279. C(8) Epimeric 8-Hydroxy-erythromycins-A

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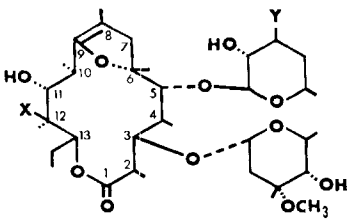
Zusammenfassung. Die sorgfältig kontrollierten Persäure-Oxydationen von 8,9-Anhydro-erythromycin-A-6,9-hemiacetal-N-oxid und 8,9-Anhydro-erythromycin-A-6,9-hemiacetal liefern nach der katalytischen Reduktion der N-Oxidgruppe direkt und stereoselektiv das (8*S*)-8-Hydroxy-erythromycin-A nebst einer geringen Menge des 8-*epi*-(8*R*)-8-Hydroxy-erythromycins-A. Die Resultate der Persäure-Oxydationen der 8,9-Anhydro-erythromycin-A- und B-hemiacetale werden verglichen.

We recently reported [1] that oxidation of 8,9-anhydro-erythromycin-B-6,9-hemiacetal (**1**) with *m*-chloroperbenzoic acid in chloroform in the presence of 5% aqueous sodium hydrogen carbonate, followed by catalytic reduction of the resulting N-oxide mixture, gave rise to the diastereomeric epoxides **2** and **3**. The pure epoxides were converted stereospecifically to the corresponding C(8) epimeric 8-hydroxy-erythromycins-B **4** and **5**. The latter, 8-*epi*-(8*R*)-8-hydroxy-erythromycin-B was found to exist as a mixture of interconverting hydroxyketone **5a** and hemiacetal **5b** tautomers.

We have now found that treatment of 8,9-anhydro-erythromycin-A-6,9-hemiacetal-N-oxide (**6a**) [2] in chloroform with a slight excess of *m*-chloroperbenzoic acid in the presence of 5% aqueous sodium hydrogen carbonate, followed by reduction of the N-oxide product, leads directly to the formation of (8*S*)-8-hydroxy-erythromycin-A (**7**). The latter, **7**, exists as a mixture of two interconverting tautomers, the hydroxy ketone **7a**, and either the 6,9-hemiacetal **7b** or the 9,12-hemiacetal **7c**. (8*S*)-8-Hydroxy-erythromycin-A (**7**) was characterized both spectroscopically, and by its conversion to (8*S*)-8-hydroxy-erythromycin-A-6,9;9,12-acetal (**9**).

Careful chromatography of the reaction product yielded a minor product which has been identified as 8-*epi*-(8*R*)-8-hydroxy-erythromycin-A (**8**) which appears to exist predominantly as the 6,9- or the 9,12-hemiacetal (**8b** or **8c**).

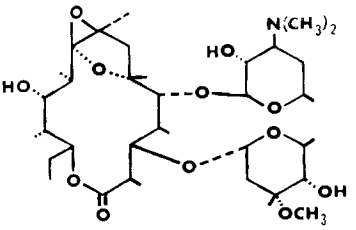
Buffered epoxidation (chloroform/5% aqueous sodium hydrogen carbonate) of 8,9-anhydro-erythromycin-A-6,9-hemiacetal (**6b**) with two to four equivalents of *m*-chloroperbenzoic acid gave a much less pure, and much more difficultly purifiable product than that prepared as described above. Brief, unbuffered oxidation of **6b** with monoperphthalic acid in ether, followed by catalytic reduction of the N-oxide product gave (8*S*)-8-hydroxy-erythromycin-A (**7**) of good quality. Unbuffered oxidation of **6b** with excess *m*-chloroperbenzoic acid in chloroform gave predominantly (8*S*)-8-hydroxy-erythromycin-A-6,9;9,12-acetal (**9**) together with some erythromycin-A-6,9;9,12-acetal (**12**) [3] and 8,9-*seco*-8-oxo-erythromycin-A-9-oic-acid-6,9-lactone (**10**).



1 : X = H; Y = $N(CH_3)_2$

6a: X = OH; Y = $N^{\rightarrow O}(CH_3)_2$

6b: X = OH; Y = $N(CH_3)_2$



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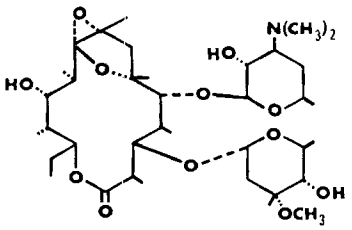

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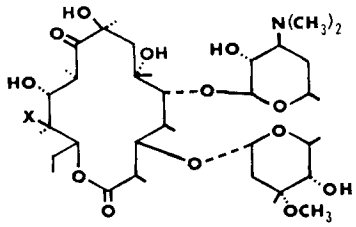
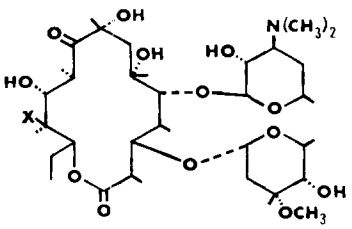


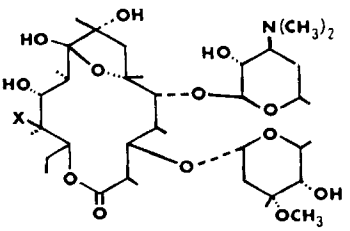
4 : X = H

7a: X = OH



5a: X = H

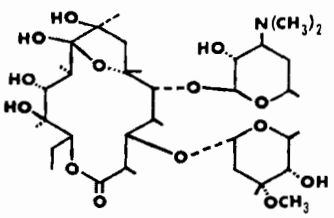
8a: X = OH



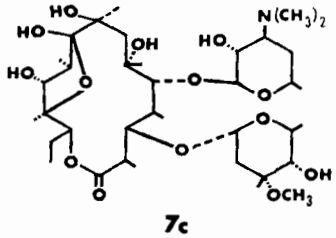
5b: X = H

8b: X = OH

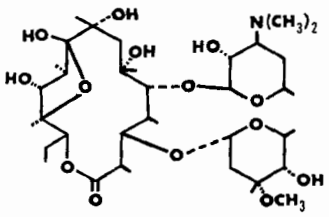
In no case were there detected any 8,9-anhydro-erythromycin-A-6,9-hemiacetal-8,9-epoxides analogous to those (**2** and **3**) formed by oxidation of 8,9-anhydro-erythromycin-B-6,9-hemiacetal (**1**). This difference between the nature of the products formed by peracid oxidation of the 8,9-anhydro-erythromycin-A- and -B-6,9-hemiacetals **6** and **1** is most probably the result of a neighboring group effect by the C(12)hydroxyl of **6**. Some precedence exists in the literature for such neighboring group effects on the course of similar peracid oxidations [4].



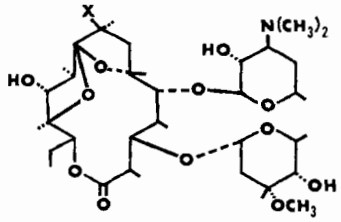
7b



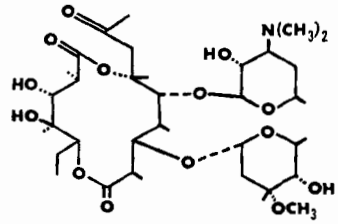
7c



8c



9: X=OH
12: X= H



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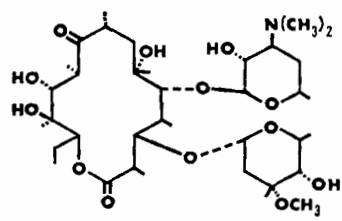

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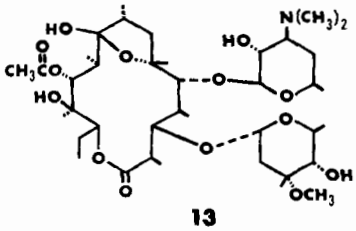

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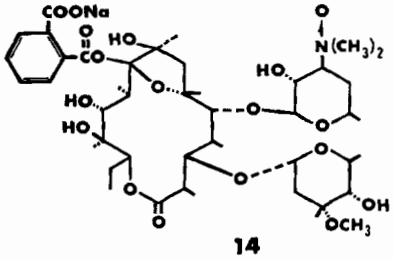

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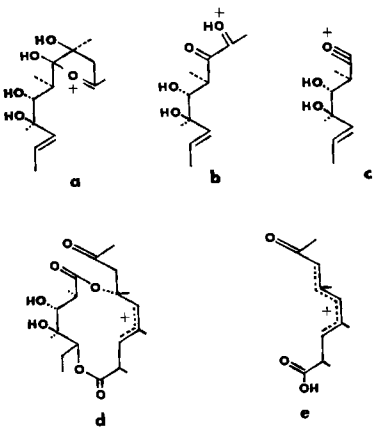
Structural Assignments. The high resolution mass spectra of the C(8) epimeric 8-hydroxyketones **7** and **8** were in agreement with the assigned structures. In the mass spectrum of (8*S*)-8-hydroxy-erythromycin-A (**7**) the molecular ion exhibited a signal at $m/e = 749$ which was too weak for an exact mass determination; the molecular ion of 8-*epi*-(8*R*)-8-hydroxy-erythromycin-A (**8**) was observed at $m/e = 749$ ($C_{37}H_{67}NO_{14}$, 1.6%). The fragmention ions of particular interest for the structure proofs of **7** and **8** arose from the *McLafferty*-rearrangement products of their molec-

ular ions [1] [5]. The ion **a**[1]) of $m/e = 273$ ($C_{14}H_{25}O_5$, 0.4%) arose by α-cleavage of the 5,6-bond of the rearranged molecular ion of **7**. The sequential loss of two molecules of water from **a** resulted in fragments at $m/e = 255$ ($C_{14}H_{23}O_4$, 0.9%) and at $m/e = 237$ ($C_{14}H_{21}O_3$, 1.9%), respectively. The signal of the C(8)epimeric ion corresponding to **a** resulting from a similar cleavage of **8** was too weak for an exact mass determination. However, the ions arising by dehydration of this fragment at $m/e = 255$ ($C_{14}H_{23}O_4$, 0.3%) and $m/e = 237$ ($C_{14}H_{21}O_3$, 1.3%) were identified in the mass spectrum of **8**. The low abundance of the ions **b** ($C_{11}H_{19}O_4$) at $m/e = 215$ resulting from the α-cleavages of the 7,8-bonds of both **7** and **8** precluded exact mass determinations. The signals observed in the mass spectra of both **7** and **8** at $m/e = 197$ ($C_{11}H_{17}O_3$, 0.8% and 0.6%, respectively) and $m/e = 179$ ($C_{11}H_{15}O_2$, 3.2% and 1.9%, respectively) must have arisen from **b** by the loss of one and two molecules of water.



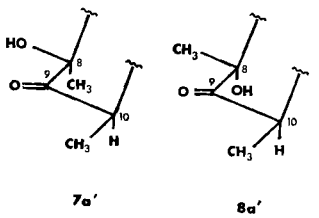
The α-cleavages of the 8,9-bonds in the rearranged molecular ions of **7** and **8** resulted in the fragments **c** of $m/e = 171$ ($C_9H_{15}O_3$, 1.5% and 1.0%, respectively) and ions at $m/e = 153$ ($C_9H_{13}O_2$, 3.0% and 1.6%, respectively) which are dehydration products of **c**. The presence of signals in the mass spectra of **7** and **8** of **b** and **c** and their dehydration products clearly located the position of the additional hydroxyl group in **7** and **8** at C(8).

Several ions observed in the high resolution mass spectrum of the *seco*-compound **10** confirmed its structure. The molecular ion gave rise to a signal at $m/e = 747$ ($C_{37}H_{65}NO_{14}$, 2.3%). An ion at $m/e = 704$ (M-43, $C_{35}H_{62}NO_{13}$, 0.8%) resulted from the cleavage of the 7,8-bond of the molecular ion with the loss of an acetyl radical. The cleavage of both sugars from the molecular ion gave rise to **d**[2]) at $m/e = 397$ ($C_{21}H_{33}O_7$, 6.2%) which represents the aglycone portion of **10**. The observation of a signal for the ion **e** at $m/e = 209$ resulting from a second *McLafferty*-rearrangement of **d**[2]) could be explained only in terms of the *seco*-structure **10**. Metastable defocusing carried out at $m/e = 209$ clearly indicated **d** as the precursor of **e**.

[1]) The formulation of **a** as the hemiacetal was preferred for chemical reasons.

[2]) The fragment **d** is most likely present in the form of one or both of its possible *McLafferty*-rearrangement products.

As in the case of C(8) epimeric 8-hydroxy-erythromycins-B [1] circular dichroism measurements confirmed the assignments of configuration at C(8) to the C(8) epimeric 8-hydroxy-erythromycins-A. The hypsochromic shift relative to erythromycin-A ($[\theta]_{290} = -6600$) [6] of the CD. maximum of the keto carbonyl of (8*S*)-8-hydroxy-



7a' **8a'**

erythromycin-A (**7a**) ($[\theta]_{284} = -1640$) requires an equatorial 8-hydroxy group (**7a'**) while the bathochromic shift of the CD. maximum of the keto carbonyl of 8-*epi*-(8*R*)-8-hydroxy-erythromycin-A (**8a**) ($[\theta]_{294} = -1064$) relative to **7a** requires an axial 8-hydroxy group (**8a'**). Formation of the (8*S*)-8-hydroxy-erythromycins-A- and -B as the major products of oxidation of the 8,9-anhydro-erythromycin-A- and -B-hemiacetals, **1** and **6**, is presumably the consequence of the attack of the oxidizing agent from the more accessible face of the 8,9-double bonds of **1** and **6** as previously discussed [1].

The NMR. spectrum of (8*S*)-8-hydroxy-erythromycin-A (**7**) is complicated by a slow interconversion between the hydroxy ketone **7a** and a hemiacetal tautomer (**7b** or **7c**) which results in separate resonances for the two forms. In chloroform-*d* solutions two resonances attributable to the C(8) methyl are found at 1.61 and 1.69 ppm with one form predominating to the extent of about 65%. In pyridine-d_5 solution the tautomeric forms are equally populated. The elevated temperature spectrum in pyridine-d_5 is simplified by the coalescence of many doubled peaks, but detailed interpretation is not possible due to substantial resonance broadening.

The NMR. spectrum of 8-*epi*-(8*R*)-8-hydroxy-erythromycin-A (**8**) is not complicated by slow interconversion of tautomers. Substitution at C(8) is shown by the appearance of a simple *AB* quartet for the C(7) methylene protons and by an additional singlet methyl resonance at 1.55 ppm attributable to the C(8) methyl protons. The remaining parameters (Table) reveal that no other portion of the molecule has been oxidized, but several unusual chemical shifts of possible structural significance are apparent. The upfield shift of H(10) from its position in the spectrum of erythromycin-A (**11**) may be a consequence of the loss of the C(9) keto carbonyl and suggests a predominance of a hemiacetal tautomer (**8b** or **8c**). The marked downfield shifts of H(2) and H(3) relative to those of erythromycin-A (**11**) are unusual and reminiscent of the shifts observed for erythromycin-A-6,9;9,12-acetal (**12**). In addition, the coupling constants $J_{2,3}$, $J_{3,4}$ and $J_{4,5}$ are almost the same as those of **12**, but are quite different from those of **11**. Clearly the data support a close conformational relationship between 8-*epi*-(8*R*)-8-hydroxy-erythromycin-A (**8**) and erythromycin-A-6,9;9,12-acetal (**12**) in the C(1) through C(6) ring segment. Since it has been shown in the case of 11-O-acetyl-erythromycin-A (**13**), which exists as the 6,9-hemiacetal [7], that formation of a 6,9-oxygen bridge does not result in such a *con-*

formational change in the C(1) to C(6) ring segment, the similarities between **8** and **12** may be presumed to arise from a 9,12-oxygen bridge common to both compounds. Thus 8-*epi*-(8*R*)-8-hydroxy-erythromycin-A is formulated as the novel 9,12-hemiacetal **8c**.

The structure of (8*S*)-8-hydroxy-erythromycin-A-6,9;9,12-acetal (**9**) was established by its NMR. spectrum. Substitution at C(8) was indicated by the presence of a simple doublet for H(7a) and an additional deshielded methyl singlet (C(8) methyl) at 1.60 ppm. The remaining NMR. parameters of **9** very closely correspond to those of erythromycin-A-6,9;9,12-acetal (**12**) in every respect, including the chemical shifts of H(2), H(3) and H(11) as well as in the magnitude of $J_{10,11}$.

The structure of 8,9-*seco*-8-oxo-erythromycin-A-9-oic-acid-6,9-lactone (**10**) was confirmed by its NMR. spectrum (Table). The C(7) methylene proton resonances of **10** are approximately 1.0–1.5 ppm farther downfield than are the C(7) methylene resonances of either erythromycin-A (**11**) or (8*S*)-8-hydroxy-erythromycin-A (**7**) and, thus suggest oxidation of C(8) to a carbonyl group. In addition, the C(8) methyl group of **10** appears as a singlet at 2.4 ppm which is indicative of the methyl group of a methyl ketone. The low-field chemical shift of the C(10) proton of **10**, relative to those of the acetals **9** and **12**, and the hemiacetal **13** is characteristic of the presence of the C(9)-carbonyl.

The minimum inhibitory concentrations of (8*S*)-8-hydroxy-erythromycin-A (**7**) and 8-*epi*-(8*R*)-8-hydroxy-erythromycin-A (**8**) *vs Staphylococcus aureus* SMITH in an agar dilution assay were 1.56 and 6.25 mcg per ml, respectively, compared with a value of 0.20 mcg per ml for erythromycin A.

The authors are indebted to Prof. *L. A. Mitscher* and Dr. *M. S. Bathala* for the determination and interpretation of the circular dichroism data. We wish also to thank Mrs. *Ruth Stanaszek* for determining the NMR. spectra, Mr. *W. H. Washburn* for the IR. spectra, Mrs. *Julie Hood* for microanalyses, Mrs. *Esther Hirner* for assistance with column chromatography and Dr. *R. B. Hasbrouck* and Mr. *J. E. Leonard* for many thin layer chromatographies.

Experimental Part

General Remarks. Optical rotations were measured with methanol solutions with a *Hilger* and *Watts* polarimeter. IR. spectra were determined with deuteriochloroform solutions using a *Perkin-Elmer* Model 521 grating spectrometer. CD. spectra were obtained with a *Durrum-Jasco* Model ORD/UV-5 instrument equipped with a CD. attachment and operating at ambient temperatures in spectral grade methanol. Mass spectra were recorded with an *A.E.I.* MS-902 mass spectrometer with an ionizing energy of 70 eV; samples were introduced into the source by a direct inlet system. NMR. spectra were determined at 100 MHz using a *Varian* HA-100 spectrometer with deuteriochloroform solutions unless specified otherwise; chemical shifts are reported in ppm from internal tetramethylsilane (0δ) and coupling constants are reported in Hz. Silica gel 60 (*Merck*, Darmstadt) was used in partition chromatography [8].

8,9-Anhydro-erythromycin-A-6,9-hemiacetal-N-oxide (**6a**). A solution prepared from 10,0 g of erythromycin-A-N-oxide [9] and 50 ml of glacial acetic acid was allowed to stand at room temperature for 3 h. The major portion of the acetic acid was evaporated under reduced pressure, and the residue was shaken with a mixture of 500 ml of chloroform and 400 ml of ice-cold, saturated, aqueous sodium hydrogen carbonate. The chloroform solution was separated and washed with two 200-ml portions of water. The aqueous solutions were washed in series with two 400-ml portions of chloroform. The chloroform solutions were combined and dried over anhydrous magnesium sulfate. The chloroform solution was concentrated to a small volume, and heptane

Nuclear Magnetic Resonance Parameters*

	Chemical Shifts					
	8	**9**	**10**	**11**	**12**	**13**
H(2)	3.29	~3.3	2.86	2.87	3.27	2.69
H(3)	4.92	4.45	4.11	4.01	4.35	4.11
H(4)	~2.3	~2.1	~2.4	2.00	~2.1	~ 2.2
H(5)	3.59	3.41	3.81	3.57	3.50	3.59
H(7a)	2.80	2.59	3.33	1.91	–	1.91
H(7e)	2.06	–	3.03	1.70	–	–
H(10)	2.69	–	2.74	3.10	3.01	2.22
H(11)	4.54	3.37	3.73	3.86	3.47	4.93
H(13)	4.73	5.18	4.85	5.04	5.12	5.03
H(1′)	4.31	4.23	4.47	4.40	4.24	4.19
H(2′)	3.17	–	3.26	3.22	3.22	3.25
H(1″)	4.92	5.19	4.96	4.89	5.21	4.85
H(4″)	2.97	2.97	3.03	3.00	2.96	3.00
H(5″)	4.03	4.00	4.01	4.05	4.01	4.07
CH_3(6)	1.36	1.31	1.73	1.46	1.43	1.50
CH_3(8)	1.55	1.60	2.14	1.16	1.07	1.23

	Coupling Constants					
	8	**9**	**10**	**11**	**12**	**13**
$J_{2,3}$	3.5	3.0	5.0	9.0	3.0	8.0
$J_{3,4}$	7.0	7.8	2.0	2.0	7.6	~ 1
$J_{4,5}$	5.0	4.5	6.5	7.5	5.0	7.6
$J_{7a,7e}$	14.0	14.0	15.0	15.0	–	14.4
$J_{10,11}$	1.0	10.0	7.5	1.5	11.0	2.0
$J_{13,14a}$	9.5	10.0	10.5	10.6	10.8	9.6
$J_{13,14e}$	4.0	4.0	3.0	2.4	3.5	2.8
$J_{1',2'}$	7.0	7.5	7.5	7.0	7.0	7.1
$J_{2',3'}$	10.0	–	10.5	10.2	10.0	10.2
$J_{1'',2a''}$	3.5	4.5	5.0	4.5	4.5	4.4
$J_{1'',2e''}$	< 1	~ 1	< 1	~ 1	~ 1	< 1
$J_{4'',5''}$	9.5	9.5	9.5	9.5	9.2	9.4

* Measured in $CDCl_3$ at 55°.

was added to initiate crystallization. The product (6.50 g) melted at 182–184°, $[\alpha]_D^{26}$ – 51° (c = 1.05); λ_{max} (CH_3OH) 209 nm (ε = 6697).

$C_{37}H_{65}NO_{13}$	Calc.	C 60.71	H 8.95	N 1.91	O 28.42%
(731,898)	Found	,, 60.60	,, 9.25	,, 2.11	,, 28.12%

m-*Chloroperbenzoic Acid Oxidation of 8,9-Anhydro-erythromycin-A-6,9-hemiacetal-N-oxide* (**6a**) *in the presence of 5% aqueous Sodium Hydrogen Carbonate.* To a vigorously stirred mixture of 8,9-anhydro-erythromycin-A-6,9-hemiacetal-N-oxide (**6a**) in 150 ml of chloroform and 240 ml of 5% aqueous sodium hydrogen carbonate[3]) was added, dropwise, a freshly prepared solution of 1.84 g of *m*-chloroperbenzoic acid in 60 ml of chloroform. After the addition was complete, stirring was

[3]) When the reaction was carried out in the absence of the aqueous sodium hydrogen carbonate buffer, tlc. (after N-oxide reduction) established that only a trace of the 8-hydroxy-erythromycins-A were present in the reaction mixture. The major products were (8*S*)-8-hydroxy-erythromycin-A-6,9;9,12-acetal (**9**) and erythromycin-A-6,9;9,12-acetal (**12**).

continued at room temperature for 18 h. To the resulting mixture was added, dropwise, a solution of 15 ml of cyclohexene in 60 ml of chloroform. Stirring was then continued at room temperature for 6 h. The resulting mixture was shaken with a mixture of 150 ml of 5% aqueous sodium hydrogen carbonate and 130 ml of chloroform. The chloroform solution was washed to neutrality with water. The aqueous solutions were washed in series with five 200-ml portions of chloroform. The chloroform solutions were combined, and the chloroform was evaporated under reduced pressure. The residual chloroform was removed by co-distillation with methanol under reduced pressure to yield 6.14 g of product as the N-oxide. The N-oxide was catalytically reduced in the usual manner [1] to yield 5.98 g of free amine as a white glass.

A portion of the product (1.42 g) was passed through a column (2.8 × 75 cm) of Sephadex LH-20 prepared and eluted with hexane/chloroform 1:1. The first fractions contained fast moving minor components which were discarded. Later fractions contained mixtures of (8*S*)-8-hydroxy-erythromycin-A (**7**) and 8-*epi*-(8*R*)-8-hydroxy-erythromycin-A (**8**). Final eluates gave pure **7**. Repeated chromatography of the fractions containing mixtures of **7** and **8** gave additional quantities of pure **7** for an overall yield of 600 mg of pure (8*S*)-8-hydroxy-erythromycin-A (**7**) as a glass which resisted all attempts at crystallization, $[\alpha]_D^{24} = -52°$ ($c = 1.06$), $\tilde{\nu}_{max}$ 3550–3450, 1728, 1712 (shoulder) cm^{-1}.

$C_{37}H_{67}NO_{14}$ (749.914) Calc. C 59.26 H 9.01 N 1.87% Found C 58.91 H 8.99 N 1.68%

Fractions enriched in 8-*epi*-(8*R*)-8-hydroxy-erythromycin-A (**8**), but still containing considerable amounts of (8*S*)-8-hydroxy-erythromycin-A (**7**) were chromatographed on a column (1.8 × 15 cm) of silica gel packed in benzene. Elution with increasing concentrations of acetone in benzene gave eluates containing **8** and several minor unidentified components. These fractions were combined and evaporated to leave 36 mg of a colorless oil which was passed through a column (1.4 × 90 cm) of Sephadex LH-20 prepared in hexane/chloroform 1:1[4]). Fractions containing only 8-*epi*-(8*R*)-8-hydroxy-erythromycin-A (**8**) were combined and evaporated to yield 24 mg of pure **8**. Crystallization from methanol gave needles, m.p. 138–141°; $\tilde{\nu}_{max}$ 3445, 1710 cm^{-1}; M^+ 749.4568, calc. for $C_{37}H_{67}NO_{14}$ 749.4560.

Monoperphthalic Acid Oxidation of 8,9-Anhydro-erythromycin-A-6,9-hemiacetal (**6b**). A solution prepared from 1.7 g of 8,9-anhydro-erythromycin-A-6,9-hemiacetal (**6b**), 30 ml of ether, and 30 ml of tetrahydrofuran was cooled to 0° and treated with 30 ml of 0.5 M monoperphthalic acid. The resulting solution was allowed to stand at 0° for 45 min, then poured into 300 ml of 5% aqueous sodium hydrogen carbonate and immediately extracted with chloroform (2 × 50 ml) to give extract A. The aqueous phase was allowed to stand at room temperature for 2 h[5]) and again extracted with chloroform to give extract B.

The chloroform was evaporated under reduced pressure from both extracts A and B, and residual chloroform was removed by co-distillation with methanol under reduced pressure. The residual N-oxides were catalytically reduced in the usual manner [1] to the free amines. From fraction A was isolated 574 mg of 8,9-anhydro-erythromycin-A-6,9-hemiacetal (**6b**) while from fraction B was isolated 904 mg of (8*S*)-8-hydroxy-erythromycin-A (**7**). These compounds were identified by their NMR. spectra and by tlc. comparisons with authentic samples described above.

(8*S*)-*8-Hydroxy-erythromycin-A-6,9;9,12-acetal* (**9**). A solution prepared from 422 mg of (8*S*)-8-hydroxy-erythromycin-A (**7**), 7 ml of acetic acid and 7 ml of water was allowed to stand at room temperature for 21 h. The resulting solution was added dropwise to a stirred solution of sodium carbonate in water. Excess solid sodium hydrogen carbonate was added, and the product, 374 mg of a white glass, was isolated by chloroform extraction. Partition column chromatography gave 135 mg of (8*S*)-8-hydroxy-erythromycin-A-6,9;9,12-acetal (**9**), $[\alpha]_D^{24} = -39°$ ($c = 1.03$), $\tilde{\nu}_{max}$ 3540, 1728 cm^{-1}.

$C_{37}H_{65}NO_{13}$ (731.898) Calc. C 60.72 H 8.95 N 1.91% Found C 60.51 H 9.28 N 1.87%

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[4]) It was later found that a column of Sephadex LH-20 prepared in methanol effected a much more efficient separation.

[5]) The time lag in the extractability with chloroform of (8*S*)-8-hydroxy-erythromycin-A (**7**) from the aqueous sodium hydrogen carbonate solution suggests a transient, water-soluble intermediate such as **14** which is converted to the N-oxide of **7** in the aqueous medium. Similar ester intermediates have been isolated from peracid oxidations [10].

8,9-seco-8-Oxo-erythromycin-A-9-oic-acid-6,9-lactone (**10**). 8,9-*seco*-8-Oxo-erythromycin-A-9-oic-acid-6,9-lactone (**10**) was formed as a by-product (10–20%) in a number of oxidations of 8,9-anhydro-erythromycin-A-6,9-hemiacetal (**6b**) with excess *m*-chloroperbenzoic acid both in the presence and absence of aqueous sodium hydrogen carbonate. Yields were erratic, and the conditions favoring its formation were not defined. **10** was isolated by countercurrent distribution employing the biphasic system adapted from that employed for partition column chromatography [8]. The more hydrophilic fractions were chromatographed on Sephadex LH-20 in chloroform/hexane 1:1. **10** was recrystallized from chloroform/hexane, and melted 162–168° (dec.), $[\alpha]_D^{26} = -57°$ ($c = 0.82$), $\tilde{\nu}_{max}$ 3540, 1724, 1705 (shoulder) cm^{-1}.

$C_{37}H_{65}NO_{14}$ (747.898) Calc. C 58.42 H 8.68 N 1.70% Found C 59.42 H 8.76 N 1.87%

REFERENCES

[1] *P. Kurath, J. R. Martin, J. Tanadier, A. W. Goldstein, R. S. Egan & D. A. Dunnigan*, Helv. *56*, 1557 (1973).
[2] *P. Kurath*, U. S. Pat. 3,674,773 (July 4, 1972).
[3] *P. Kurath, P. H. Jones, R. S. Egan & T. J. Perun*, Experientia *27*, 362 (1971).
[4] *G. O. Aspinall, R. R. King & Z. Pawlak*, Canad. J. Chemistry *51*, 388 (1973); *G. O. Aspinall & R. R. King*, Canad. J. Chemistry *51*, 394 (1973).
[5] *R. L. Foltz, L. A. Mitscher & M. I. Levenberg*, 17th Annual Conference on Mass Spectrometry and Allied Topics, May 18–23, 1969, Dallas, Texas, Paper No. 119, p. 343; *L. A. Mitscher, R. L. Foltz & M. I. Levenberg*, Org. Mass Spectrom. *5*, 1229 (1971); *P. Kurath & R. S. Egan*, Helv. *54*, 523 (1971).
[6] *L. A. Mitscher, B. J. Slater, T. J. Perun, P. H. Jones & J. R. Martin*, Tetrahedron Letters *1969*, 4505.
[7] *P. H. Jones, T. J. Perun, E. K. Rowley & E. J. Baker*, J. med. Chemistry *15*, 631 (1972).
[8] *N. L. Oleinick & J. W. Corcoran*, J. biol. Chemistry *244*, 727 (1969).
[9] *E. H. Flynn, M. V. Sigal, Jr., P. F. Wiley & K. Gerzon*, J. Amer. chem. Soc. *76*, 3121 (1954).
[10] *D. N. Kirk & J. M. Wiles*, Chem. Commun. *1970*, 518.

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280. Die Isolierung weiterer Alkaloide aus *Pleiocarba talbotii* WERNHAM

150. Mitteilung über Alkaloide[1])

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(8. X. 73)

Summary. From the leaves of the *Apocynaceae Pleiocarpa talbotii* WERNHAM three new indole alkaloids were isolated and identified as 3,4,5,6-tetradehydrotalbotine (**2**), 5,6-dehydrotalbotine (**3**) and deformyl talbotinic acid methylester (**4**). From the stem bark of the same plant normacusine B (tombozine) was isolated.

Aus der in Westafrika heimischen *Apocynaceae Pleiocarpa talbotii* WERNHAM wurden bisher die folgenden Indolalkaloide isoliert: Aus den Blättern Talbotin (**1**) [2] und aus der Stammrinde Talcarpin [3], Talpinin [3] und 16-epi-Affinin [3].

Bei der Chromatographie der Talbotin-Mutterlaugen konnten neben Talbotin (**1**) in kleinen Mengen drei weitere, neue Alkaloide isoliert werden: 3,4,5,6-Tetradehydro-

[1]) 149. Mitt., vgl. [1].