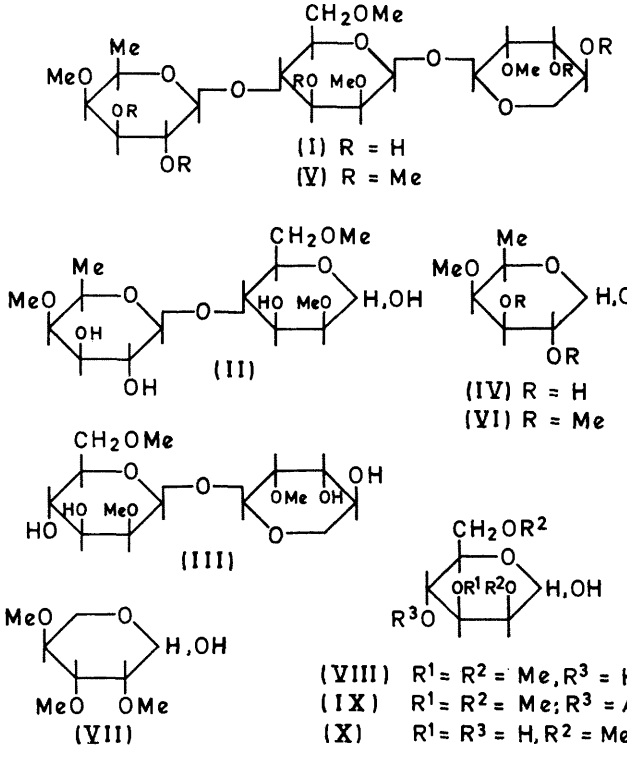
Structure of Evertriose, a Trisaccharide Component of Everninomicin D[1]

By ASHIT K. GANGULY* and OLGA Z. SARRE

(*Natural Products Research Department, Schering Corporation, Bloomfield, New Jersey* 07003)

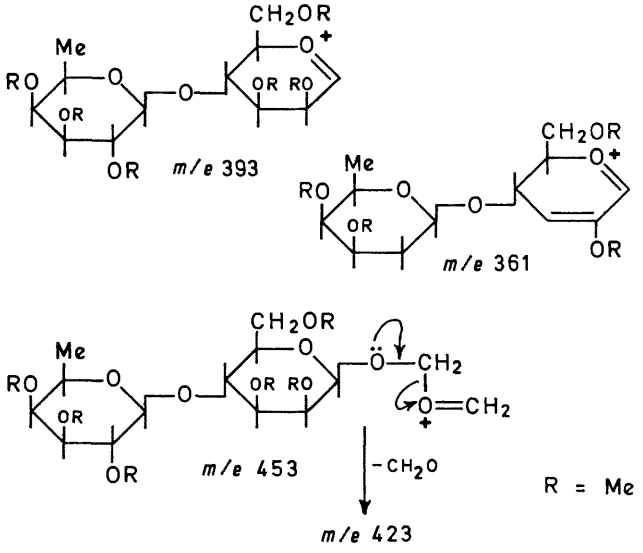
Summary Evertriose, a trisaccharide component of everninomicin has been shown to be (I).

EVERNINOMICIN D on hydrolysis with aqueous acid yields a mixture of compounds from which we have isolated a trisaccharide, evertriose (I) and a disaccharide (II).



Evertriose, $C_{21}H_{38}O_{14},H_2O$,† $[\alpha]_D$ — 41·6° (H_2O, 24 h), is an amorphous solid. It is non-reducing, shows no selective absorption in the u.v., and in the i.r. (Nujol) it does not show any carbonyl absorption. The n.m.r. spectrum (D_2O) of (I) shows the presence of four methoxy-groups, a secondary methyl (δ 1·3; *J* 6·5 Hz), and three anomeric protons at δ 4·31 (*J* 7 Hz), 4·95 ($J_{w/2}$ 1·5 Hz), and 5·33 (*J* 2·5 Hz). Evertriose (I), on further hydrolysis, yielded everninose[1] (III), m.p. 200—201°, $[\alpha]_D$ — 74·1° (water) and D-curacose (IV), m.p. 125—127°, $[\alpha]_D$ + 98·4° → + 86·4° (water), which formed a tosylhydrazone identical with an authentic sample of D-curacose tosylhydrazone,[2] (m.p., mixed m.p., $[\alpha]_D$, t.l.c., i.r.). The identity of everninose was proved by direct comparison with an authentic sample. We have previously reported[1] the structure of everninose as (III).

Evertriose on methylation[3] yielded a permethylated compound (V), $C_{26}H_{48}O_{14}$ (*m/e* 584), $[\alpha]_D$ — 47·7°. The n.m.r. spectrum of (V) showed nine methoxy-groups, one secondary methyl group at δ 1·33 (d, *J* 6·5 Hz), and three anomeric protons. Two of the anomeric protons at δ 5·30 (d, *J* 2 Hz) and 4·73 (d, *J* 1·5 Hz) belonged to the everninose portion of the molecule, and the third one at δ 4·36 (d, *J* 7 Hz) was attributed to the curacose portion and therefore established the β-anomeric linkage. The mass spectrum of (V), besides showing the molecular-ion peak at *m/e* 584, showed prominent peaks at *m/e* 393, 361, 155, 453, 452, 423, 439, 379.



The mass-spectral fragmentation pattern of (V) indicated that in evertriose (I), D-curacose (IV) was linked to the 4-position of the hexose moiety of everninose (III). Prolonged acid hydrolysis of (V) yielded a mixture which was separated using preparative t.l.c. into (VI), (VII), and 2,3,6-tri-*O*-methyl-D-mannose (VIII), syrup, $C_9H_{18}O_6$, $[\alpha]_D$ + 6·9° → + 5·9° (methanol). The structure of (VIII) was proved by acetylation to (IX), crystallized from acetone–hexane, m.p. 95°, $C_{13}H_{22}O_8$, $[\alpha]_D$ + 28·4°. The n.m.r. spectrum of (IX) showed the presence of three methoxy-groups (δ 3·88, 3·56, and 3·45), two acetoxy-groups (δ 2·11 and 2·15), an anomeric proton at δ 6·28 (*J* 2 Hz), and a 1H triplet at δ 5·28 (*J* 9 Hz) ascribed to the 4-H. These facts established the structure and stereochemistry of evertriose as (I). The stereochemistry of the anomeric linkage (curacose part) was shown from the n.m.r. spectrum and was further confirmed by the application of Klyne's rule.[5]

The structure of the reducing disaccharide (II), syrup, $C_{15}H_{28}O_{10}$, $[\alpha]_D$ + 12·9° → + 13·8° (water) followed from the evidence below. The n.m.r. spectrum of (II) showed the presence of three methoxy-groups (δ 3·36, 3·45, and 3·56), two anomeric protons (δ 5·31, *J* 2 Hz; δ 4·3, *J* 7 Hz) and a methyl doublet (δ 1·3; *J* 6·5 Hz). The mass spectrum

† Satisfactory elemental analyses were obtained for all new compounds; unless otherwise noted, i.r. spectra were recorded in chloroform solution and n.m.r. spectra were taken at 60 MHz in $CDCl_3$ with internal $SiMe_4$ standard; optical rotations were measured in chloroform solution at 25°.

of the *O*-trimethylsilyl ether of (II) did not show a molecular-ion peak but showed[4] peaks at *m/e* **567** (M — $OSiMe_3$), **535** (M — $OSiMe_3$ — CH_3OH), and **551** M — CH_3 — $SiMe_3OH$). Other prominent peaks were at *m/e* **479**, **464**, **153**, **335**, and **305**. On hydrolysis with aqueous acid the disaccharide (II) yielded D-curose (IV) and 2,6-di-*O*-methyl-D-mannose (X).[1]

Elucidation of the structure of evertriose (I) thus establishes the sequence of a three-sugar fragment in evernimicin D.

(*Received, May 11th*, 1970; *Com.* 715.)

[1] Previous publication: A. K. Ganguly, O. Z. Sarre, and J. Morton, *Chem. Comm.*, 1969, 1488.
[2] E. G. Gross, *Carbohydrate Res.*, 1966, **2**, 56. We thank Dr. E. G. Gross, Universidad de Buenos Aires, Argentina, for sending us a sample of D-curacose tosylhydrazone.
[3] H. G. Walker, jun., M. Gee, and R. M. McCready, *J. Org. Chem.*, 1962, **27**, 2100.
[4] N. K. Kochetkov, O. S. Chizhov, and N. V. Molodtsov, *Tetrahedron*, 1968, **24**, 5587.
[5] W. Klyne, *Biochem. J.*, 1950, **47**, XLI.