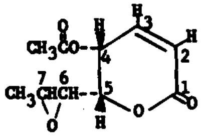
Tetrahedron Letters No.18, pp. 1969-1973, 1966. Pergamon Press Ltd.
Printed in Great Britain.

THE STRUCTURE OF U-13,933, A NEW ANTIBIOTIC

A. D. Argoudelis and J. F. Zieserl
Research Laboratories
The Upjohn Company
Kalamazoo, Michigan

(Received 7 February 1966)

Antibiotic U-13,933 is a new crystalline compound produced by *Aspergillus nidulans*. Isolation of this compound[1], which has shown antibacterial activity *in vitro*, has been achieved by extraction of the clear broth with ethyl acetate, followed by Florisil chromatography and crystallization from a Skellysolve B-ethyl acetate mixture. We now wish to present evidence which assigns structure I to antibiotic U-13,933.

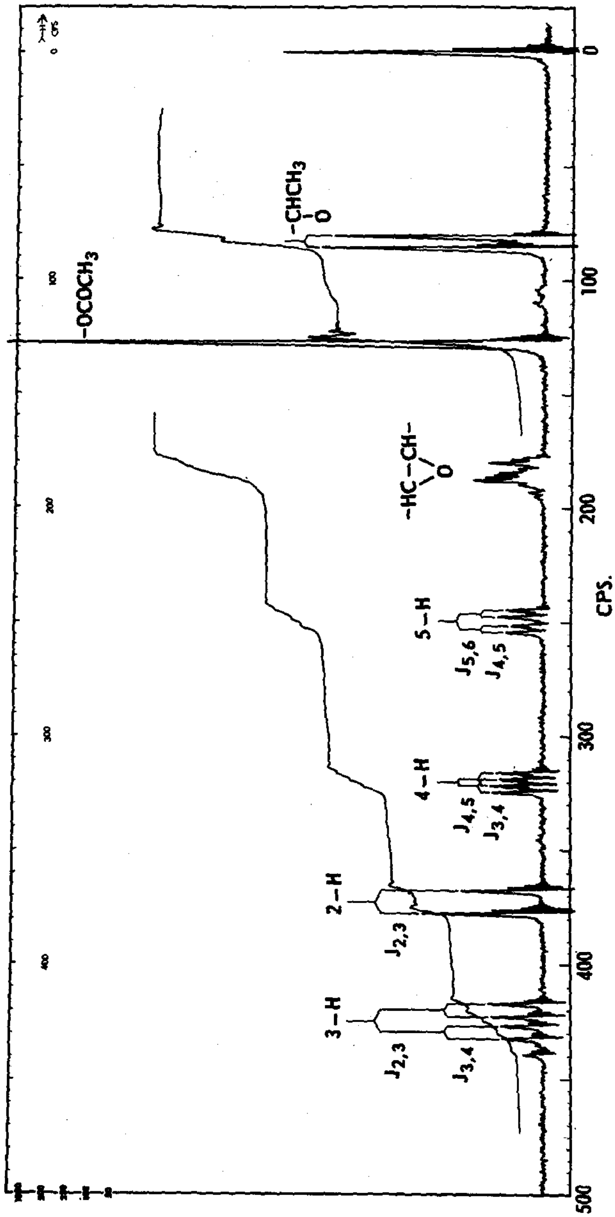


I

U-13,933 has the molecular formula $C_{10}H_{12}O_5$, m.p. 71-73°, $[\alpha]_D^{25}$, +345° (c, 0.9, 95 per cent ethanol). *Anal*. Calc'd for $C_{10}H_{12}O_5$: C, 56.65; H, 5.66; O, 37.69; mol. wt. 212. *Found*: C, 56.95; H, 5.88; O, 37.70; mol. wt. (isothermal distillation in acetonitrile), 208. Potentiometric titration showed the absence of titratable groups. The presence of an α,β-unsaturated δ-lactone (or ester) in U-13,933 is indicated by its ultraviolet spectrum $[\lambda_{max.}^{EtOH}$ 204 mμ $(\varepsilon = 12{,}000)]$[2,3] and infrared absorption at 1715 and 1652 cm^{-1}. The infrared spectrum, which is characterized by

FIG. 1

N.M.R. Spectrum of Antibiotic U-13,933



the complete absence of any absorption in the -OH,NH region, showed an additional ester carbonyl absorption at 1735 cm^{-1}, which is due to an acetate grouping. This is confirmed by the presence of a sharp absorption peak at 128 c.p.s. (3H) in the n.m.r. spectrum of U-13,933.

The n.m.r. spectrum of U-13,933 in deuterochloroform[4] (Figure 1) showed absorptions at ca 425, 373, 320, and 249 c.p.s., each due to a single proton, at 185 c.p.s. due to two protons, and at 128 and 83 c.p.s. each due to three protons. Absorptions at 373 and 425 c.p.s. are assigned to single protons on C-2 and C-3 of I, respectively, coupled to each other, $J_{2,3}$ = 10 c.p.s. indicating cis olefinic function.[5] The presence of the carbonyl group (at C-1) accounts for the shift of the vinyl-hydrogen absorptions to frequencies lower than normally expected.[6] While the C-2 proton appears as a doublet, the C-3 proton appears as a doublet of doublets due to additional coupling to a single proton at C-4, $J_{3,4}$ = 5.5 c.p.s. The proton on C-4 appears also as a doublet of doublets at 320 c.p.s. This is due to coupling to a single proton on C-5, $J_{4,5}$ = 3 c.p.s. To account for the low field position of the protons on C-4 (320 c.p.s.) and C-5 (249 c.p.s.), C-4 and C-5 must be attached to oxygen functions. While the oxygen on C-5 is part of the δ-lactone ring, the oxygen on C-4 is part of the acetate grouping. The latter assignment accounts for the deshielding of the C-4 proton and is in agreement with the lack of -OH or NH absorptions in the I.R. spectrum. The proton on C-5, which appears also as a doublet of doublets, shows coupling to a single proton on C-6, $J_{5,6}$ = 6.5 c.p.s.; this C-6 proton must be in the area of absorption of two protons at 185 c.p.s. Furthermore, the doublet absorption at 83 c.p.s. (3H) is due to a methyl group coupled to a single proton, which must be the second proton absorbing at 185 c.p.s. Hence, the complex absorption at 185 c.p.s. is due to two tertiary protons, one of which (on C-6) is coupled to the proton on C-5 and the other (on C-7) to the C-CH$_3$ group. The complexity of these absorptions indicates that these two protons are coupled to each other. The frequencies of the two protons (185 c.p.s.) and of the methyl doublet suggest oxygen function on both C-6 and C-7.[7] However, this would require a C-6/C-7 epoxide group. This assignment is supported by the proton signals at 185 c.p.s. (2H)[8] and by absorption bands at 1252, 912 and 830 cm^{-1} in the I.R. spectrum.[9]

Aside from absolute configuration considerations, the observed coupling, $J_{4,5}$ = 3.c.p.s., suggests a *cis* arrangement of the hydrogens on C-4 and C-5.[10] The stereochemistry at C-6 and C-7 (*trans* or *cis* arrangement on the epoxide ring) and the absolute stereochemistry remain to be examined. We hope that work in progress will resolve these questions.

Acknowledgments - The authors are grateful to members of the Physical and Analytical Chemistry Department for microanalytical and spectral data and to Drs. George Slomp and P. F. Wiley for helpful discussion and criticism.

References

(1) A. D. Argoudelis, J. H. Coats and R. R. Herr, *Antimicrobial Agents and Chemotherapy*, 1965 (in press).

(2) A. I. Scott, *Interpretation of the Ultraviolet Spectra of Natural Products*, p. 82, Pergamon Press, New York, New York (1964).

(3) U. Eisner, J. A. Elvidge and R. P. Linstead, *J. Chem. Soc.*, 1372 (1953).

(4) N.m.r. spectra were observed in deuterochloroform on a Varian A-60 spectrometer using internal tetramethylsilane as a reference.

(5) L. M. Jackman, *Applications of Nuclear Magnetic Resonance Spectroscopy in Organic Chemistry*, p. 85, Pergamon Press, New York, New York, 1959.

(6) K. Nakanishi, *Infrared Absorption Spectroscopy*, p. 224, Holden-Day, Inc., San Francisco, 1962.

(7) K. Nukada, O. Yamamoto, T. Suzuki, M. Takeguchi, and M. Ohnishi, *Anal. Chem.* *35*, 1892 (1963).

(8) N. S. Bhacca and D. H. Williams, *Applications of N.M.R. Spectroscopy in Organic Chemistry*, p. 99, Holden-Day, Inc., San Francisco, 1964.

(9) Reference 6, page 36.

(10) Stereomodels indicated that, with the lactone ring in any of the possible chair-like or boat-like conformations, the dihedral angle for the hydrogens on C-4 and C-5 would be large (160-170°) if both substituents (acetate on C-4, alkyl chain on C-5) were equatorial and small (35-60°) if one of the substituents was equatorial and the other axial. (Diaxial arrangement of the substituents on C-4 and C-5 is considered to be energetically unfavorable.) The high dihedral angle is inconsistent with the low coupling constant ($J_{4,5} \doteq 3$ c.p.s.) (Ref. 8, pp. 50-51), therefore suggesting a *cis* (equatorial-axial) relationship between the hydrogens on C-4 and C-5.