5.44; N, 6.31. Found: C, 59.63; H, 5.52; N, 6.42) may be illustrated by expressions II and III—though no definite information on



II III

absolute configuration is available. A similar situation would apply in the case of oxytetracycline[3] (I, R = H, R_2 = OH) and chlorotetracycline[4] (I, R_1 = Cl, R_2 = H).

The evidence leading to this conclusion, taking the case of tetracycline as an example, is as follows: The ultraviolet absorption spectrum (*cf.* ref. 1 and Fig. 1) of epitetracycline (quatrimycin) differs significantly from that of tetracycline only in the region where the chromophoric group[5] A (see I) makes its contribution—absorption due solely to group B being essentially identical in each isomer. This relationship limits the site of isomerization to the relatively few atoms associated with group

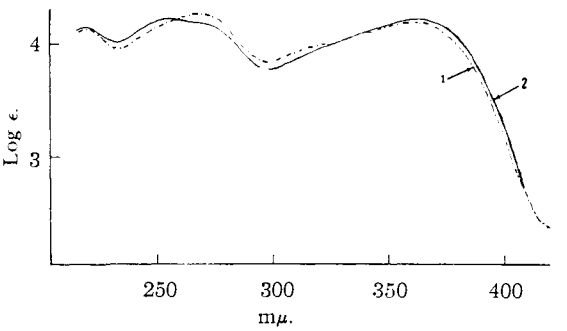


Fig. 1.—Ultraviolet absorption spectra in 0.01 *N* methanolic HCl: 1, · · · · · · tetracycline; (2), ——— epitetracycline.

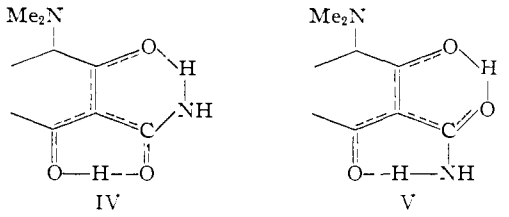
(3) Terramycin is the registered trade-mark of Chas. Pfizer & Co., Inc., for the antibiotic oxytetracycline.

(4) Aureomycin is the registered trade-mark of the American Cyanamid Company for the antibiotic chlorotetracycline.

(5) *Cf.* C. R. Stephens, L. H. Conover, R. Pasternack, F. A. Hochstein, W. T. Moreland, P. P. Regna, F. J. Pilgrim, K. J. Brunings and R. B. Woodward, THIS JOURNAL, **76**, 3568 (1954), and earlier papers for a discussion of the ultraviolet chromophores in the tetracycline series. It has been shown (R. B. Woodward, private communication) that the configuration of the C.12a center has a distinct effect on chromophore B.

A. Comparable degradation studies on tetracycline and epitetracycline confirm this spectral evidence in establishing the area involved.

In group A only two possibilities exist for isomerization. These include epimerization at C.4 and enol tautomerism involving such structures as IV and V in which the strongly chelated carboxamide group could assume different orientations.



IV V

The latter possibility can be excluded since the epimers give isomeric nitrile derivatives, 10-benzenesulfonyltetracylinonitrile ($(\alpha)^{25}D$ (1% in dimethylformamide) −470°, *Anal.* Calcd. for $C_{28}H_{26}N_2O_9S$: C, 59.36; H, 4.62; N, 4.94. Found: C, 59.46; H, 5.02; N, 4.64) and 10-benzenesulfonylepitetracyclinonitrile ($(\alpha)^{25}D$ (same solvent) −431°, *Anal.* C, 59.83; H, 4.89; N, 5.19) on treatment with benzenesulfonyl chloride in pyridine solutions.[6] The nitrile derivatives show absorption differences similar to those observed with the parent epimeric amides (*cf.* Fig. 2).

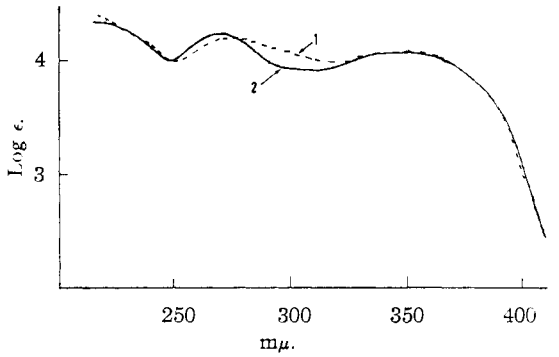


Fig. 2.—Ultraviolet absorption spectra in 0.01 *N* methanolic HCl: 1, - - - 10-benzenesulfonyltetracyclinonitrile; 2, ——— 10-benzenesulfonylepitetracyclinonitrile.

Further confirmation of the epimerization reaction is found in a study of the properties of desdimethylaminotetracycline[6] (m.p. 210–215° (dec.) $(\alpha)^{25}D$ (0.5% in methanol, 0.1 *N* in HCl) −260°, *Anal.* H, 4.72; N, 3.47). This compound, differing from tetracycline only in that the $-NMe_2$ is replaced by H (thus removing the asymmetry at C.4), undergoes no change when subjected to conditions which rapidly epimerize either tetracycline or epitetracycline.

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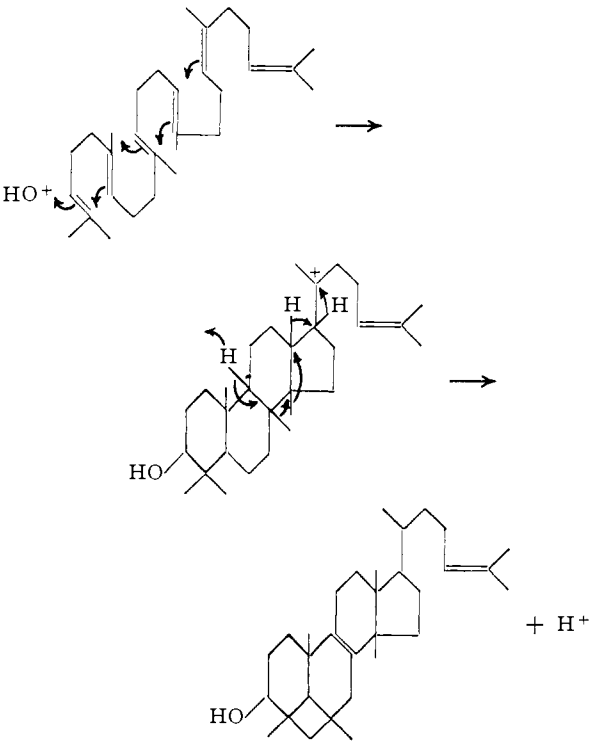
(6) Reference 5 includes a discussion of similar products from chlortetracycline and oxytetracycline.

ON THE MECHANISM OF CYCLIZATION OF SQUALENE[1]

Sir:

The transformation of squalene to lanosterol[2] and of lanosterol to cholesterol[3] has been reported recently. In order to rationalize the biogenetic relation between squalene and the steroids, and the origin of lanosterol in particular, it had earlier been proposed[4] that in the cyclization process the triterpenoid chain assumes the folded form shown in Fig. 1.[5] In their comprehensive theoretical treatment of the mechanism of steroid and triterpene



biogenesis, Ruzicka[6] and Eschenmoser, *et al.*,[7] arrive at the important conclusion that the transformation of squalene to lanosterol is a concerted or non-stop process, *i.e.*, it occurs without formation of stabilized intermediates. In their formulation the cyclization is initiated by the attack of an hypothetical electrophilic OH^+. We now wish to present the results of studies which provide strong experimental evidence in support of a concerted reaction mechanism. Moreover, it is demonstrated that molecular oxygen is involved in this oxidative cyclization.

In the first series of experiments, squalene was incubated with liver homogenate in the presence of

(1) Supported by grants-in-aid from the National Science Foundation and the Life Insurance Medical Research Fund.

(2) T. T. Tchen and K. Bloch, THIS JOURNAL, **77**, 6085 (1955).

(3) R. B. Clayton and K. Bloch, *J. Biol. Chem.*, **218**, 305 and 319 (1956).

(4) R. B. Woodward and K. Bloch, THIS JOURNAL, **75**, 2023 (1953); W. G. Dauben, *et al.*, *ibid.*, **75**, 3038 (1953).

(5) Since the limited space does not permit a discussion of the stereochemistry of the cyclization process, the configuration of the methyl groups and of the hydrogens is not indicated in Figure 1.

(6) L. Ruzicka, *Experientia*, **9**, 359 (1953).

(7) A. Eschenmoser, L. Ruzicka, O. Jeger and D. Arigoni, *Helv. Chim. Acta*, **38**, 1890 (1955).