ISOLATION AND STRUCTURAL ELUCIDATION OF NAPHTHOMYCINS B AND C*

WALTER KELLER-SCHIERLEIN and MICHAEL MEYER

Organisch-chemisches Laboratorium der Eidgenössischen Technischen Hochschule, CH-8092 Zürich, Switzerland

AXEL ZEECK, MANFRED DAMBERG and REINHARD MACHINEK

Institut für organische Chemie der Universität Göttingen Tammannstr. 2, D-3400 Göttingen, W. Germany

HANS ZÄHNER and GÜNTHER LAZAR

Institut für Biologie II, Lehrstuhl Mikrobiologie I, Universität Tübingen, Auf der Morgenstelle 28, D-7400 Tübingen, W. Germany

(Received for publication January 8, 1983)

Two new ansamycin antibiotics, the naphthomycins B and C, were isolated from two different strains of Streptomyces. The structures were determined by comparison of the spectra (UV, ^{1}H NMR, ^{13}C NMR) with those of the known naphthomycin A, by spin decoupling experiments (300 MHz) and in one case by a two dimensional NMR analysis. Naphthomycin B (**II**) is 30-chloronaphthomycin C. Strikingly, naphthomycin A (**I**) differs from B and C not only by the presence of an additional methyl group at C (2), but also in the configuration of some of the double bonds. A fourth ansamycin antibiotic of the naphthomycin subgroup, actamycin, is 30-hydroxynaphthomycin C.

The antibacterial and antifungal ansamycin antibiotic, naphthomycin (thereafter called naphthomycin A), was first isolated from a strain of *Streptomyces diastatochromogenes* (synonym: *S. collinus*), strain Tü 105[2]. Its structure (**I**) was determined by WILLIAMS[3] by a careful ^{1}H NMR investigation and slightly revised upon a chemical degradation study by BRUFANI *et al.*[4]. A different strain of Streptomyces, strain Tü 353, belonging to the species *Streptomyces galbus* subsp. *griseosporeus,* isolated from a soil collected in Kouroussa (Guinea), produced an antibiotic closely related to naphthomycin A, which was called naphthomycin B. A third compound of the same family, naphthomycin C, was obtained during the isolation of the ansatrienins[5,6] from *Streptomyces diastatochromogenes* var. *diastatochromogenes,* strain Tü 1892. In this paper, the characterization and structural elucidation of the new naphthomycins are described.

Naphthomycin B

During the chromatographic purification of an ethyl acetate extract of the culture filtrate from strain Tü 353 this antibiotic moved as a single deeply yellow band. By recrystallization from acetone - hexane it was obtained as fine yellow needles of wax-like consistency. The two major peaks in a FAB mass spectrum (728, M+Na; 706, M+H) were consistent with the molecular formula, $C_{39}H_{44}ClNO_9$, which, however, agreed only roughly with the elemental analyses. Obviously, the crystals strongly retained variable and non stoichiometric amounts of solvent, predominantly hexane from the crystallization, whose presence is also observable in the NMR spectrum. The above molecular formula shows 1C atom and 2H atoms less than that of naphthomycin A[3].

* 220th communication of the series "Metabolites of Microorganisms". For preceding paper see reference 1.

The UV spectrum of naphthomycin B is nearly superimposable with that of naphthomycin A[2]; and the IR spectrum is very similar in the CO region with maxima at 1665, 1630 (sh.), 1605 and 1575 cm^{-1} (Fig. 4). The ^1H NMR spectra show one major difference: in the 2 ppm region of naphthomycin B we find only 3 methyl signals instead of 4 in naphthomycin A[2]. This suggested that naphthomycin B differs from A by replacement of a methyl group at a double bond by a hydrogen atom. This is confirmed by comparison of the ^{13}C NMR spectra (Table 1): naphthomycin A shows 7 signals for methyl carbon atoms, one of them (20.5 ppm) lacking in the spectrum of B. On the other hand, we find in the sp^2 region of naphthomycin B one doublet more (142.1 ppm) and one singlet less (132.7) than in the spectrum of naphthomycin A, whereas all other signals are the same in both spectra with only minor differences of chemical shifts.

The position of the lacking methyl group could be determined by NMR spin decoupling experiments at 300 MHz (Fig. 1 and Table 2). From these it follows that the six carbon atoms of the triene system carry a proton each, giving signals (from HC (2) to HC (7)) at 5.95, 6.60, 6.81, 6.74, 6.20 and 5.39 ppm.

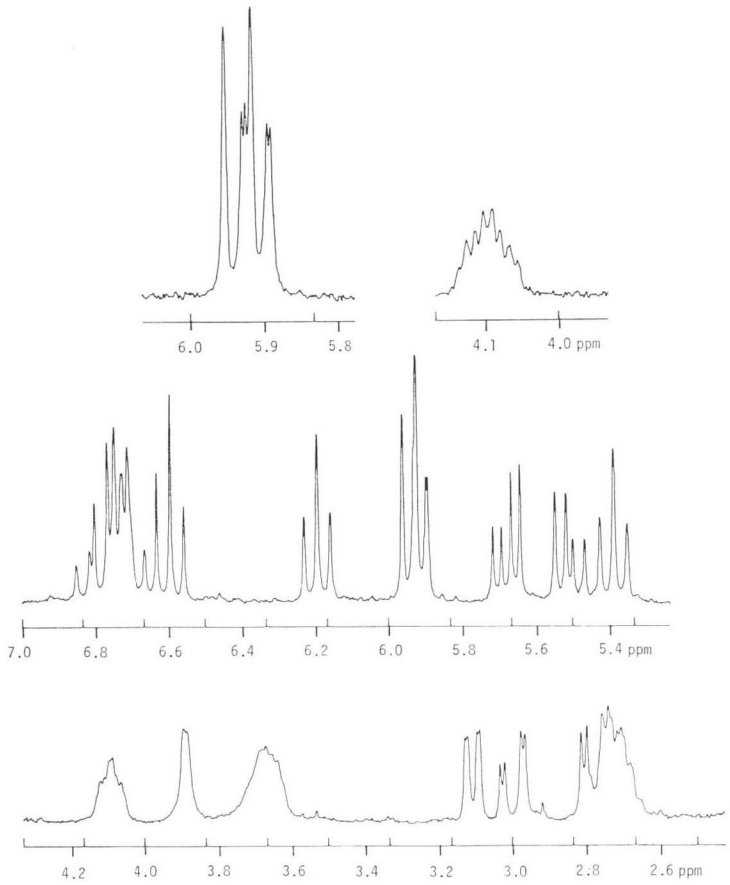
The mode of splitting in the overlapping region (3H) from 6.6 to 6.9 ppm becomes more transparent in the decoupling experiments. Therefore, it is the methyl group at C (2) of naphthomycin A which is absent in naphthomycin B.

Table 1. ^{13}C NMR spectral data of naphthomycins A, B, and C in $CDCl_3$.

	Naphthomycin A (25 MHz)	Naphthomycin B (25 MHz)	Naphthomycin C (50 MHz)		Naphthomycin A (25 MHz)	Naphthomycin B (25 MHz)	Naphthomycin C (50 MHz)
C=O region:					132.1 d	133.5 d	134.1 d
Ketone C atoms	203.8 s*	203.6 s*	203.5 s**		131.4 d	131.4 d	131.6 d
	201.9 s	201.7 s	202.4 s		128.8 d	128.9 d***	129.4 d***
Quinone C atoms	179.0 s	178.8 s	186.2 s		126.9 d	128.9 d***	129.4 d***
	178.0 s	178.1 s	179.4 s		123.1 d	119.9 d	118.4 d
Amide C atom	168.9 s	164.4 s	167.0 s		—	—	118.3 d
sp^2 region:				sp^3 region:			
Singlets	161.5 s	161.0 s	161.2 s	CH–O	77.0 d	76.6 d	76.2 d
	137.8 s	138.1 s	138.3 s		72.8 d	73.5 d	72.9 d
	137.6 s	137.7 s	138.2 s		72.2 d	72.4 d	71.4 d
	136.4 s	136.8 s	137.9 s	CH	45.0 d	41.6 d	41.4 d
	134.4 s	134.3 s	135.4 s		41.6 d	39.6 d	39.4 d
	133.6 s	133.3 s	132.1 s		33.7 d	33.6 d	33.6 d
	132.7 s	—	—	CH_2	40.6 t	40.8 t	40.6 t
	131.6 s	129.2 s	—		36.1 t	36.4 t	36.4 t
	121.7 s	121.5 s	122.1 s	CH_3	20.5 q	—	—
	120.0 s	120.3 s	119.4 s		17.2 q	17.7 q	17.4 q
Doublets	147.2 d	147.4 d	146.0 d		16.5 q	16.4 q	16.3 q
	143.0 d	142.9 d	145.9 d		16.1 q	16.2 q	15.7 q
	—	142.1 d	142.6 d		12.6 q	12.4 q	12.8 q
	140.0 d	139.4 d	140.4 d		11.1 q	11.2 q	11.1 q
	136.6 d	137.2 d	138.0 d		10.5 q	10.7 q	10.5 q
	134.1 d	135.8 d	137.1 d				

* Multiplicity under off resonance conditions.
** Multiplicity by the multiplets selection method.[11]
*** Overlapping of 2 signals.

Fig. 1. ^1H NMR spectrum of naphthomycin B in $CDCl_3$ (300 MHz).
The sections on top were measured under slightly modified conditions.



The signals of olefinic protons not belonging to the triene system are assigned as follows: The doublets of doublets at 5.51 and 5.68 ppm belong to a *trans*-disubstituted double linkage ($J_{16,17}=15$ Hz), flanked by a methine (2.21) and a carbinol proton (4.10). A doublet of doublets at 5.91 ppm shows a long-range coupling with a methyl group (2.04 ppm) and a vicinal coupling with a methine proton (2.72 ppm) and is therefore assigned to HC (21). From this it follows that a methyl group is present at C (22). The signal of HC (13) is partly hidden in the multiplet near 6.75 ppm integrating for 3H. WILLIAMS[3] had assigned the double bond C (6)=C (7) an *E*- and the double bond C (4)=C (5) a *Z*-configuration in naphthomycin A (formula I). To our surprise, the coupling constants in naphthomycin B ($J_{6,7}=11$, $J_{4,5}=15$, $J_{2,3}=11$) prove that C (4)=C (5) has an *E*-geometry, whereas the other two double linkages of the triene group are *Z*-configurated as demonstrated in formula II. Therefore, we have extended our 300 MHz irradiation experiments to the olefinic part of naphthomycin A (Fig. 2 and Table 3). Upon irradiation at 6.12 ppm (m, 2H; HC (4) and HC (5)), a doublet at 6.65 ppm (HC (3)) collapsed to a singlet, and at the same time the doublet of doublets at 6.51 (HC (6)) became a doublet with $J_{6,7}=15$ Hz. Irradiation at 6.51 ppm changed the two-proton multiplet at 6.12 and at the same time a signal at 5.47 ppm (dd→d, $J=10$), which is assigned to HC (7) in agreement with WILLIAMS' assignments based on chemically induced shift experiments.

Table 2. ^{1}H NMR spin decoupling experiments in naphthomycin B (300 MHz, $CDCl_3$).

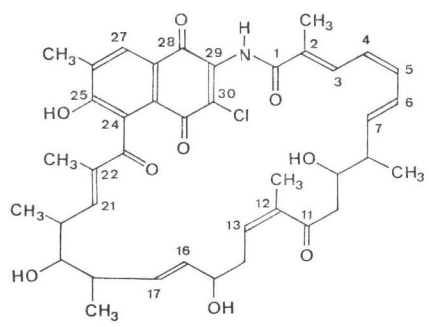
δ (ppm) H (x)[a]	Irradiation[b] (ppm)	Changed to/ J (Hz)	Assignment H (x)	δ (ppm) H (x)[a]	Irradiation[b] (ppm)	Changed to/ J (Hz)	Assignment H (x)
2.04 d (3H)	5.91	s	CH_3C (22)	5.39 dd	2.73	d/11	HC (7)
2.21 m	3.11	ch	HC (18)		6.20	d/11	
	5.51	ch		5.68 dd	4.10	d/15	HC (16)
2.30 m (2H)	4.10	ch	H_2C (14)	5.91 dd	2.72	d/2	HC (21)
2.72 m	5.91	ch	HC (20)	5.95	6.60	s	HC (2)
	3.11	ch		6.20 dd	5.39	d/11	HC (6)
2.73 m	3.66	ch	HC (8)		6.74	d/11	
2.77 dd	3.66	d/17	H_aC (10)	6.60 dd	6.81	d/11	HC (3)
3.00 dd	3.66	d/17	H_bC (10)		5.95	d/11	
3.11 dd	2.72	d/9	HC (19)	6.74 dd	6.81	d/11	HC (5)
3.66 m	2.73	ch	HC (9)		6.20	d/15	
4.10 ddd	5.68	dd/9+2	HC (15)	6.81 dd	6.60	d/15	HC (4)

[a] Coupling constants in Hz: 2/3=11, 3/4=11, 4/5=15, 5/6=11, 6/7=11, 7/8=11, 9/10a=5, 9/10b=4, 10a/10b=17, 14a/15=9, 14b/15=2, 15/16=7, 16/17=15, 17/18=9.5, 18/19=9, 19/20=2.5, 20/21=8, 21/CH_3=2.

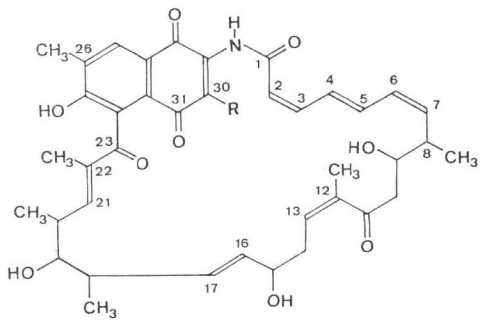
[b] No decoupling experiments: δ 0.82 (d, J=7, CH_3C (18)); 0.97 (d, J=6.5, CH_3C (20)), 1.19 (d, J=7, CH_3C (8)), 1.70 (s, broad, CH_3C (12)); 2.39 (s, CH_3C (26)), 5.51 (dd, J_1=15, J_2=9.5; HC (17)); 2.29, 3.68 and 3.89 (OH, exchangeable); *ca.* 6.75 (overlapped; HC (13)); 7.98 (s, HC (27)); 8.16 (s, exchangeable, NH); 9.64 (s, exchangeable, HO-C (25)).

Table 3. NMR spin decoupling in naphthomycin A (300 MHz, $CDCl_3$).

δ (ppm) H (x)	J (Hz)	Irradiation (ppm)	Changed to	Assignment H (x)
5.47 dd	$J_{7,8}$=10 $J_{7,6}$=15.5	6.51	d, J=10	HC (7)
6.12 m	(2 H)	6.51	ch	HC (5) and HC (4)
6.51 dd	$J_{6,7}$=15.5 $J_{6,5}$=10	6.12	d, J=15.5	HC (6)
6.65 d	$J_{3,4}$=9	6.12	s	HC (3)



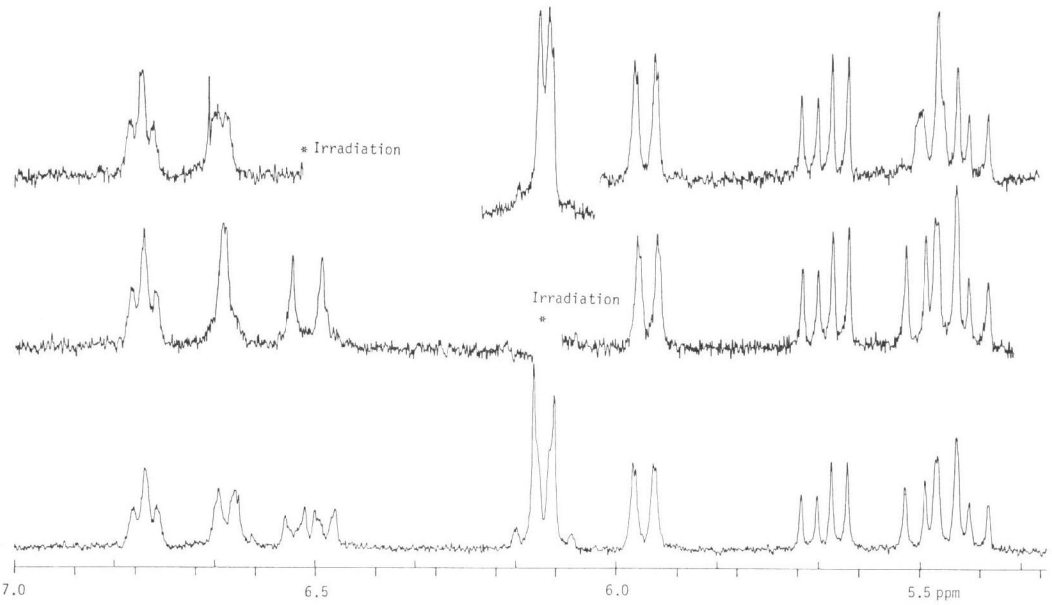
I



II R=Cl, III R=H

Thus, the *E*-configuration proposed by WILLIAMS for C (6)=C (7) in naphthomycin A is correct, and naphthomycin B is not simply 2-demethylnaphtomycin A, but also differs from it in the configuration of at least one double bond. On the other hand, the multiplet at 6.12 ppm for HC (4) and HC (5)

Fig. 2. ^1H NMR spin decoupling of naphthomycin A (olefinic section, 300 MHz, $CDCl_3$).



shows higher order spin-spin interactions. WILLIAMS' assignment of a *Z*-configuration to this double bond can therefore not be confirmed by our experiments, although it is probable from the induced shift experiments[8].

The signals of HC (16) and HC (17) at 5.66 and 5.43 ppm show a coupling constant of 15 Hz. The isolated double bond is, therefore, *trans*-substituted as in naphthomycin B. In the latter, there remain to be determined the configurations of the trisubstituted double linkages, C (12)=C (13) and C (21)=C (22). We tried to solve this problem by the application of nuclear Overhauser experiments. Irradiation at 2.04 ppm (CH_3C (22)) gives a strong NOE at 2.7 ppm (HC (20)) and weak enhancements at 3.1 (HC (19)) and 5.9 ppm (HC (21)), thus proving the *E*-configuration of the C (21) double bond. On the other hand, irradiation at 1.7 ppm (CH_3C (12)) gives only weak and doubtful effects at 2.3 and 6.7 ppm, which does not allow an assignment of the configuration. In formula **II** for naphthomycin B the double bond C (12)=C (13) is drawn arbitrarily. Whith naphthomycin A no NOE experiments were carried out.

In addition to naphthomycin B strain Tü 353 produced ansatrienins, identical with those from the original ansatrienin producer, strain Tü 1892, which were predominantly present in the mycelial cake.

Naphthomycin C

Naphthomycin C was obtained as a green-yellow amorphous powder which decomposed above 218°C. It is nearly inactive against bacteria and fungi. Its molecular formula, $C_{39}H_{45}NO_9$, was determined by high resolution mass spectrometry and corresponds formally to a dechloronaphthomycin B. In the ^{13}C NMR spectrum the only remarkable difference from naphthomycin B is an additional doublet at 118.3 ppm instead of a singlet at 129.2 ppm, in agreement with the above conclusion. A shift of a quinone carbonyl signal from 178.8 ppm in B to 186.2 ppm in C confirms a structural change in the quinone ring. All other signals correspond closely with those of naphthomycin B (Table 1). The ^1H NMR spectrum in $CDCl_3$ (Table 4) shows nearly the same signals as that of naphthomycin B.

Table 4. ^{1}H NMR spectral data of naphthomycin C (200 MHz).

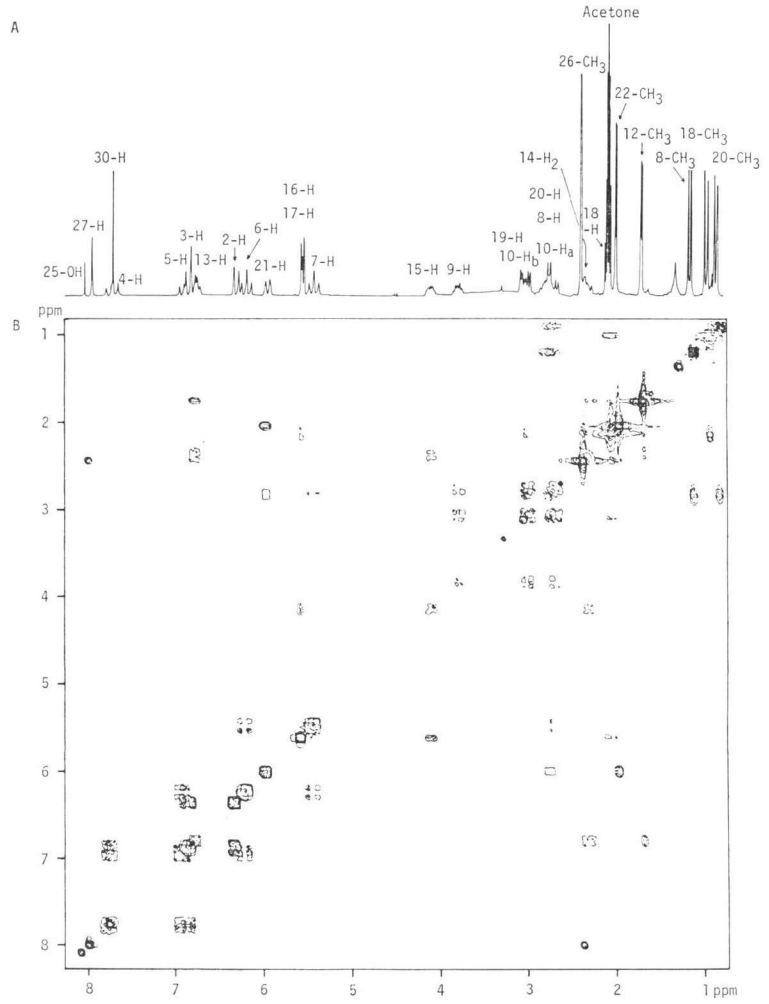
Acetone-d_6		$CDCl_3$	Assignment
δ (ppm)	J (Hz)	δ (ppm)	
0.82 d	6.5 (3H)	0.85	CH_3C (20)
0.93 d	6.5 (3H)	0.99	CH_3C (18)
1.12 d	6.5 (3H)	1.21	CH_3C (8)
1.68 d	1 (3H)	1.69	CH_3C (12)
1.96 d	1.2 (3H)	2.05	CH_3C (22)
2.06	overlapped	2.1~2.34	HC (18)
2.32 m	(2H)	2.1~2.34	H_2C (14)
2.36 d	0.8 (3H)	2.34	CH_3C (26)
2.69 dd	17/5.5	2.81	H_aC (10)
2.77 m	(2H)	2.72	HC (8), HC (20)
3.00 dd	17/4.5	3.03	H_bC (10)
3.03 m		3.11	HC (19)
3.78 m		3.61	HC (9)
4.08 m		4.15	HC (15)
5.42 dd	11/11	5.48	HC (7)
5.56 m	(2H)	5.55	HC (17)
		5.67	HC (16)
5.96 dd	11/1.2	5.94	HC (21)
6.20 dd	11/11	6.30	HC (6)
6.32 d	11	5.86	HC (2)
6.77 m		6.84	HC (13)
6.84 dd	11/11	6.72	HC (3)
6.93 dd	15/11	6.84	HC (5)
7.73 s		7.68	HC (30)
7.75 dd	15/11	7.34	HC (4)
7.97 d	0.8	7.99	HC (27)
8.06 s	exchangeable	9.73	HOC (25)
9.17 s	exchangeable	8.44	NH
—		2.81 exchangeable	
—		3.31 exchangeable	3 HO
		3.96 exchangeable	

A characteristic difference is an additional singlet at 7.68 ppm for the proton at C (30) which replaces the chlorine atom of naphthomycin B. The correlation of signals could be carried out by means of a two dimensional spectrum[7] taken in acetone-d_6. The correlations revealed unambiguously the two partial formulae from C (2) to C (10) and from CH_3C (12) to CH_3C (22). The structure of the quinone moiety follows from the close similarity of the spectra (UV, ^{1}H NMR, ^{13}C NMR) with those of naphthomycins A and B and, when available, with those of actamycin[8,9], an antibiotic closely related to the naphthomycins. Actamycin is constitutionally 30-hydroxynaphthomycin C; however, a stereochemical correlation is not possible at present.

A comparison of the ^{1}H NMR data of naphthomycins B and C (Tables 2 and 4) shows that the coupling constants of corresponding olefinic protons are the same in both antibiotics. Thus, naphthomycin C has the same configuration at all disubstituted double bonds as naphthomycin B, *i.e.* (*Z*)-(*E*)-(*Z*) in the triene system, and (*E*) at the isolated double bond C (16)=C (17).

Nearly identical chemical shifts of the methyl protons at C (12) and C (22), and of the olefinic protons at C (13) and C (21) in both antibiotics also indicate that the configurations at the trisubstituted double linkages are the same. The two antibiotics possess therefore the structures given in formulae

Fig. 3. ^1H NMR spectrum of naphthomycin C in acetone-d_6 (200 MHz).
A: Normal FT-spectrum; B: 2D FT-spectrum (contour plot), $^1H/^1H$ shift correlation, 512×512 data points.



II and III with uncertain configuration at C (12)=C (13).

A striking difference is found for the chemical shifts of HC (4), 6.60 ppm in naphthomycin B and 7.34 in naphthomycin C (both in $CDCl_3$). A plausible explanation for this is that some carbon atoms of the triene chain are twisted out of the plane by the large chlorine atom in naphthomycin B, whereas this system is planar in naphthomycin C.

Experimental

General

IR spectra in pressed KBr disks were recorded using a Perkin Elmer model 297 spectrometer, UV spectra using a Perkin Elmer model 402 or Zeiss DMR 21 spectrometer. FAB-mass spectrum was obtained on a M-Scan Ltd. instrument, EI-mass spectrum on a Varian MAT 731 (70 eV) using the direct probe insert, high resolution with perfluorokerosine as a standard. ^1H NMR spectra were determined at 200 MHz with a Varian XL-200, at 300 MHz with a Bruker WM-300. The program

HOMOCOR (Varian) was used for the two dimensional spectrum; the NOE experiments were done by Fourier difference analysis. Chemical shifts (δ in ppm) are reported relative to internal tetramethylsilane. ^{13}C NMR spectra were determined on a Varian XL-100 (25.2 MHz) or a Varian XL-200 (50.4 MHz). Optical rotations were taken with a Perkin Elmer model 241 polarimeter.

Fermentation and Isolation of Naphthomycin B (II)

Strain Tü 353 was cultured at 27°C in a 200-liter, Giovanola b, fermenter in a nutrient medium containing 2% soybean meal and 2% glucose, pH 7.5 before sterilization. Agitation: 800 rpm; aeration: 50 liters/minute.

After 66 hours of cultivation, the culture was harvested, 2% Celite added, and the mixture filtered. From the filter cake ansatrienins A and B, identical with those from strain Tü 1892[5,6], could be obtained by extraction with acetone. The culture filtrate was extracted twice with ethyl acetate, the extracts concentrated under reduced pressure to a small volume and the crude naphthomycin B precipitated by the addition of petroleum ether.

This precipitate (*ca.* 0.5 g) was chromatographed on a column of 25 g silica gel with $CHCl_3$ - MeOH, 19: 1 as the eluant. The first yellow fractions gave 120 mg of chromatographically pure substance, followed by 86 mg of slightly contaminated material. Recrystallization from acetone - hexane gave orange yellow needles with mp 156~165°C (decomp.). $[\alpha]_D^{25}+412°$ (*c* 0.5, $CHCl_3$). On TLC plates (silica gel, Merck F_{254}; ethyl acetate - H_2O - formic acid, 100: 30: 2.5, upper phase) the Rf value (0.52) was lower than that of naphthomycin A (0.62). UV (EtOH): λ_{max} (log ε) 235 nm (4.63), 307 (4.56), 360 (sh. 4.05); UV (0.01 N NaOH in EtOH): 235 (4.59), 297 (4.54), 345 (sh. 4.14), 434 (4.17), *ca.* 570 (broad, 3.08). IR (KBr) see Fig. 4. ^1H NMR (300 MHz, $CDCl_3$), see Fig. 1 (high field part not depicted) and Table 2; the CH_3 region is somewhat disturbed by the presence of hexane. ^{13}C NMR ($CDCl_3$), see Table 1. The antimicrobial activity of naphthomycin B is very similar to that of naphthomycin A[2]: high inhibition of Gram-positive bacteria and many fungi.

Anal. Calcd. for $C_{39}H_{44}ClNO_9$ (706.23): C 66.32, H 6.28, N 1.98, Cl 5.02.
Calcd. for $C_{39}H_{44}ClNO_9 \cdot C_6H_{14}$: C 68.21, H 7.38, N 1.77, Cl 4.47.
Found: C 66.43, H 6.63, N 1.82, Cl 4.89.
C 67.28, H 7.03, N 1.72, Cl 4.30.

Two extreme values out of 8 analyses are given.

Titration in methyl cellosolve - H_2O (8: 2): *pKa'* 7.10, equivalent weight found 739. FAB-MS: *m/z* 728 (M+Na), 706 (M+H), 688 (706−H_2O), 670 (706−$2H_2O$), 652 (706−$3H_2O$).

Isolation of Naphthomycin C (III)

A crude ansatrienin mixture from strain Tü 1892 (1.8 g) was chromatographed on a column (30×5.0 cm) of silica gel with $CHCl_3$ - MeOH, 96: 4 as eluant (flash chromatography). A first deeply yellow fraction (220 mg) contained ansatrienins A, A_2 and A_3[5,6,10], a second faintly yellow fraction (480 mg) the ansatrienin B components together with naphthomycin C. The latter fraction was further separated by preparative (thick) layer chromatography (silica gel P UV_{254}, Macherey & Nagel, plates 20×40 cm) with the above eluant. A fast moving yellowish green zone gave by extraction and precipitation with

Fig. 4. IR spectrum of naphthomycin B in KBr.

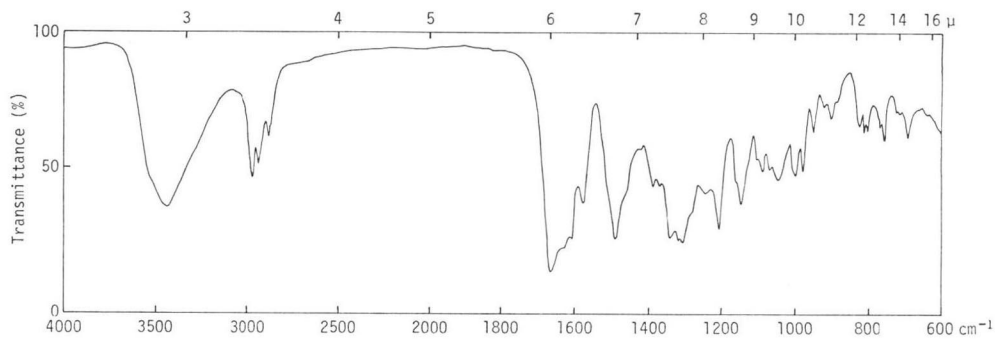
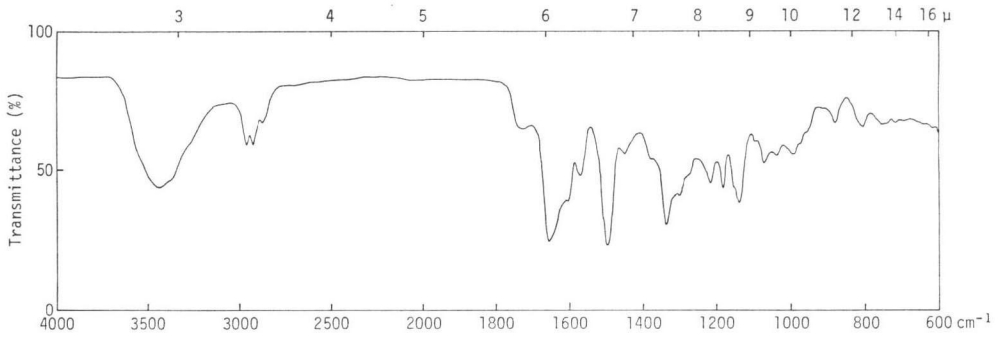


Fig. 5. IR spectrum of naphthomycin C in KBr.



pentane 50 mg naphthomycin C as an amorphous greenish yellow powder. $[\alpha]_D^{20}+117.7°$ (c 0.8, $CHCl_3$). Rf 0.31 (TLC plates, Polygramm SIL G/UV 254, $CHCl_3$ - MeOH, 96: 4, ansatrienin A: Rf 0.47). EI-MS: m/z (abund.) 671 (0.5%, M^+; high resolution found: 671.3085, calcd. for $C_{39}H_{45}NO_9$: 671.3079), 653 (3%, M−18, $C_{39}H_{43}NO_8$), 635 (2), 299 (14), 298 (15), 258 (18, $C_{14}H_{12}NO_4$), 230 (28, $C_{12}H_8NO_4$), 91 (100). UV (MeOH): λ_{max} (log ε) 232 nm (4.51), 286 (sh), 307 (4.45), 350 (sh, 4.26). IR (KBr), see Fig. 5. ^1H NMR, see Fig. 3 and Table 3. ^{13}C NMR in $CDCl_3$, see Table 1. Naphthomycin C was nearly inactive against bacteria and fungi.

Acknowledgment

We gratefully acknowledge the cooperation of the following colleagues and technicians: Mr. D. MANSER, Zürich (microanalyses); Miss B. BRANDENBERG, Zürich (NMR spectra); Dr. B. JAUN, Zürich (NOE experiments); Dr. G. REMBERG, Göttingen, Dr. J. MEILI and Prof. Dr. J. SEIBL, Zürich (mass spectra).

The authors from Göttigen and Tübingen thank the Deutsche Forschungsgemeinschaft for financial support. The investigations in Zürich were graciously supported from a special research fund of ETH-Zürich.

References

1) KELLER-SCHIERLEIN, W.; B. JOOS, H. P. KAISER & P. GASSMANN: Strukturaufklärung von Niphimycin. 2. Die Konstitution von Desmalonyl-niphimycin Iα und Iβ. Helv. Chim. Acta 66: 226~258, 1983
2) BALERNA, M.; W. KELLER-SCHIERLEIN, C. MARTIUS, H. WOLF & H. ZÄHNER: Stoffwechselprodukte von Mikroorganismen. 72. Naphthomycin, ein Antimetabolit von Vitamin K. Arch. Mikrobiol. 65: 303~317, 1969
3) WILLIAMS, T. H.: Naphthomycin, a novel ansa macrocyclic metabolite. Proton NMR spectra and structural elucidation using lanthanid shift reagent. J. Antibiotics 28: 85~86, 1975
4) BRUFANI, M.; L. CELLAI & W. KELLER-SCHIERLEIN: Degradation studies of naphthomycin. J. Antibiotics 32: 167~168, 1979
5) WEBER, W.; H. ZÄHNER, M. DAMBERG, P. RUSS & A. ZEECK: Stoffwechselprodukte von Mikroorganismen. 201. Ansatrienin A und B, fungistatische Antibiotica aus *Streptomyces collinus*. Zbl. Bakt. Hyg. I. Abt. Orig. C 2: 122~139, 1981
6) DAMBERG, M.; P. RUSS & A. ZEECK: Die Konstitution der fungistatischen Ansamycin-Antibiotica Ansatrienin A und B. Tetrahedron Lett. 23: 59~62, 1982
7) FREEMAN, R. & G. A. MORRIS: Two dimensional fourier transformation in NMR. Bull. Magn. Reson. 1: 5~26, 1979
8) ALLEN, M. S.; I. A. MCDONALD & R. W. RICKARDS: The ansamycin antibiotic actamycin. I. Definition of structural features by deuterium labelling. Tetrahedron Lett. 22: 1145~1148, 1981
9) MCDONALD, I. A. & R. W. RICKARDS: The ansamycin antibiotic actamycin. II. Determination of the structure using carbon-13 biosynthetic labelling. Tetrahedron Lett. 22: 1149~1152, 1981
10) LAZAR, G.; H. ZÄHNER, M. DAMBERG & A. ZEECK: Ansatrienin A_2 and A_3: Minor components of the ansamycin complex produced by *Streptomyces collinus*. J. Antibiotics 36: 187~189, 1983
11) LE COCQ, CH. & J. Y. LALLEMAND: Precise carbon-13 NMR. Multiplicity determination. J. Chem. Soc., Chem. Comm. 1981: 150~152, 1981